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SYNTHESIS AND CHARACTERIZATION OF CHITOSAN USED IN THE TREATMENT OF EPITHELIAL LESIONS

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Abstract. *The skin is considered the largest organ of the body of bofh animals and humans. Due to the high incidence of skin lesions, the biomedical engineering is constantly developing materials to optimize the dynamics of epithelial healing. Among these materials, there is chitosan, a polymer which exhibits high biocompatibility, accelerates wound healing, protection against microbial attack and allows the passage of water vapor and air. It is extracted from the deacetylation of chitin biopolymer present in cell walls of fungi, exoskeletons of insects and crustaceans. Based on that, the chitosan was synthesized from the exoskeletion of the lobster – Palinurus elephas – through the processes of demineralization, deproteinization and deacetylation. Was performed XRD in all the processes of synthesis of chitosan confirming the characteristics of its structure and molecular chain that assist in epithelial healing.*

Keywords: *Chitosan, lesion, crystallinity structure.*

1. INTRODUCTION

The skin is considered the largest organ of the body of both animals and humans. It is a tissue rich in nerve endings responsible for receiving environmental information and send them to the central nervous system (Junqueira and Carneiro, 2005; Veiga, 2009 a). Performs many functions such as protecting against injury physical, chemical and biological, prevents water loss by evaporation, serves as a wide receiver for general sensations (pain, pressure, touch, temperature), protects against ultraviolet radiation converts into vitamin D precursor molecules , works on thermal regulation and excrete certain substances through sweat glands (Ross *et al*, 1993; Rigau, 1996; Martin and Paul, 1997). As this is the first barrier protecting the body, the skin is exposed to environmental assaults that can result in loss of continuity. Thus, lesions with or without tissue loss may cause a physiological imbalance to this body and the structures protected by him, leaving the body vulnerable to the entry of pathogens micro-organisms and opportunistic (Veiga, 2009 b).

The tissue lesions decrease the body's normal defense against infection and, in addition, the fluid exiting the lesion offers a favorable environment for growth of microorganisms. The lesion or ulcer is a sore on the skin or mucosa, and gradual disintegration of tissue necrosis (Ferreira, 1999). Once installed the wound, starts immediately tissue repair. The healing is a physiological and biochemical phenomenon that has as main objective to restore the anatomical integrity of the injured tissue.

It is in this sense that in recent decades there has been an increased need for medical device development, with high compatibility with the body and to enable integration between cells and synthetic membranes for a variety of biomedical applications (Chuang, 1999). The difficulty of selecting an ideal biomaterial to mount this framework is justified precisely on the challenge of adding the same structure a material that presenting a profile physico-chemical and biological properties ideal for assisting the skin physiological reconstruction, such as: biocompatibility, is not cytotoxic, biodegradable (Ritner *et al*, 2004) and proper biomechanics are the mechanical properties required for its biological utilization (Griffitih, 2002), porosity, toxicity, antibacterial and antifungal action, hydrophilicity, suitable pH,

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ease of adhesion proteins, among others, all desirable for a good performance within the implantation site. (Resieh *et al*, 2010)

In this world, a number of metallic, ceramic, polymeric (synthetic and biógenos) and composites have been tested. A natural polymer, chitosan More specifically, it has been shown to be an alternative that would have aroused great interest of scientists and technologists as functional polymeric materials.

Chitin and chitosan are polysaccharides nontoxic, biodegradable, biocompatible and produced by renewable natural sources, with applications in several areas: agriculture, food industry, textile industry, pharmaceutical industry, developing cosmetics and biomaterials, such as gels, films, polymeric membranes and nanofibers (Tonhi *et al*, 2002).

Chitin is a natural polymer, monomer unit linear b-(1-4)-N-acetyl-D-glucosamine like show the figure 1. Precursor of chitosan, is the second most abundant biopolymer found in nature after cellulose (Canella *et al*, 2001). Is present in cell walls of fungi, crustaceans (shrimp, crabs, crabs, lobsters) is insoluble in water, organic solvents, dilute acids and alkalis. It is obtained by demineralization, deproteinization and deodorizing waste the exoskeleton of crustaceans. After these steps, is performed the chitin deacetylation to obtain a chitosan.

