Article

PRODUCTION OF K-CARRAGEENAN BEADS BY PRILLING PROCESS[†]

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Running title: Production of K-carrageenan beads by prilling process

ABSTRACT

The objective of this study is to prepare, evaluate and understand carrageenan gelation. By using these properties of κ-carrageenan, beads are created by thermal gelation. A warm κ-carrageenan solution is extruded dropwise into a cold air column; the particles are collected. The relationship between the process variables on the shape and size of κ-carrageenan drops after gelation is established with the aid of image analysis. Thanks to these measurements, a model is developed to present satisfactory performance in industrial application.

Mathematical model for κ-carrageenan prilling, its simulated results, the parameters of equipment for the innovated process and its application are addressed. This article is protected by copyright. All rights reserved

Keywords: κ-carrageenan, prilling tower, thermal gelation, sphericity, mathematical modeling

INTRODUCTION

One of the most usual methods of encapsulation of biocatalysts, especially living cells, is entrapment in alginate beads. The active is dispersed in alginate solution, extruded dropwise in calcium bath, converting drops in hydrogel beads by gelation.[1] The method is soft, realized at room temperature and alginate is probably the most efficient gelling material.[2] The main drawback of this method is the gel sensitivity to chelating compounds such as phosphate or citrate.[3]

κ-carrageenan has been proposed as alternative to alginate.[2] This polysaccharide is gaining popularity as a material for bioencapsulation, linked to its ability to form a thermo-reversible gel.[4] It provides stronger and more stable beads than alginate. Two approaches to form κ-carrageenan beads are mainly reported in the literature:

- A warm solution of κ-carrageenan is extruded dropwise in a cold KCl solution;[5]
- A warm solution of κ-carrageenan is dispersed in oil. Then, the resulting emulsion is dropped in cold KCl solution.[6]

In these methods, potassium ions replace sodium ions in the gel network leading to more compact and solid gel. However, the main process of gelation is due to the temperature drop. The first method provides generally large and quasi mono-disperse beads, while the second method produces small size-dispersed beads. The second method is easier to scale-up.[7]

Most researches stop at the stage of collecting wet beads but, industrially, beads must be dried before storages transport and application. All methods described above require filtering and washing the beads. On top of the cost associated to these operations, part of the encapsulated active may be release, reducing the encapsulation efficiency. For large quantity, drying is realized by passing air through the beads. However, operation is difficult due to the sticking behaviour of the wet beads. Often the process is inefficient and bead agglomeration is observed.

Tosa et al.[8] have proposed an alternative consisting in extruding drop wise a warm κ-carrageenan solution directly in cold air. Resulting beads may be collected or directly directed

to a dryer. Such an approach would avoid filtration and washing steps; the drying process is simplified and more efficient.

The aim of this work is to experiment κ-carrageenan droplet gelation in cold air, propose and identify a model and through some simulations analyze the impact of different parameters on the process performance.

MATERIALS AND METHOD
Preparation of κ-Carrageenan Solution (50 g/L)

Fifty grams of κ-carrageenan (Carra GEL PGU 5289, supplied by Gelymar, Chile) are suspended in 1L distilled water at room temperature, heated at 80 °C and stirred for 1 hour until complete dissolution. The solution is then cooled down to 70 °C before beads production.

Differential Scanning Calorimetry (DSC) Measurements

DSC measurements are performed with DSC Q-VII (TA Instruments, UK) equipped with 30 mg carrageenan solution sample in sealed aluminum pans and empty aluminum as reference. The cells are cooled to 20 °C, equilibrated for 1 min, then heated to 90 °C. After heating and equilibrated at 90 °C for 5 min, the sample is cooled down to 20 °C. This cycle is carried out twice. The rate of heating/ cooling is set at 1°C/min.

