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EURYGASTER INTEGRICEPSDAN XITOZAN OLISH

Annotatsiya: Butun dunyoda tabiiy xomashyo manbaalaridan xitin moddasini ajratib olish va u asosida xitozan sintez qilishga hamda olingan moddalarni zamonaviy asbob-uskunalar yordamida fizik-kimyoviy konstantalarini aniqlashga qaratilgan tadqiqotlar jadal jivojlanib bormoqda. Ushbu maqolada biz uchun mahalliy xomashyo boʻlgan Eurygaster integricepsdan xitin, xitozan moddalarini olishning optimal sharoitlarini aniqlash, olingan moddalarni IQ va UB spektrlarini taxlil qilish natijalari toʻgʻrisida soʻz yuritiladi.

Kalit soʻzlar: Eurygaster integriceps, xitin, xitosan, IQ spektr, UB spektr, xomashyo, ekstraksiya.

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ПОЛУЧЕНИЕ ХИТОЗАНА ИЗ EURYGASTER INTEGRICEPS

Аннотация: Во всем мире активно развивается исследования, направленные на извлечение хитина из природного сырья и синтез хитозана на его основе и определение физико-химических констант полученных веществ с помощью современных оборудования. В статье описаны оптимальные условия

извлечения хитина и хитозана из местного для нас сырья *Eurygaster integriceps*, а также результаты ИК- и УФ-спектрального анализа полученных веществ.

Ключевые слова: *Eurygaster integriceps*, хитин, хитозан, ИК-спектр, УФ-спектр, насекомое, сырьё, экстракция.

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EXTRACTION OF CHITOSAN FROM EURYGASTER INTEGRICEPS

Abstract: All over the world, research aimed at extracting chitin from natural raw materials and synthesizing chitosan based on it and determining the physico-chemical properties of the obtained compounds with the help of modern equipment is actively developing. This article describes the optimal conditions for extracting chitin and chitosan from *Eurygaster integriceps*, a local raw material for us, and the results of IR and UV spectra analysis of the obtained compounds.

Keywords: *Eurygaster integriceps*, chitin, chitosan, Infrared spectrum, insect, raw material, extraction.

Introduction

Chitin is the building block of insects. It is the second most widely distributed biopolymer in nature (after cellulose). It represents β -1,4-homopolymer of N-acetylglucosamine.

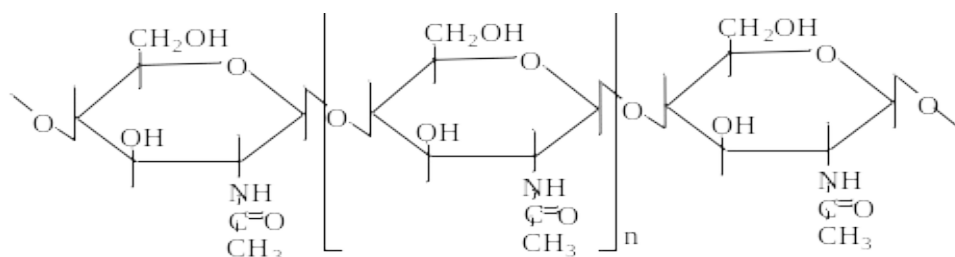


Figure 1. The structural formula of chitin

The substances chitin and chitosan, like most other compounds containing the elements of carbon and nitrogen, participate in the global cycles of these elements in

nature. In nature, chitin has three main sources: the shell of crustaceans, the cuticle of insects, and the cell wall of filamentous fungi. It is the building material of most animals and mainly performs an auxiliary function. In crustacean shells it is associated with proteins and calcium, and in the cuticle of insects it is present in the form of a chitin-melanin complex. In the cell wall of fungi, chitin is found predominantly in complex with $\beta(1,3)$ -glucan and is the main structural polysaccharide of mycelial hyphae. The cell walls of zygomycete fungi contain chitin and chitosan, produced by deacetylases [1-2].

As a result of many studies, scientists have found that the structure of chitin is not the final structure, as it can be transformed into chitosan by further processing. Chitosan is a linear polysaccharide, an N-deacetylated derivative of chitin polysaccharide [3-4].

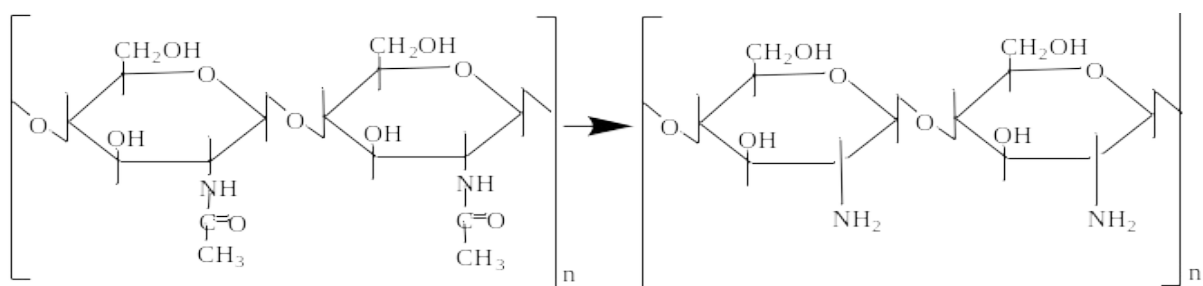


Figure 2. Deacetylation reaction of chitin with NaOH

As part of this work, the goal was to isolate chitosan from the *Eurygaster integriceps*, a pest of local plants [5-6]. Also during the study, optimal conditions for the extraction of chitosan were developed.