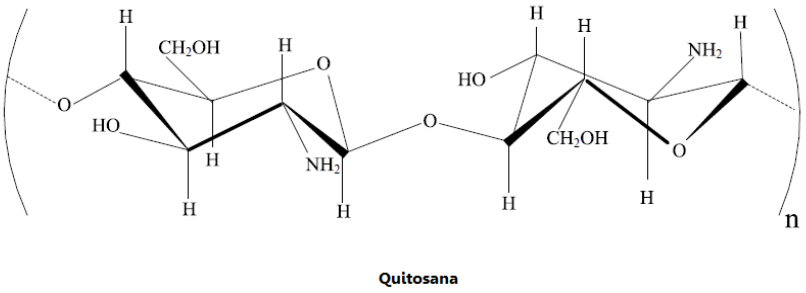

Figure 1. Structure of chitosan with their respective structural units (adapted from Sacchetin 2009)

Thus, an increasing number of researches on chitin and chitosan as biomaterials for the treatment of injuries, motivated from several studies that prove its properties in the acceleration of wound healing, protect against microbial attack and the passage of water vapor , air and has high antiseptic power.

For synthesis of chitosan was used the exoskeleton of the lobster and analyzes performed by X-ray diffraction for characterization and confirmation of the semi-crystalline structure.

2. MATERIALS AND METHODS

The amount of raw material required for the development of the research was obtained by producers of waste Lobster - Palinurus elephas, such as hotels, restaurants and markets in the city of Natal / RN. Then, was performed the processing of residues required for chitin production through the following steps:

Grinding - obtaining 0.5 mm grain size to increase the contact surface of the material and consequently facilitate the next steps. Before grinding exoskeletons were sun dried for 24 hours at a temperature of 29 ° C;

Demineralization - reduce the ash content, noting the elimination of carbonates and phosphates (HCL). Was added HCl 1M to 7.0% v/v with a bath ratio of 1:5 (10 g, dry milled material in 50 ml of aqueous solution). The mixture was placed in a water bath at 75 ° C for better interaction of the reaction for 1 hour, then demineralized powder was filtered and washed with water until neutrality and the filtrate placed in an oven at 80 ° C for 1h and then placed in the desiccator until constant temperature;

Despoteinização - to reduce the nitrogen content of protein (NaOH). We used an aqueous solution of NaOH to 7% w/v, with bath ratio of 1: 50, the mixture was heated in a water bath at 75 ° C to accelerate the reaction for 8 hours. This material was then filtered and washed with water until neutrality of the filtrate followed by further drying of. Figure 2 show the chitin obtained.

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Figure 2. Chitin obtained after the process.

The treatment method of chitin to chitosan production is by deacetylation (NaOH). Was used NaOH 40% w/v, with bath ratio of 1: 50 (in 1 g of chitin was added 50 ml of NaOH) the mixture was heated in a water bath at 100 ° C for 4 hours. The material was washed thoroughly with water until neutrality of the filtrate, removing the excess of the reagent which has been found by measuring the pH, followed by further drying.

At each step of synthesis of chitosan analyzes were performed by X-ray diffraction (XRD) to characterize the biomaterial.

3. RESULTS AND DISCUSSES

Measurements of X-ray diffraction were performed on diffractometer Universal X-ray model Shimadzu XRD-6000 with Cu radiation with a power of 112 kV and current of 31 mA from 5 to 80 ° (2Θ).

The diffractogram (Fig. 3) is the initial powder of the lobster exoskeleton. Observed a several prominent peak in 222,30 cps, corresponding to 2Θ = 29,8776°. This peak is characteristic of the calcium carbonate existent in rigid lobster carapace that was retired after the first chemistry process.