Experimental Set-Up and Bead Production

Figure 1

The experimental set-up for producing κ-carrageenan beads is presented in Figure 1. κ-carrageenan solution is introduced in 1 L bottle (C) maintained at 70 °C. The bottle is placed under pressure controlled by a manometer (B) leading to solution extrusion dropwise through a nozzle (D). The droplets fall inside a column (E) fed with airflow provided by a fan (G) and cooled down by passing through a cylinder containing dry ice (I). The jellified droplets are collected at the bottom of the column (F). Experiments were run by varying the temperature and height of the cooling column.

Bead characterization: Size and Sphericity Factor (SF)

κ-carrageenan beads were observed using a digital microscope (Dino-lite Pro, Carb & Diam). Mean size and standard deviation are estimated from 100 bead measurements. The sphericity factor (SF) indicates the roundness of the beads according to the following:

$$SF = 1 - \frac{2d_{min}}{d_{max} + d_{min}} \tag{1}$$

where d_{max} is the maximum diameter and d_{min} is the minimum diameter measured on the bead. Sphericity factor varies from unity for a perfect sphere to zero for an elongated particle. Beads may be classified[9] as spherical, oblate, egg-shape and tear-pear shape, based on the microscope observation and their sphericity factor value (Table 1).

THEORETICAL ASPECT

To define the conditions of thermo-gel bead formation in a cold air column, two questions have to be answered:

- How will the thermo-gel droplet fall in the column?
- How fast does droplet temperature drop to reach the gelation point?

Modelling of the Droplet Falling

In developing the model, several hypotheses have been taken in consideration:

- The droplets are spherical, assimilated to solid sphere,
- Turbulent regime is assumed (500<Re<200 000),
- If so, the Drag coefficient, C_D, is constant and equal to 0.44[10],
- As density of air (ρ_a=1,3 kg/m^3) is largely smaller than droplet density (ρ_d≈1030 kg/m^3), the Buoyancy force has been neglected.

In such conditions, the acceleration acting on the droplet is the gravity force minus the friction forces:

$$m\frac{du}{dt} = mg - \frac{1}{2}C_D\, S\, \rho_a\, u^2 \tag{2}$$

where m is the mass of the droplet ($\frac{\pi}{6}d^3\rho_d$), d the droplet diameter, S the section or projected area of the droplet ($\frac{\pi}{4}d^2$), g the gravity constant and u the droplet velocity. The gravity and drag forces will have a tendency to equilibrate, leading to a terminal velocity for the droplet:

$$u_t^2 = \frac{2mg}{C_D S \rho_a} \tag{3}$$

Combining Equations (2) and (3) results in the following:

$$\frac{u_t^2}{g}\frac{du}{dt} = u_t^2 - u^2 \tag{4}$$

Or by rearranging Equation (4), we get the following:

$$\frac{du}{u_t^2 - u^2} = \frac{g}{u_t^2}dt \tag{5}$$

After integration, one will obtain the following:

$$u = u_t \tanh\left(\frac{gt}{u_t} + \alpha\right) = \frac{dy}{dt} \qquad \text{with } \alpha = \operatorname{arctanh}\left(\frac{u_0}{u_t}\right) \qquad \text{for } u_o < u_t \tag{6}$$

$$u = u_t \qquad\qquad\qquad\qquad\qquad \text{for } u_o = u_t \tag{6a}$$

$$u = \frac{u_t}{\tanh\left(\frac{gt}{u_t} + \alpha'\right)} = \frac{dy}{dt} \qquad \text{with } \alpha' = \operatorname{arctanh}\left(\frac{u_t}{u_0}\right) \qquad \text{for } u_o > u_t \tag{6b}$$

where u_0 is the initial droplet velocity and y is the vertical distance between the droplet and the nozzle. By integrating Equation (6) and (6a), it results in the following:

$$y = \frac{u_t^2}{g}\ln\left(\cosh\left(\frac{gt}{u_t} + \alpha\right)\right) + \mathrm{k} \qquad \text{for } u_o < u_t \tag{7}$$

$$y = u_t t \qquad\qquad\qquad\qquad\qquad \text{for } u_o = u_t \tag{7a}$$

$$y = \frac{u_t^2}{g} \ln \left(\sinh \left(\frac{g\,t}{u_t} + \alpha' \right) \right) + k' \qquad \text{for } u_o > u_t \qquad (7b)$$

where k and k' are constants.

