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Editors:
Caue Ribeiro
Odílio Benedito Garrido de Assis
Luiz Henrique Capparelli Mattoso
Sergio Mascarenhas

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Studying the possible application of chitosan in the formulation of bovine rumen delivery systems

N. L. G. D. de Souza (PG)^{1*}, H. M. Brandão (PQ)², J. C. Carneiro (PQ)², J. M. Marconcini (PQ)²,
A. C. A. Duque (PG)² and L. F. C. de Oliveira (PQ)¹

⁽¹⁾ NEEM - - Departamento de Química – ICE - Universidade Federal de Juiz de Fora

⁽²⁾ Embrapa gado de leite de Juiz de Fora.

*nelson.luis@ice.ufjf.br

Abstract – Chitosan is a linear polysaccharide obtained from the deacetylation of chitin, and the physico-chemical properties of chitosan are dependent on the average degree of acetylation and the average molar mass. In this study it has been evaluated the stability of chitosan in the ruminal environment by means of infrared and UV-vis spectroscopic techniques in order to determine the possible use in ruminal bovine delivery systems.

Devices for the administration of intra-ruminal drugs are designed to provide a long delivery period and release of drugs in the reticulum and rumen of bovines. It is well known the administration of drugs during the grazing is complicated by the need to manipulate the animals before administering the drug; for circumventing this problem, as well as to decrease the costs and improve efficiency of drug, several types of sustained release devices have been developed to satisfy this need [1]. Chitosan, a natural linear biopolyaminosaccharide, can be obtained by alkaline deacetylation of chitin, which is the second more abundant polysaccharide next to cellulose; it has been also used for building delivery systems, due to its biocompatibility, high charge density, non-toxicity and mucoadhesion [2]. The purpose of this work is to analyze the chemical stability of chitosan in the ruminal environment of bovines in order to determine its possible use as a carrier for ruminal drug delivery.

The experiment was carried out in the experimental farm of Embrapa Dairy Cattle Research Center, located in Coronel Pacheco-MG (Brazil), using three *girolando* cows adapted with a ruminal fistula. The animals had free access to water, but with restricted feeding at pasture *braquearea ad libitum*. Two chitosan samples of different density were appropriately incubated in bovine rumen and removed daily, analyzed by vibrational (infrared) and absorption spectroscopy (UV-visible), for seven days.

The vibrational infrared spectrum in the of chitosan before incubation showed some characteristic bands, as for instance the one at 3438 cm⁻¹ which is assigned to $\nu(\text{O-H})$, superimposed with the NH stretching band; the bands at 2921 and 2877 cm⁻¹ can be assigned to $\nu(\text{C-H})$ modes. The band at 1650 cm⁻¹ can be related to the $\nu(\text{C=O})$, also known as amide I mode; the band at 1596 cm⁻¹ is assigned to scissoring mode of the (N-H) group; the band at 1421 cm⁻¹ is attributed to the symmetrical deformation of CH₃ and CH₂ groups; in the region between 1310 and 1380 cm⁻¹ are bands which can be assigned to $\nu(\text{C-N})$ belonging to the amide III mode. Other bands can also be assigned, such as the ones at 1256 cm⁻¹ (twisting vibration of O-H), 1080 cm⁻¹ (hydroxyl stretching), 1033 cm⁻¹ (C-O-C of glucose stretching), as well as the glycoside bond in chitosan, related to the $\beta(1 \rightarrow 4)$ bond, assigned to bands at 1154 cm⁻¹ and 897 cm⁻¹ [3]. The infrared spectra of chitosan after incubation indicate changes in the macro structure, such as the depolymerization of the polymer chain with subsequent ring opening monomers and nitrogen consumption. These changes can be inferred by some deviations in the spectral region of the $\beta(1 \rightarrow 4)$ glycoside bond in chitosan, the C-O-C stretching of the glucose species and the bands related to the amine and amide groups, all of them described above. The analysis by UV-visible spectroscopy show the appearance of a new electronic transition around 250 and 280 nm, which can be ascribed to double bonds formed after the main chain breaking of the polymer, followed by the ring opening [4]. However, this band does not increase in intensity with time, and can not be made straight relationship between the amount of chitosan degradation and the time.

As a preliminary conclusion, the interaction between chitosan and the rumen is reproductive and is followed by some changes in the chemical structure; in this sense, the use of chitosan in systems for controlled release ruminal delivery systems becomes difficult, because the low stability of chitosan in the ruminal environment.

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