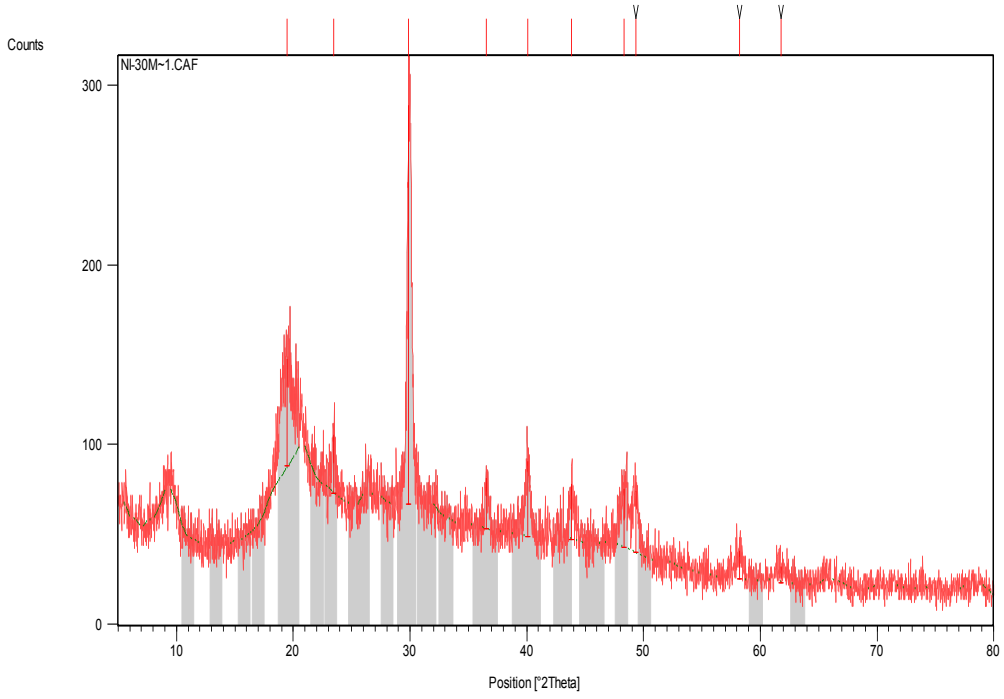

Figure 3. XRD of the initial powder.

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The diffractogram (Fig. 4) represents the demineralization step. It can be obervar the crystal structure of the powder decalcified, with several prominent peaks and being more intense at (243.31) and (484.55) cps, corresponding to 2Θ = 9.393° and 2Θ = 19.011 °.

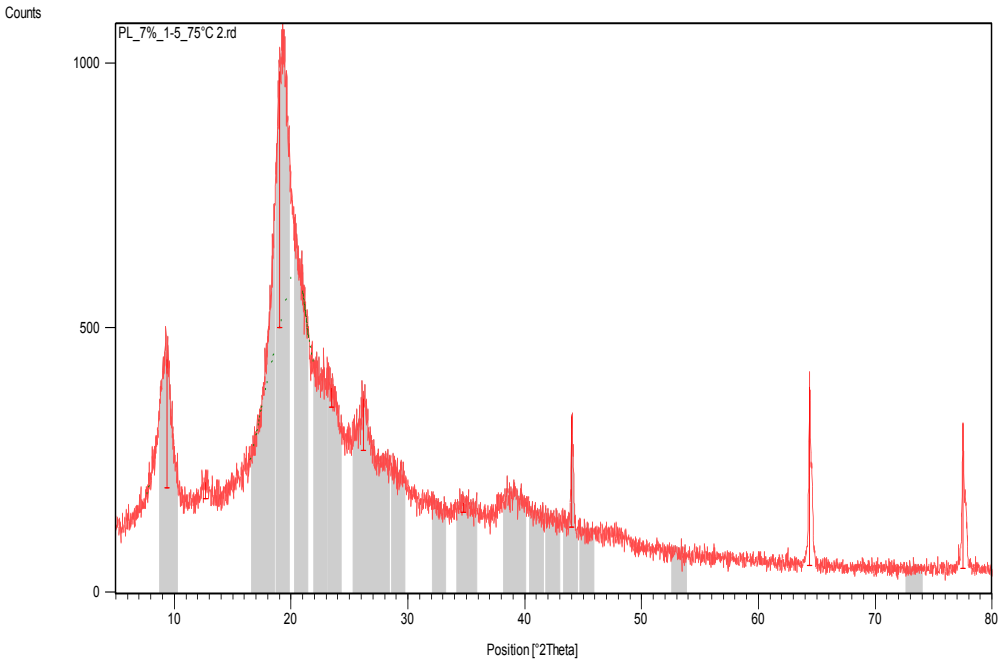

Figure 4. X-ray Diffractogram from 5 to 80 ° (2Θ). Demineralization phase.

Fig. 5 is the graph referring and deproteinization. This phase was characterized chitin and its crystal structure, with the most intense peaks (524.24) and (1151.26) cps, corresponding to 2Θ = 9.4040 ° and 2Θ = 19.1916 °.