Modelling of the heat exchange

In this study, the following assumptions are considered:

- Heat transfer is limited by the transfer at the air-droplet interface,
- Temperature is homogeneous in the beads,
- Temperature is constant along the vertical axis in the cooling tube,
- κ-carrageenan is at low concentration (maximum 5 %; above this concentration, there is an extrusion problem (clogged nozzle)),
- Gelation enthalpy of the solution is low and covers a range of temperatures,
- Then, the specific heat capacity is similar between κ-carrageenan and water,
- No contraction is observed during gelation.

If so, the temperature profile could be defined by the following:[11]

$$m\,c_p\,\frac{dT_d}{dt} = a\,h\,(T_a - T_d) \qquad (8)$$

where c_p is the specific heat capacity, a the surface of the droplet ($\pi\,d^2$), T_a, and T_d respectively the air and droplet temperatures. The air-to-droplet heat transfer coefficient, h, could be predicted by the following equation:[12]

$$h = \frac{0{,}003\,\lambda_a\,Re^{1{,}8}}{d} \qquad (9)$$

with the Reynold number, $Re = \frac{\rho_a U_t d}{\mu_a} > 4000$ \qquad (10)

Combining Equations (8) and (9) leads to the following:

$$\frac{dT_d}{T_a - T_d} = \beta\,dt \qquad \text{where } \beta = \frac{6h}{\rho_d c_p d} \qquad (11)$$

By integration, one gets the following:

$$ln\frac{T_a-T_d}{T_a-T_{do}} = -\beta\ t \tag{12}$$

To confirm that the transfer at the interface is the limiting factor, the Biot number, *Bi*, has to be computed:

$$Bi = \frac{h\ d}{\lambda_d} \tag{14}$$

Biot number compares the transfer at the interface (h) and inside the droplet ($\frac{\lambda_d}{d}$), where λ_d is the heat specific conductivity inside the droplet. If Biot number is lower than 0.1, one may assume that the temperature inside the beads is homogeneous.

Combining Falling and Thermic Model

In order to ensure gelation of the beads, the temperature of the cooling column is fixed at carrageenan gelation temperature minuc 10 °C. The time to reach this temperature may be deduced from Equation (12):

$$t_{gel\ total} = \frac{1}{\beta}ln\frac{T_a-T_{do}}{T_a-T_{dg}} \tag{15}$$

Introducing this value in Equation (7) or (7a) will provide the required cooling column height to get the gelation.

RESULTS & DISCUSSION

Impact of the column temperature and collecting distance on the bead formation

Thirty-six experiments are run using a 5 % κ-carrageenan solution at 70 °C by varying the column temperature and the collecting distance (Table 2). As the drop detaches from the nozzle, it has a tear-shape.[9] The gelation must not be too fast allowing the drop to take a spherical shape. Preliminary tests showed that the nozzle has to place at least 20 cm on top of the cooling column. However, collecting distance, y, must be sufficient to jellify the drop prior the impact with the collecting surface. At room temperature, no spherical bead was collected. While temperature inside the column is equal to 3 °C, spherical beads obtained for

collecting distance superior to 1.25 m. Dropping the temperature to -10°C allows to get spherical beads for collecting distance down to 1 m. Qualitatively, this confirms that the lower the temperature inside the column is, the lower is the dropping height to get spherical capsules, and to assume a correct gelation of the droplets.

For a more quantitative analysis, sphericity factor was plotted versus the collecting distance (Figure 2a). The minimum distance to reach sphericity factor higher than 0,95 maybe deduced and is plotted in Figure 2b (dashed line).

