STUDIES OF GENTAMYCIN ADSORPTION ON CHITOSAN

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Abstract
Chitosan as a dietary supplement has been used in the treatment of obesity due to its capability of binding bioactive compounds. A molecule of a cationic polymer, such as chitosan, may bind gentamycin. The aim of the study was to determine the binding capability of Gentamycin by chitosan's contained in weight-loss supplements. The Gentamycin absorption was investigated by means of a dynamic method in a biopharmaceutical model imitating in vitro conditions including the required level of nutrients. Mean absorption of gentamycin by chitosan ranged from 2.1% to 34.1% depending on the environmental pH. The highest absorption rate was observed at above pH 6.8. Concluding, the studies confirmed that the investigated gentamycin interact with chitosan, what leads to decrease in their amount and affects the bioavailability of the drugs.

Key words: *Gentamycin, chitosan; absorption.*

1. Introduction

The combinations of polymers and biopolymers with biologically active small-molecule compounds have become the subject of extensive investigations recently. A small molecular weight active substance when combined with a polymer often presents a modified action. On the other hand, the use of improper polymers may lead to drug-polymer incompatibility. Especially important are interactions in the form of adsorption and formation of complex bonds diminishing the effect of gentamycin. Chitosan, due to its capacity of binding lipids, cholesterol, fatty acids, triglycerides, bile acids, has been used in the treatment of obesity. It is an effective source of soluble fiber. It does not undergo digestion; it dissolves in the acid environment of the stomach, where it binds many molecules of water, forming a stable absorption gel. A molecule of a cationic polymer, such as chitosan, is capable of binding acid drugs [1, 2].

For this reason the study was undertaken to explain the mechanism of interactions of antibiotics drug: Gentamycin with dietary supplements containing chitosan. Qualitative study of adsorption will be conducted in future work.

2. Materials and method

Natural chitosans (Primex, Chito-Clear, Huasu) with deacetylation of 85% to 95%, degraded by 5 to 30 kGy radiation dose were used in the study. Also, dietary supplements containing chitosan were used (Vitana®, Hitec Nutrition®, Chromdiet®, Bio-Active Tech-Food Trading®). The adsorption of the gentamycin was investigated by means of a dynamic method in a biopharmaceutical model imitating the conditions in vitro. The amount of gentamycin absorbed by chitosan was calculated from the concentration of the investigated drug prior to and after sorption. The calculated amounts of bound drug were used to calculate mean percentage of absorbed dose. 0.03 g portions of chitosan end equivalent drug (0,060 mg) using were weighed and put to 5 ml glass centrifuge vials and next 2 ml of 0.05 M HCl were added to achieve pH 2 of the solution, what corresponds to natural fasting gastric pH [5 - 8].

Next the drugs were added to the vials: Gentamycin the doses of 0.01 g. Next 0.1 M Na_2CO_3 was added to the vials to achieve pH 6.4 (corresponding to the duodenal pH) and stirred for 0.5 hour (300 r.p.m.). The solution was alkalized with sodium carbonate to achieve pH 7.0 - 7.6, corresponding to the pH of the intestinal juice. The samples were then incubated at 37 °C in a shaker (300 r.p.m.) for 2.5 hours, after which the samples were brought to room temperature and centrifuged (2100 ×g) for 20 minutes.

Then left at standstill in order to align the layers for a further 30 min. The mixture was filtered through filter paper, and the clear solution was diluted 1:1 with 0.05 M HCl using a spectrophotometric method were tested at a wavelength of 218 nm. The concentration of antibiotic in the test solution was calculated by interpolation of the results of absorbance with respect to the equation of a straight absorbance antibiotic concentration in the solution and the result obtained by multiplying the value of the sample used in the dilution.

Measurements were carried out on the basis of three independent sample, which lead to the arithmetic and assessed statistically. Calculation of standard deviation was made, and then determining the coefficient of relativity repeatability was calculated. Standard deviation ranged from 0.001 to 0.057 and the determined relativity coefficient ranged from 1.81% to 4.82%, what confirms high accuracy of measurements [9 - 12].

Research of chitosan viscosity used in the work was carried out by. the procedure described in previous work [12].

3. Results and discussion

3.1. Gentamycin binding by degraded and non-degraded chitosans

Examination of adsorption of the investigated drug: Gentamycin confirm that the amount of bounded drug depends on the degradation rate of chitosan and its origin. The analysis of the effect of intrinsic viscosity on the capability of absorption of the investigated drug by chitosan's reveals an increase in the amount of absorbed drug [2].