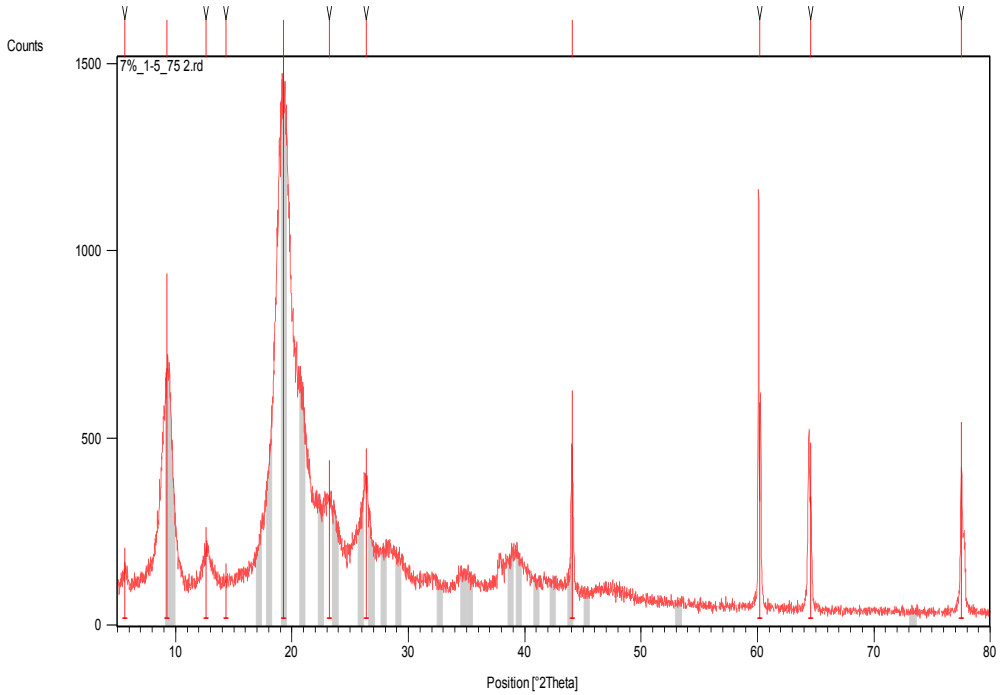

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Figure 5 - X-ray Diffractogram from 5 to 80 ° (2Θ). Chitin lobster - Palinurus elephas.

In fig. 6 is a graph of deacetylation of chitin. Chitosan is deacetylated chitin compound and thus has a semicrystalline structure because the terminal amino groups contribute to an amorphous structure for the secondary connections together as hydrogen and contributes to the change in the angle of connection between the chitosan molecules. It is exactly the semicrystalline structure of chitosan that assists in the process of epithelial healing, because the potential for absorption and degradation of exudates from ulcers. It is observed in Fig. The three most intense peaks (57,664.11) and (88,714.92) cps corresponding to 2Θ ° = 19.2878 ° and 2Θ = 9.5706, this graph shows that the deacetylation was performed material was removed all impurities and chemical structures is not necessary.

