EVALUATION OF MOLECULAR WEIGHT OF CHITOSAN IN THIN-LAYER AND SPOUTED BED DRYING

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ABSTRACT

Chitosan is chitin in deacetylated form and is the main constituent of crustacean exoskeletons. Commercially, chitosan is dried in tray driers, and during the operation, polymerization may occur as the chitosan is composed of carbohydrates. The aim of this work was to analyze chitosan in spouted bed and thin-layer drying, considering viscosity average molecular weight of the chitosan samples in the process. Results showed that spouted bed-dried chitosan presented a molecular weight value similar (160 kDa) to that of the raw one (150 kDa). However, when dried on tray dryers, the molecular weight was 300 kDa, indicating that molecule polymerization occurred.

PRACTICAL APPLICATIONS

Chitosan, a deacetylated form of chitin, is considered a functional food. The use of chitosan in various areas is directly related to the molecular weight and degree of deacetylation, which depends on the conditions of chitin deacetylation and chitosan drying. One of the main steps of the chitosan production process is drying. Industrial chitosan is dried in tray dryers; however, it is composed mainly of carbohydrate monomer units capable of undergoing polymerization during the drying operation. The spouted bed dryer is a method that promotes direct particle–fluid contact, with high rates of heat and mass transfer. Also, it presents an alternative to the spray-dryer in an attempt to obtain dry products with the same quality, at low costs. This work presents the study of chitosan drying in spouted bed and thin layer, in paste form, considering the viscosity average molecular weight of the chitosan samples in the operation.

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INTRODUCTION

Chitosan, a deacetylated form of chitin, is considered a functional food that presents unique properties in absorption and excretion of ingested fats (Rudrapatnam-Tharanathan and Kittur 2003). It is a substance that has been used in various industrial processes, such as pharmaceutical products, biomedical, food and treatment of effluent wastes (Chen and Hwa 1996; Ayers and Hunt 2001).

Chitosan characteristics depend on the degree of deacetylation, chain size and distribution of molecular weight (Tsai *et al.* 2002). When the degree of deacetylation of the material exceeds 50%, it becomes soluble in aqueous acid solutions and is called chitosan (Tsaih and Chen 1997). Because chitosan is commercially available with different degrees of deacetylation and molecular weight (Zhang and Neau 2001), it is important to take into consideration the effects of these parameters.

In some fields, especially in medicine and the food industry, the application of this polysaccharide is limited by its high molecular weight, which results in its low solubility in aqueous media (Ilyina *et al.* 2000). Chitosan is used in food as a clarifying agent and enzymatic browning inhibitor in apple and pear juices and in potatoes, and as an antioxidant in sausages. Chitosan can also be used as an antimicrobial film for covering fresh fruits and vegetables (Devlieghere *et al.* 2004). Because chitosan displays muco-adhesive properties, strong permeation-enhancing capabilities for hydrophilic compounds and a safe toxicity profile, it has received considerable attention as a novel excipient drug delivery system and has been included in the European Pharmacopoeia since 2002 (Mao *et al.* 2004).

Molecular weight distribution is influenced by factors such as time, temperature, reagent concentration and atmospheric conditions of the deacetylation reaction for obtainment of chitosan. In many cases, reduction of highly deacetylated chitosan with a very low degree of depolymerization is desirable because these parameters influence not only its physical-chemical characteristics, but also its biodegradability and immunological activity (Tolaimate *et al.* 2003).

One of the main steps of the production process of chitosan is drying. Industrially, chitosan is dried in tray dryers (Brzeski 1982); this operation is widely used in agriculture and chemical engineering, in deep or thin-layer drying (Sun and Woods 1997). Chitosan is composed mainly of carbohydrate monomer units capable of undergoing polymerization during the drying operation.

Spouted bed dryers have been indicated as the appropriate equipment for drying suspensions as well as for pastes and solutions (Costa *et al.* 2006). This method promotes direct particle-fluid contact, with high rates of heat and mass

transfer (Shuhama *et al.* 2003; Devahanstin *et al.* 2006). Also, it presents an alternative to the spray-dryer in an attempt to obtain dry products with the same quality, but at low costs (Cunha *et al.* 2000).