Regardless of the used chitosan an increase in the adsorption of the antibiotic on the chitosan with medium viscosity is noted. Low and high viscosity Chitosans are characterized by reduced adsorption, only chitosan type 352 for the variety of high viscosity has good adsorption properties. Presumably, this is due to the adsorption of particles with atoms with free electron pairs, and we have to expect a qualitative analogy of their specific adsorption to the amino groups of chitosan and probably there is the impact of a large dipole moment of the-NH_2 groups and blocking the active sites of the adsorbent surface by the solvent is observed.

The analysis of viscosity-average molecular weights demonstrated that absorption of individual drug increases with decrease in intrinsic viscosity of all the investigated chitosans. The decrease in molecular weight induces lower strength of the polymer chain and the capability of formation of larger branches in the polymer network (***Figure 1*** see page 178).

The binding of Gentamycin by chitosan's contained in dietary supplements confirms the hypothesis of aggregative character of chitosan in relation to these drug. The highest amounts of drug are bound by chitosan contained in Vitana® dietary supplement (***Figure 2*** see page 178).

The investigated chitosan's contained in dietary supplements available on the weight-loosing products market are capable of binding on an average of 0.292 – 0.371 g of Gentamycin per 1 g of chitosan end equivalent drug (0.06 g) . The highest absorption rate was observed at pH above 7.6 (intestinal juice).

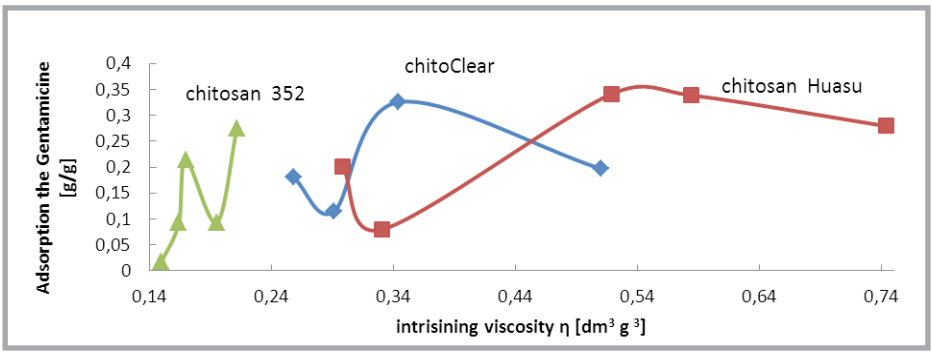

Figure 1. *The graph illustrating the dependence between binding the Gentamycin near dose 0.01 g in relation to essential intrinsic viscosity [η] (temp. 37 °C and pH 7.6).*

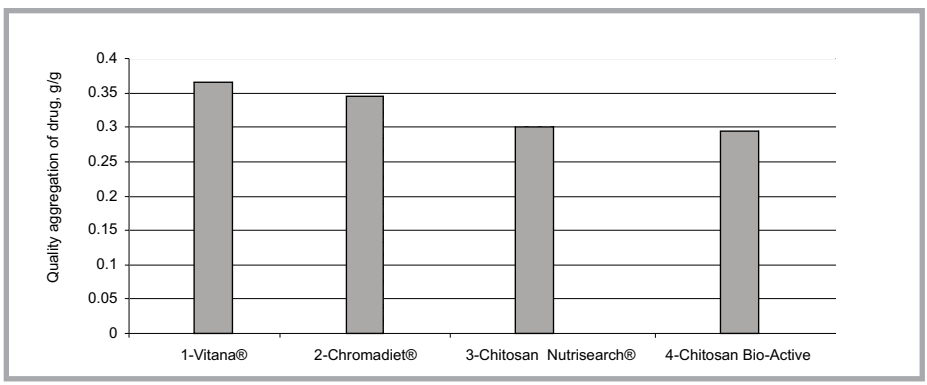

Figure 2. *The amount of Gentamycin bound by 1 g of chitosans present in dietary supplements in g/g (temp. 37 °C and pH 7.6).*

4. Conclusion

In conclusion, the studies have shown that there is an interaction between drug and chitosan, what decreases their quantity and affects the bioavailability of these drugs. At pH above 7.6, corresponding to the environment of the intestinal contents, the mean absorption for the highest dose of the drug on chitosan ranged from 2.1 to 34.1%, what is associated with an increased essential surface of the polymer and its sorption capability. Basing on the above considerations, it can be stated that the investigated drug and polymer interact antagonistically in the form of absorption of drug from this group on a polymer such as chitosan.

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