The collecting distances versus temperature were plotted on Figure 3a. The dash line divides the graph in two regions; below, one obtains non-spherical beads (triangle) and above, spherical *beads* (cross). The continuous line corresponds the estimation of the gelation distance using the model described above. In Figure 4, three photographs of carrageenan beads correspond to various measures of sphericity.

Over the studied range of air cooling temperature, the transition from oval beads to spherical beads corresponds quite well with the model prediction of the gelation height (Figure 3b). The model then correctly simulates the gelation of the κ-carrageenan droplet gelation in a cooled air column.

Validation Model and Its Hypotheses

Figure 5 shows κ-carrageenan analysis is done by micro differential scanning calorimetry (DSC). A small peak is observed around 46 °C corresponding to an enthalpy of -0.9125 J/g. Pivette[13] has built similar model for the solidification of melt fatty acid droplets, taking into account the solidification enthalpy but more complex. However, intrinsically the κ-carrageenan gelation is a less energetic process and the solution contains only 5 % of κ-carrageenan. The model could then be simplified, assuming a mean and constant specific heat capacity, c_p, over the working temperature range. From Figure 5, it was evaluated to 4,2 kJ/kg.K, which is very similar to water.

The size of the droplets and the beads are very similar (around 2 mm). No contraction was evident during gelation while a volume reduction was observed during solidification of melt droplets.[13]

Figure 6 presents an example of the simulated evolution of the bead temperature and velocity as well as its drop distance versus time in the cooling column. As expected, the droplet velocity increase to reach a maximum corresponding to the terminal velocity, u_t. However, as the gelation occurs in less than 2 s, the column could be limited to around 1 m and the velocity would reach only 4 m/s, i.e. 57 % of the terminal velocity.

The Reynolds number computed is such condition is equal to 5130, confirming that we are in the turbulent regime. The Biot number is 0.0012, showing that the heat transfer is limited at the air-droplet interface and not inside the droplet. One may then expect that gelation takes place homogeneously in the beads, not moving from the surface to the center of the droplet.

It could then be concluded that the hypotheses used to develop the model are valid.

Simulation of the Gelation in Gravity Drop Wise Conditions

At low flow rate, the extruded κ-carrageenan solution will exit the nozzle dropwise. Under simple gravity the size of the droplet is generally around 2.5 to 3 mm. However, applying for example an electrostatic potential to the nozzle lower droplet size may be obtained.[14] The drop starts to fall with a zero velocity.

Figure 7 presents evolution of the droplet velocity, temperature and falling height in function of the time for different droplet sizes. The time of gelation is quasi-independent of the size while the vertical distance for correct gelation increases with droplet size. In fact, the smaller drops fall at lower speed than larger ones. The heat exchange at the surface is reduced compensating the fact that less energy is needed to jellify small droplets.

The impact of the column temperature is presented in Figure 8. Both time and height of gelation increase quickly as the column temperature increases, showing the real interest to maintain a low temperature in the column.

For biocatalyst encapsulation, high temperature must be avoided. Tests have been realized with κ-carrageenan presenting lower melting temperature. Using similar conditions, no spherical beads were obtained. Reducing the κ-carrageenan gelation temperature and the

initial temperature of the κ-carrageenan solution impose to either decrease the column temperature or increase its height.

K-Carrageenan Bead Production in Jet Breakage Regime

For larger scale production, especially producing small beads, the liquid flow rate is increased to reach the formation of a liquid jet. The jet is then broken in droplet either by applying a specific vibration on the jet[5] or by passing the jet through a rotating cutting wheel.[15] As such, the productivity by nozzle is increased by a factor of 10 or more, reaching liters per hour.

The initial drop velocity is not zero and mainly equal to the jet velocity. The evolution of the droplet velocity over time is presented in Figure 9. Depending on the initial velocity, the drop will be accelerated or decelerated to finally reach the terminal velocity.