Spouted bed dryers with inert bodies have been extensively studied for drying bio-resources such as foodstuffs and phyto-pharmaceutical products (Marreto *et al.* 2006; Oliveira *et al.* 2008). Recently, spouted beds have been applied to drying medicinal extracts (Peixoto *et al.* 2004; Cordeiro and Oliveira 2005).

The objective of this work was to analyze chitosan drying in a spouted bed dryer and in a thin-layer dryer, as well as analyzing final product quality and evaluating the viscosity average molecular weight of chitosan.

MATERIALS AND METHODS

Raw Material

The studied raw material, purified chitosan, was obtained in the Laboratory of Unit Operations/FURG according to Weska *et al.* (2007).

Drying Chitosan

Spouted Bed. A schematic diagram of the equipment utilized in the spouted bed drying experiments is shown in Fig. 1.

The spouted bed drying experiments were performed in a CSB (conventional spouted bed) dryer constituted of a stainless steel cylindrical cone column with glass cones. The conical basis with enclosed angle of 60° had 0.15 m height and the cylindrical column had diameter and height of 0.175 and 0.75 m, respectively. Also, the drier presented a 1:6 ratio between column and air inlet diameters.

Air supplied to the system, provided by a radial blower with 6 kW power and 0.1 m^3/s maximum outflow, was heated in a system composed of three electric resistances with 800 W each. Heat control of the exit air stream was performed by a temperature controller. Drying air velocity was measured in an orifice meter. It measured the pressure drop through the stream bed with a U-tube manometer. Temperature readings were carried out in type K copper-constantan thermocouples. The dry chitosan was collected in a Lapple cyclone.

Inert particles used in the spouted bed were polyethylene "pellets" (diameter, 0.003 m; sphericity, 0.7; density, 960 kg/m^3). The cell was filled with 4.0 kg of inert particles, corresponding to approximately 80% of the maximum "spoutable" bed depth (H_m).

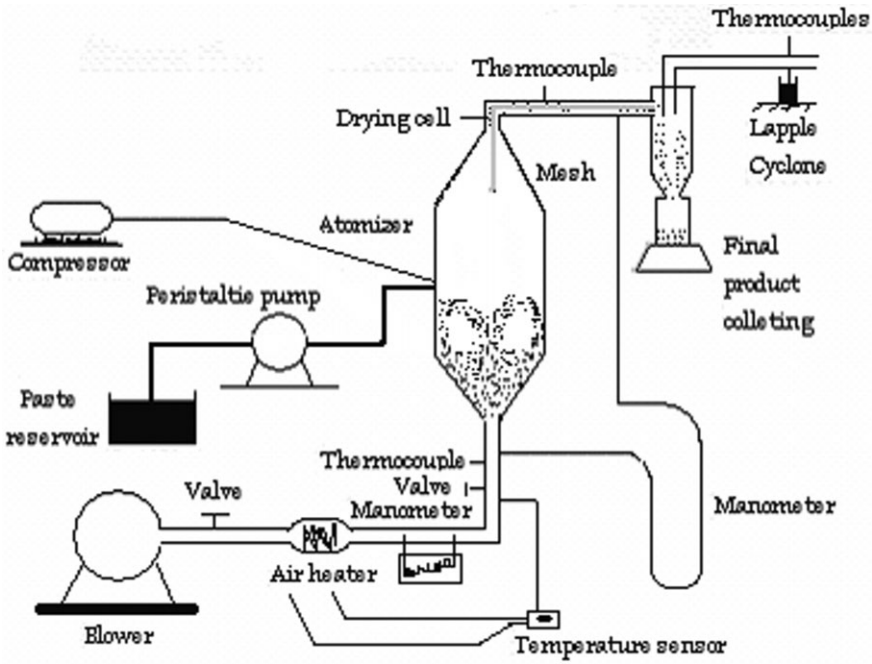

FIG. 1. SCHEMATIC DIAGRAM OF THE EQUIPMENT FOR SPOUTED BED DRYING

The experimental spouted bed-drying procedure was carried out as follows: initially, fluid dynamics of the equipment for determination of minimum spouting velocity were carried out, employing a circulation rate of 100% larger than the minimum spouting velocity, as recommended by Mathur and Epstein (1974), for drying of pastes.