Chitin is usually found in natural sources together with protein, various minerals, lipids and other coloring substances. When extracting chitin using the classical method, it is first purified from water-soluble substances. Next raw materials are cleaned from lipids using non-polar solvents, mineral substances using dilute solutions of acids, protein substances using weak alkali solutions, coloring additives using hydrogen peroxide solution. Chitin is extracted from it. The resulting chitin is

deacetylated with a high concentration NaOH solution and chitosan is formed. Each stage is carried out at different times and temperatures depending on the type of raw materials used. In addition, the used alkali, acid and hydrogen peroxide solutions are selected depending on the amount of additives in the raw materials [7].

Research methodology

During our research, various physico-chemical research methods were used to determine the composition and structure of substances extracted from *Eurygaster integriceps* and synthesized on their basis, including:

IR spectrum was obtained using INVENIO S (BRUKER, 2021. 4000-400 cm^{-1}).

UV spectrum was obtained using UV/Vis Spectrophotometer (UV-755) (APL Instruments Shanghai, China) 190-1100 nm.

Analysis of the obtained results

There are many and varied sources of chitin. They can include the tissues of most fungi and some algae, cuticles of insects, shells of crustaceans, upper shells of worms, and some species of molluscs. Taking into account that the above-mentioned sources are not available in the conditions of Uzbekistan or their reproduction is low, it was aimed to extract chitosan from an alternative source (*Eurygaster integriceps*). A certain amount of *Euryaster integriceps*, which was first collected and dried, was boiled and washed for 2 hours using distilled water to remove various additives. Then, for deproteinization, it was boiled with a 3.2% NaOH solution, the next stage was the demineralization stage, in which the obtained residue was treated with 3.5% HCl. In the next step, the obtained residue was boiled in a water bath with 4% NaOH for complete deproteinization (after treatment with alkali and acid solutions, the sample was washed to a neutral environment and dried).

In the next step, acetyl groups were removed by heating the chitin with a 30% caustic sodium solution, and the chitosan (chitin-chitosan mixture) was isolated, this step is called deacetylation. Below is the IR spectrum of the chitosan obtained:

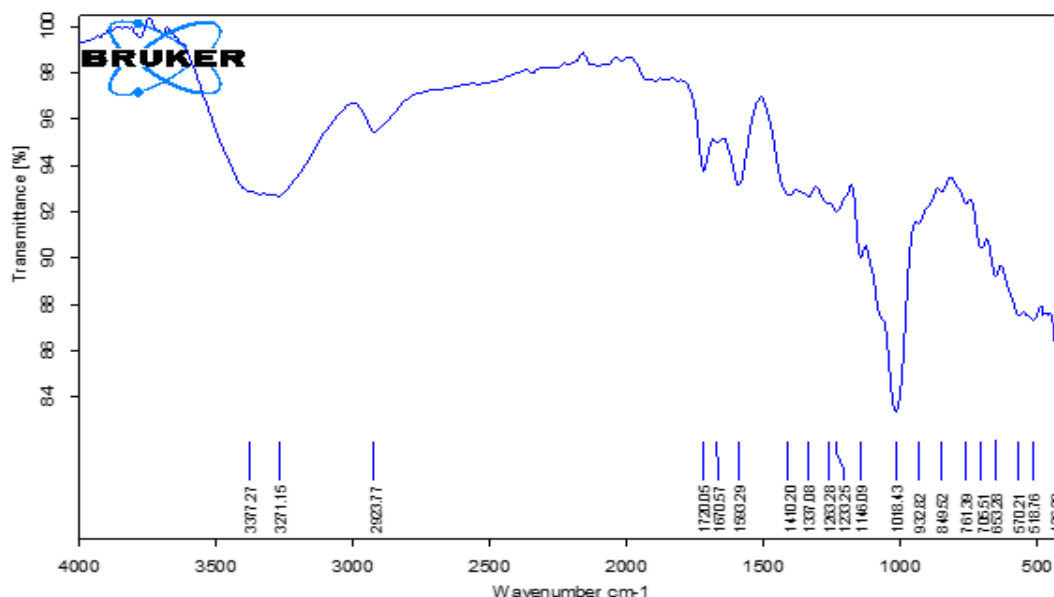


Figure 3. IR spectrum of the obtained chitosan

Chitosan molecule has functional groups such as -OH , -NH_2 , -NHCOCH_3 , -C-O-C . In the IR spectrum of chitosan isolated from *Eurygaster integriceps* (Fig. 3), these functional groups and their associated chemical bond vibrations can be seen. In particular: in the IR spectrum, the peak with average broad band intensity in the region of 3377 cm^{-1} is -OH , the region of 3271 cm^{-1} belongs to free amino groups, and the width of the band represents the presence of hydrogen bonds in the chitosan molecule. In the 2923 cm^{-1} area, the absorption peaks formed by the vibration of sp^3 hybridized carbon between C-H bonds in the structure were observed. In the 1720 cm^{-1} area, the absorption peaks caused by the valence vibrations of the -C(O)- bond, in the 1593 cm^{-1} area by the -C-N-H bond, and in the 1016 cm^{-1} area by the valence vibrations of the -C-O-C- bond we can observe.

These absorption peaks were found to be very close when compared with infrared spectra of chitosans isolated from other sources.

Optical properties of chitosan biopolymer isolated from *Eurygaster integriceps* were analyzed using a UV-visible spectrometer. Due to glucosamine (GlcN) and N-acetylglucosamine (GlcNAc) groups in the sample, the absorption maximum in a hardly wide range can be seen at 310 nm (Fig. 4).

The resulting UV spectrum (Fig. 4, right) was found to be very similar to the UV spectrum of chitosan obtained from *Apis mellifera* (Fig. 4, left) reported in the literature.

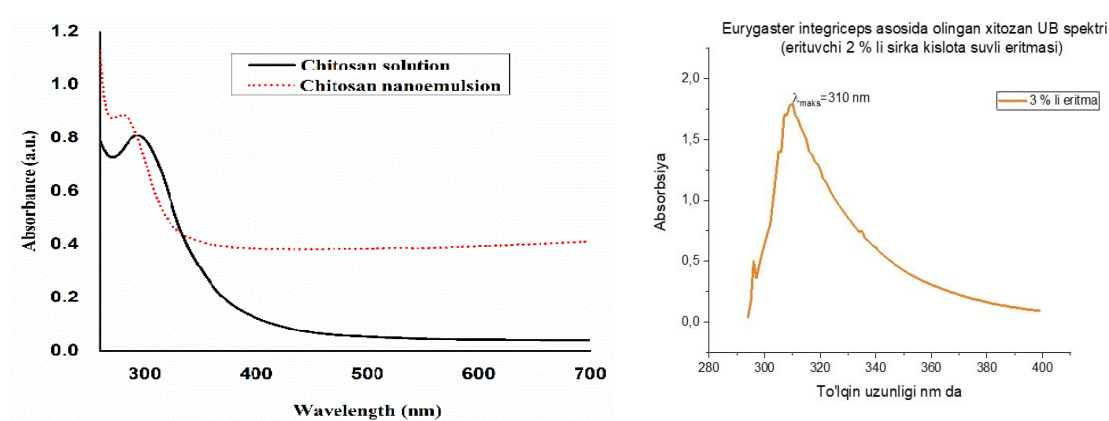


Figure 4. UV spectrum of chitosan from *Apis mellifera* and *Eurygaster integriceps*

Chitosan absorption depends on several factors. An increase in concentration causes an increase in the peak wavelength. Also, increasing the concentration of acetic acid used as a solvent decreases the height of the peak.

Experimental part

Extraction of chitin from raw materials. 10 g of dried samples of *Eurygaster integriceps* were taken and crushed in a porcelain mortar. This raw material was placed in a heat-resistant conical flask and deproteinized by adding 60 mL of 3.2% NaOH for 45 minutes. In the next step, the dried residue was demineralized by pouring 60 ml of 3.5% HCl solution over it for 40 minutes. After both steps, the mixture was washed with distilled water under vacuum in a Buchner funnel until it has become neutral. It was filtered and dried. To completely deproteinize the dried residue, it was heated in 60 ml of 4% NaOH solution for 80 minutes. The mixture

was then washed with distilled water under vacuum until neutral and filtered and dried. The obtained mass of chitin was 1.26 g with the 12.6% yield.

Hydrolyzing chitin into chitosan. 1.26 g of dry chitin was placed in a flask, 50 ml of 30% NaOH solution was added to it and boiled for 40 minutes. The mixture was then washed with distilled water until neutral in a Buchner funnel under vacuum. When dried by filtration, the mass was 0.864 g. The yield was 8.64 % in relation to the original raw material.

Summary

In order to obtain chitin, the widely available local (*Euryaster integriceps*) source was used. From the conducted experiments, using different concentrations of alkali solutions with 3.2%, 4% and 30% of NaOH and 3.5% solution of HCl was found to be acceptable. The overall yield of the process was found to be 8.64%.

The infrared and ultraviolet spectra of the obtained chitosan were found to be very similar when compared with the spectra of chitosan obtained from other sources.

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