If the initial velocity is significantly lower than the terminal velocity, drop acceleration may lead to collision between droplets. If the initial velocity is significantly higher than the terminal velocity, the deceleration may also lead to droplet collision. The optimum condition is then to select a jet velocity similar to the terminal velocity.

Impact of K-Carrageenan Selection

Most of the actives that could be encapsulated in κ-carrageenan beads are thermo-sensible. One may limit the effect of temperature by realizing a quick mixing in front of the extrusion. Selecting a lower gelation temperature may also limit the detrimental effects. However, some experiences show that using lower temperature gelation κ-carrageenan was not successful.

The simulation presented in Figure 10 is realized assuming that the κ-carrageenan solution is extruded at a temperature 30 °C above the complete gelation temperature. Both the time and the falling height gelation to reach it increase when the κ-carrageenan gelation temperature decreases. To get correct gelation, height of the cooling column must be increased and/or the temperature in the column must be decreased.

Figure 10

CONCLUSION

The κ-carrageenan beads are usually made by ionic gelation. κ-carrageenan solution was extruded dropwise into a potassium chloride solution. The use of a gelling bath needs steps of filtration and drying, which are costly for industries. To eliminate these steps, it is necessary to extrude carrageenan solution in the cold air as its gelation is carried out thermally.

Our study was to develop production of κ-carrageenan by thermal gelation eliminating bath reception and the step of rinsing. A mathematical model was established on the heat transfer between carrageenan beads and air-cooled to predict the dropping height depending on the morphology of the particles. This characterization is important criterion that was defined by Chan et al.[9] Theory and experiments of this work are very encouraging.

Simulations and predictions were performed showing the evolution of droplet velocity, the falling height in function of various parameters (gelation temperature, droplet size, initial velocity, etc.). To work in favourable industrial conditions, it is necessary to have the following:

- A jet breakage regime,
- The falling height to be adapted in function of the gelation temperatuap,
- An air temperature around 0 °C,
- An initial velocity close to th e terminal velocity.

NOMENCLATURE

Bi	biot number		
C_D	drag coefficient	0.44	dimensionless
C_p	specific heat capacity of carrageenan	4200	J/kgK
d	diameter of droplet	2.5 10-3	m
d_{max}	maximum diameter of bead		m
d_{min}	minimum diameter of bead		m
g	acceleration of gravity	9.81	m/s^2
h	heat transfer coefficient	3447.7	W/m^2K
λ_a	thermal conductivity of air(T 273K)	0.024	W/mK
λ_d	thermal conductivity of droplet		W/mK
m	droplet mass		kg
R_e	Reynolds number		
S	surface of droplet		m^2
T_a	air temperature	273	K
T_d	droplet temperature		K
T_{d0}	carrageenan solution temperature	353.15	K

T_{dg} or T_{gel}	gelation temperature		K

total

U_0	initial velocity		m/s
U_t	bead thermal velocity	7.76	m/s
β		1.92	SI
ρ_a	volumic mass of air	1.272	kg/m3
ρ_d	volumic mass of bead	1030	kg/m3
μ_a	viscosity of air	1.8 10-5	Pa.s
τ		6.13	SI

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Table 1. Comparison of dimensionless shape indicators and corresponding particle shapes

Dimensionless shape parameters				
	Spherical	Oblate	Egg-shape	Tear/pear shape
Sphericity factor (SF)	>0,95	0,95 – 0,92	0,92-0,78	<0,78

Table 2. Production of carrageenan beads at different temperatures and dropping heights

Height (m)	T -10°C			T 3°C			T 25°C		
	D (mm)	SF	Aspect	D (mm)	SF	Aspect	D (mm)	SF	Aspect
0.25	1.95	0.8	🔵	2.43	0.80	🔵	NR	NR	
0.50	1.72	0.85	🔵	2.40	0.81	🔵	NR	NR	
0.75	1.94	0.88	🔵	1.83	0.90	🔵	1.97	0.80	🔵
1.00	1.72	0.94	🔴	1.79	0.93	🔵	1.97	0.89	🔵
1.25	2.08	0.95	🔴	2.13	0.94	🔴	2.02	0.92	🔵
1.50	2.07	0.98	🔴	1.85	0.97	🔴	1.88	0.93	🔵

D : diameter, SF : Sphericity factor and NR : no response
Red color corresponds to the spherical beads and blue color corresponds to egg-shape beads

Figures:

Figure 1.