After a steady velocity regime had been established, the sample feeding system was set in motion and the chitosan paste was fed to the inside of the cell through atomization, with feeding of 0.32 kg paste/h/kg inert by peristaltic pump and air compressed at a pressure of 10^5 Pa gauge. Inlet air temperature was fixed at 100C. Dry and wet bulb temperatures of the drying air were monitored in order to observe chitosan drying behavior. Chitosan, while in spouted bed drying, occurred by fluid–particle contact and also by friction between inert particles caused by the high rate of circulation of the particles in the spouted bed interior. Dried chitosan in powder form was transported pneumatically by the drying air stream and collected in a cyclone.

Tray Dryer. Chitosan in paste form was dried in a discontinuous tray dryer (Fig. 2), composed of a drying chamber of 1.8 m length, 0.25 m height

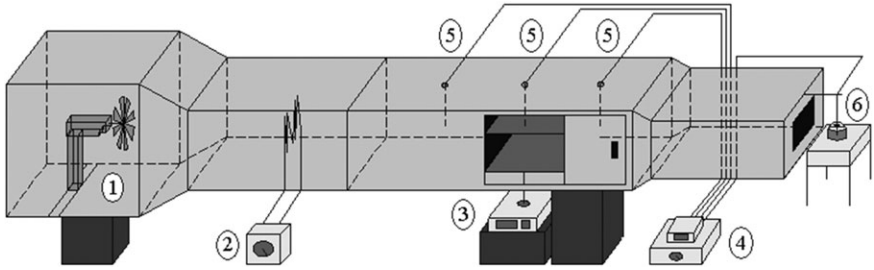

FIG. 2. DISCONTINUOUS TRAY DRYERS WITH PARALLEL AIRFLOW
(1) Fan. (2) Temperature controller. (3) Electronic scale. (4) Millivoltimeter with attached thermocouples. (5) Dry bulb thermocouples. (6) Wet bulb thermocouples.

and 0.2 m width. Stainless steel rectangular trays (0.12 m × 0.14 m), with a thickness of 0.004 m (one-side drying only), received a chitosan paste load of 4 kg/m^2. The chamber possessed a fan for moving the drying air, a heating system, thermocouples located at the dryer's exit and an anemometer for measuring air velocity. An electronic scale (AS2000C, Marte model, São Paulo, Brazil) with 0.01-g precision was used to measure mass of the utilized samples. Samples' thicknesses were measured with a digital caliper (model CD-6CS, Mitutoyo America Corp., Aurora, IL, USA) with 0.0001-m precision. Air relative humidity was measured with a thermohygrometer (model 3310-00, Cole Parmer, Vernon Hills, IL, USA) with 0.1% precision and air velocity with 0.1 m/s precision anemometer (Windmesser model MIT Thermometer, TFA, Germany).

The past-form chitosan was homogenized, weighed and placed in stainless steel trays. Drying experiments were carried out at air temperatures of 60C, air velocity of 2.5 m/s and a relative humidity of approximately 18% (Batista *et al.* 2007) until reaching commercial moisture content of the product (5–8% wet basis [w.b.]). Samples were weighed every 5 min for constructing drying curves.

Analytical Methodology

Centesimal chemical composition of wet chitosan was determined through analytical methods according to the Association of Official Analytical Chemists (1995): ashes, moisture and total-N (using conversion factor of chitosan 11.5), as well as moisture content of the final product.