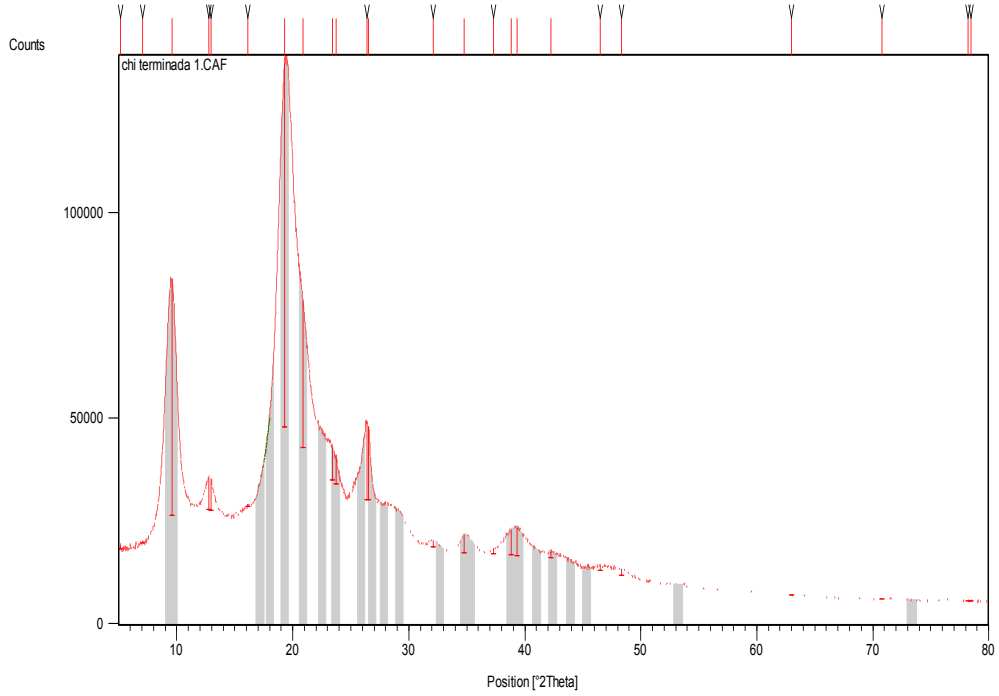

Figure 6 - X-ray Diffractogram from 5 to 80 ° (2Θ). Chitosan lobster - Palinurus elephas.

4. CONCLUSION

It was concluded that from the characterization of the biomaterial chitin has a very crystalline structure which becomes necessary for deacetylation of the material so that it carries the desired function healing. Thus becoming an alternative to the manufacture of dressings epithelial value, since chitosan is from residues of fish industry and food industry.

5. REFERENCES

Canella, K. M. C.; Garcia, R. B. Caracterização de quitosana por cromatografia de permeação em gel: influência do método de preparação e do solvente. Química Nova, v. 24, n. 1, p. 13-17, 2001.

Chuang, W.Y; Young, T.H.; Yao, C.H.; Chiu, W.Y. “Proprieties of the poly (vinyl alchol)/chitosan blend and its effect on the culture of fibroblast in vitro” Biomaterials, v. 20, p. 1479-1487, 1999.

Ferreira, A.B.H., 1999. New Aurélio XXI centry: O Dicíonário da Língua Portuguesa. 3. Ed. Rio de janeiro: Nova Fronteira, p. 1.655.

Griffith, L.G. Emerging design principles in biomaterials and scaffolds for tissue engineering, Ann. N. Y. Acad. Sci., v. 961, p. 83-95, 2002.

Hsieh, W.C.; Chang, C.P.; lin, S.M. “Morphology and characterization of 3D microporous structured chitosan scaffolds for tissue engineering.” Colloids and Surfaces B: Biointerfaces. v. 57, p. 250-255, 2007.

Junqueira, L.C.; Carneiro, J. Biologia Celular e Molecular. 8.ed. Rio de Janeiro, Guanabara Koogan, 2005.

Martin, Paul. Wound Healing-Aiming for perfect Skin Regeneration. Sience, v. 276, n. 5309, p. 75-81,1997.

Ratner, B.D.; Hoffman, A.; Schoen, F.J.; Lemons, J.E. Biomaterial Science. In: Ratner, B.D. Biological Testing of Biomaterials. Academic press, 355-396, 2004

Rigau, J. Acción de la luz laser a baja intensidad em La modulación de La función celular. 1996. 211p. Dissertação (Doutorado em Medicina). Universitat Rovira i Virgili.

Ross, Michael L.; Reith, Edward J.; Romrell, LYNN J. "Histologia Texto e Atlas". São Paulo: Panamericana, 1993.p. 47-115; 347-376.

Sacchetin, P.S.C. "Incorporação de flavobacterium columnare inativado em micropartículas de alginato e quitosana para a imunização de tilápia do nilo (oreochromis niloticus) por via oral." Faculdade de Engenharia Química, Universidade Estadual de Campinas, 2009. Dissertação (Mestrado)

Tonhi, E.; Plepis, A. M. G. Obtenção e caracterização de blendas colágeno-quitosana. *Química Nova*, v.25, p.943-948, 2002.

Veiga, I.G. Uso de xantana em substituição ao alginato em membranas coacervadas de quitosana projetadas para a cicatrização de lesões de pele. 2009. Dissertação (Mestrado em Engenharia Química) - Faculdade de Engenharia Química, Universidade Estadual de Campinas, Campinas.

6. RESPONSIBILITY NOTICE

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