Chitosan samples' color was analyzed by colorimetric method. Caramel color (1 g) was dissolved in an acetic acid solution (1% p/v) and afterwards stirred for 10 min. The solution was completed up to 50 mL with demineralized water and filtered through a medium porosity glass filter for removal of

trace impurities, resulting in a 0.02 g/mL solution. The following procedure was adopted: 0.6, 0.8, 1.2, 1.4, 1.6, 1.8 and 2.0 mL of caramel solution were introduced into test tubes, followed by different volumes of acetic acid solution (1% p/v) in order to obtain 6 mL. Absorbance values were measured at 420 nm with a digital spectrophotometer (Micronal I, model B 342 II, São Paulo, Brazil). The analytical curve was obtained after plotting the absorbance readings for the eight concentrations in milligram caramel/mL solution. Afterwards, the chitosan samples (0.5 g in dry basis [d.b.]) were dissolved in 50 mL of acetic acid solution (1% p/v), stirred and filtered for reading absorbance in 420 nm. Then, chitosan samples' color was plotted by analytical curve (expressed in gram caramel/g sample).

Grain-size analysis of the dry samples was carried out in a standardized mesh screen. The average diameter was calculated by the definition of Sauter, as shown in Eq. (1):

$$\bar{D}_{\text{Sauter}} = \frac{1}{\sum \frac{\Delta X_i}{D_{\text{mi}}}} \tag{1}$$

where $\bar{D}_{\text{Sauter}}$ is the average diameter of Sauter (m), ΔX_i is weight fraction of particles' size, and D_{mi} (%) and D_{mi} is the arithmetic average diameter between two screens (m).

Molecular Weight Determination. The viscosity average molecular weight (M_w) of chitosan samples was determined by viscosimetry method (Zhang and Neau 2001).

Chitosan samples were prepared in an aqueous solution of acetic acid 0.2 M and sodium chloride 0.1 M at five different concentrations (0.001–0.012 kg/L). For analysis of samples' viscosity, a Cannon-Fenske capillary viscometer (model Schott Gerate GMBH-D6579, Hofheim, Germany) was used, placing 10^{-2} L of the sample in the capillary at 25 ± 1C. Knowing the time flow $t(s)$ and the capillary constant k (m^2/s^2), kinematic viscosity v (m^2/s) of the samples was determined as shown in Eq. (2):

$$v = kt \tag{2}$$

The specific viscosity (η_{sp}) was determined by Eq. (3):

$$\eta_{sp} = (\eta_{solution} - \eta_{solvent})/\eta_{solvent} \tag{3}$$

Reduced viscosity (η_{sp}/c) was determined by the relation between specific viscosity and concentration. The graph of reduced viscosity (η_{sp}/c) against the

concentration (c) of the samples' solutions gives the intrinsic viscosity (η) of the solution. In order to estimate its value, Huggins equation was used, according to Simal (2002), as shown by Eq. (4):

$$\frac{\eta_{sp}}{c} = [\eta] + A[\eta]^2 c \tag{4}$$

Viscosity average molecular weight (M_w) was calculated based on the Mark-Houwink-Sakurada equation (Eq. 5):

$$[\eta] = KM_w^{\alpha} \tag{5}$$

where $K = 1.81 \times 10^{-3}$ and $\alpha = 0.93$ for chitosan in a solvent system consisting of acetic acid and sodium chloride at 25C, according to Simal (2002).

RESULTS AND DISCUSSION

Chemical Composition

Raw material results for characterization of purified chitosan were N-chitosan of 4.71 ± 0.4%, ashes of 0.6 ± 0.1 and moisture content of 95.1 ± 0.4; all values were in wet basis.

Chitosan initial moisture content used for the drying experiments corresponded to 19.6 ± 2.1 on a dry basis.

Drying in Thin Layer

Figure 3 presents the drying rate (N) versus moisture content on a dry basis (X), obtained by numerical derivation of the moisture content curve of the sample, on a dry basis, as a function of time at 60C. The existence of two distinct drying periods is verified in this figure: constant rate and falling rate.

In Fig. 3, the critical moisture content (X_c) that represents the end of the constant rate period (horizontal portion of the drying rate curve) was 5.0 kgH_2O/kg d.b., which is close to the value found by Batista *et al.* (2007). The importance of the constant rate period can be noted in this figure; it presented practically the same time duration as the falling rate period, which was around 60 min. This is because of the high value of initial moisture content value of the sample, corresponding to approximately 95% on a wet basis, requiring a prolonged time interval for the removal of unbound moisture present in the surface material. Constant drying rate was 0.28 kg/kg/min (0.0047 kg/kg/s). From the critical moisture content, the falling rate period can

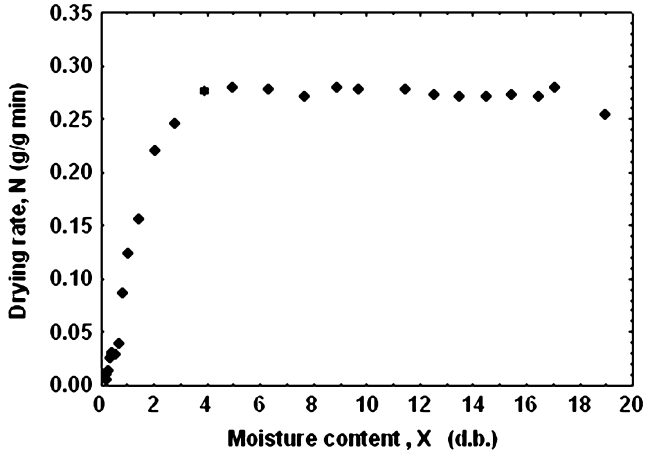

FIG. 3. DRYING RATE CURVE VERSUS CHITOSAN MOISTURE CONTENT AT 60C

be found where the drying rate is controlled by the moisture transport in the interior of material (Strumillo and Kudra 1986). The total time necessary for reaching the commercial moisture content of the product (5–8% w.b.) was 116 min, resulting in a 2 kg/h/m^2 capacity of chitosan paste.

Drying in Spouted Bed

Figure 4 presents the pressure drop graph in the type CSB versus superficial air velocity for the experiments with an inert particle load of 4.0 kg and inlet air temperature at 100C. The figure shows classic behavior for this geometry type as presented in Mathur and Epstein (1974). In Fig. 4, through the increasing and decreasing gas flows curves, a minimum spouting velocity (v_{jm}) of 0.63 m/s, as well as the maximum value (5,340 Pa) and stable value (1,520 Pa) of pressure drops, were determined for the bed.

It was observed that the chitosan paste residence time in the drying spouted bed was around 15 min, reaching the commercial moisture content of the product (5–8% w.b.). This time was determined when the outlet air temperature reached a constant value. Tacon and Freitas (2007) evaluated the effects of paste properties on residence time during drying in a spouted bed dryer with inert bodies of the polyethylene bed. These authors determined that the mean residence times varied from 12.2 to 17.7 min.

Inlet air temperature was fixed at 100C, resulting in an outlet air temperature of 75C, in order to guarantee final product quality. Marreto *et al.* (2006) evaluated the performance of CSBs with inert bodies in the drying of

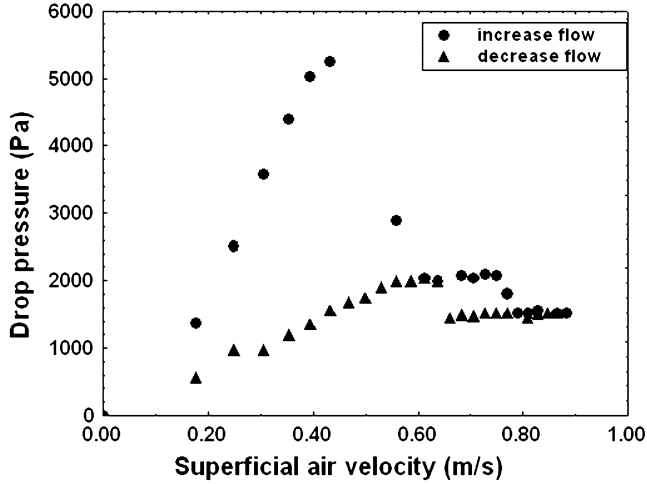

FIG. 4. PRESSURE DROP VERSUS SUPERFICIAL AIR VELOCITY CURVES OF SPOUTED BED WITH 4 kg OF INERT PARTICLES AT 100C

pharmaceutical paste based on dry-product quality. The authors demonstrated a strong dependence of inlet air temperature on the bixin and caffeine contents in annatto and guarana dry powders.

Samples were collected at 30-min intervals, in a total drying operation time of 2 h. The first 30 min was rejected, assuming that the spouted bed reached a stabilized state. Average results for moisture content and production of dry chitosan in the spouted bed-drying experiments were in the range of 5–7% and 51–57 g/h, respectively. An analysis of variance was carried out with the values of moisture content and production, and it was observed through this analysis that the answers presented no significant difference, considering a 95% level. This demonstrates the stability of the spouted bed during the running time of drying, and moisture content values were within the range of commercial chitosan.

The average Sauter diameter, according to Eq. (1), was 0.21×10^{-3} m (60–80 mesh). Chitosan particles between 40 and 140 mesh are applied in the food area, and chitosan studied in this work falls within this range.

Viscosity Average Molecular Weight and Color

Figure 5 presents the reduced viscosity graph as a function of samples' concentration.

Values of calculated intrinsic viscosity (Eq. 4), viscosity average molecular weight (Eq. 5) and color variation for the chitosan samples, *in natura*, dried

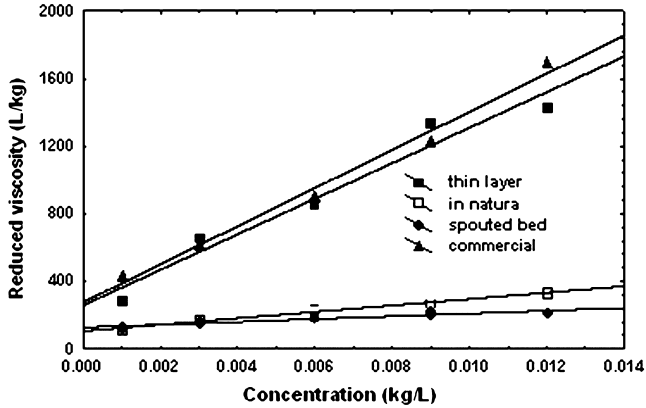

FIG. 5. REDUCED VISCOSITY GRAPH AS A FUNCTION OF CONCENTRATION FOR THE CHITOSAN SAMPLES

TABLE 1.
INTRINSIC VISCOSITY (η), VISCOSITY AVERAGE MOLECULAR WEIGHT (M_w) AND COLOR VARIATION OF THE CHITOSAN SAMPLES

Chitosan	(η)* (L/kg)	M_w^* (kDa)	Color variation (%)	Correlation coefficient (R)
In natura†	119 ± 2	150 ± 3	–	0.99
Dry spouted bed	126 ± 2	161 ± 3	12.5 ± 1.4	0.96
Dry on trays	230 ± 4	306 ± 6	41.4 ± 2.2	0.96
Commercial	285 ± 5	387 ± 6	–	0.99

* Mean values ± SD (in replicate).
† Color value of *in natura* sample: (0.36 ± 0.02) g caramel/g sample.

on the spouted bed, dried in trays and dried commercially, are found in Table 1. Linear regression used for the calculation of the intrinsic viscosity of each sample presented a good adjustment, as demonstrated by the correlation coefficient (R) values in Table 1.

According to Table 1, it is evident that the chitosan molecule practically did not present structure alterations when dried in a spouted bed, as observed through its viscosity average molecular weight value when compared with the *in natura* sample value. The minimal color deterioration during spouted bed drying (Table 1) is an appropriate indication that this process preserves chitosan. This is due to the fact that the equipment is capable of forming many uniform layers of the material, thus recovering the inert particles of the spouted bed, which possesses a high circulation rate, presenting excellent conditions of

heat and mass transfer in a short retention time (Shuhama *et al.* 2003), and forming a high-quality product (Nindo *et al.* 2003).

However, when the chitosan was tray-dried, a considerable increase of viscosity average molecular weight and color was observed. Therefore, in this equipment, it is possible to consider that all the bed material is submitted to the same drying air conditions of temperature and humidity, and submitted to the same conditions for a prolonged time, which caused polymerization. Nonenzymatic browning of foods proceeds in several ways. One of the most important is caramelization reaction, in which carbohydrates are heated in the absence of nitrogen-containing compounds. In this reaction, the carbohydrates initially undergo dehydration and then polymerization into complex molecules of varying molecular weight. Lightly colored materials occur in the initial stages, but as the reactions continue, higher molecular weight bodies are produced and the color becomes more accentuated. Molecular weight and color variation values of tray-dried chitosan and the commercial chitosan have been similar (Table 1).

According to Krokida and Maroulis (1997), considerable changes in the physical structure of products, such as reduction in volume and decrease in porosity, can be found during drying. In the case of AD, reduction of volume is usually accompanied by wrinkles, deformation and even change in color, indicating the collapse of air-dried products. This behavior has been observed for most types of foodstuffs (Ratti 1994). Srinivasa *et al.* (2004) studied the properties of chitosan films prepared under different drying conditions. Chitosan-based packaging films were prepared by employing wet casting followed by two drying methods at 80, 90 and 100C, oven-drying (OD) and infrared-drying (ID), and the film properties were compared with those prepared under ambient temperature drying (AD ≈ 27C). The authors determined that OD showed a higher color index compared with ID and AD films.

Chitosan dried in spouted bed presented a higher quality product; therefore, its viscosity average molecular weight was close to the range of the chitosan applied to the pharmaceutical area, 130–205 kDa (Carreño-Gómez and Duncan 1997), while commercial and tray-dried chitosan were close to the value recommended for industrial application, i.e., 300–400 kDa (Giunchedi *et al.* 1998).

CONCLUSIONS

The spouted bed, in the conditions employed for chitosan paste-drying, presented stable behavior during the running time, and the final product attained the commercial moisture range (5–8% w.b.) at a retention time of 15 min.

Observing the results obtained in thin layer-dried chitosan samples, it could be stated that in order to attain the desired moisture content in the drying conditions, the total drying time was approximately 2 h, with the constant rate period being equivalent to the falling rate period.

According to the results, it was also observed that the viscosity average molecular weights of chitosan dried in spouted bed and *in natura* are close to 150–160 kDa. On the other hand, an increase in molecular weight was observed with tray-dried chitosan (300 kDa), indicating polymerization therefore decreasing the final product's quality.

NOMENCLATURE

C	concentration of the samples	kg/L
$\overline{D}_i$	average diameter between screen	m
$\overline{D}_{Sauter}$	Sauter average diameter	m
ΔX_i	retained mass fraction	dimensionless
ΔXp	passing mass fraction	dimensionless
k	capillary constant	m^2/s^2
N	drying rate	$kgH_2O/kgds$
R	correlation coefficient	dimensionless
X	moisture content (dry basis)	$kgH_2O/kgds$
M_w	viscosity average molecular weight	kDa
t	flow time	s
$[\eta]$	intrinsic viscosity (Eq. 4)	L/kg
η_{sp}/c	reduced viscosity (Eq. 4)	L/kg
$\nu_{solution}$	kinematic viscosity of solution	m^2/s
$\nu_{solvent}$	kinematic viscosity of solvent	m^2/s
(η_{sp})	specific viscosity	dimensionless
ν	kinematic viscosity	m^2/s

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