Experimental set up (A: air pressure, B: manometer, C: bottle under pressure, D: nozzle (200 µm), E:coling column, F: reception bowl, I: dry ice, H: inox cylinder, G: ventilator)

Figure 2.

A. Effect of collectig distance on sphericity factor of carrageenan beads; B. Effect of air temperature on minimum distance to obtain spherical beads (SF= 0.95)

Figure 3.

A. Effect of air temperature on gelation time and height (dash line: experimental values for spehrical beads, continuous line: theoretical gelation distance for spherical beads); B. validation of model

Figure 4.

Images of carrageenan beads at 20°C. A. deformed beads, B. oval beads, C. Spherical beads

Figure 5.

Carrageenan analysis by DSC (specific heat capacity).

Figure 6.

Evolution of the droplet velocity (A), falling height (B) and bead temperature (C) versus time

Figure 7.

Evolution of the droplet velocity (A), falling height (B) and bead temperature (C) versus time for different droplet size

Figure 8.

Time of gelation (lozenge) and the falling height (square) versus air temperature

Figure 9.

Evolution of droplet velocity (Ut) according to initial volocity (U0)

Figure 10.

Time of gelation (lozenge) and the falling height (square) versus gelation temperature.
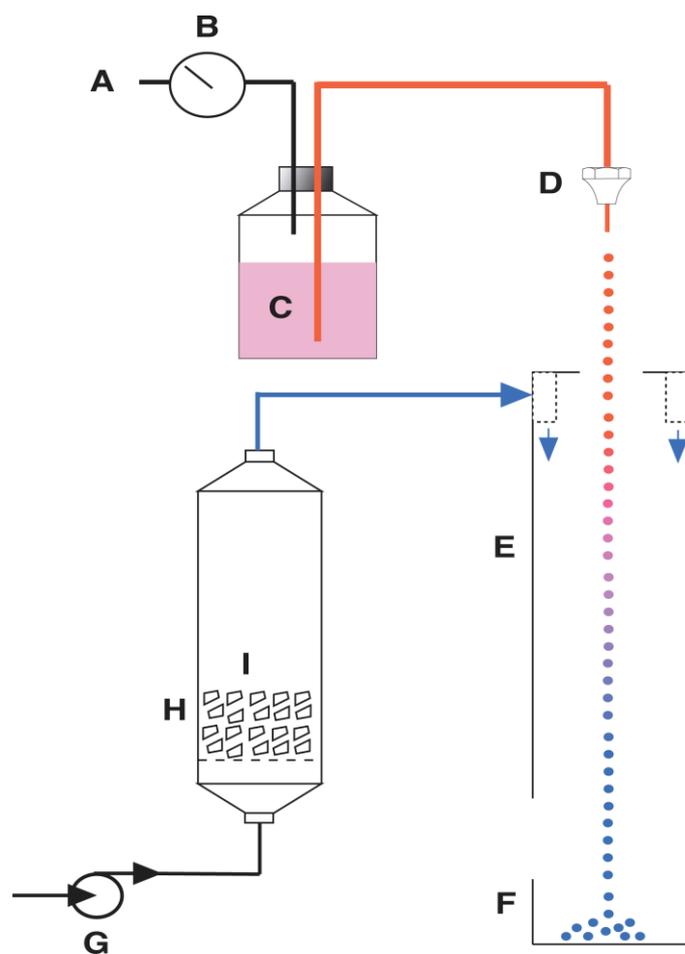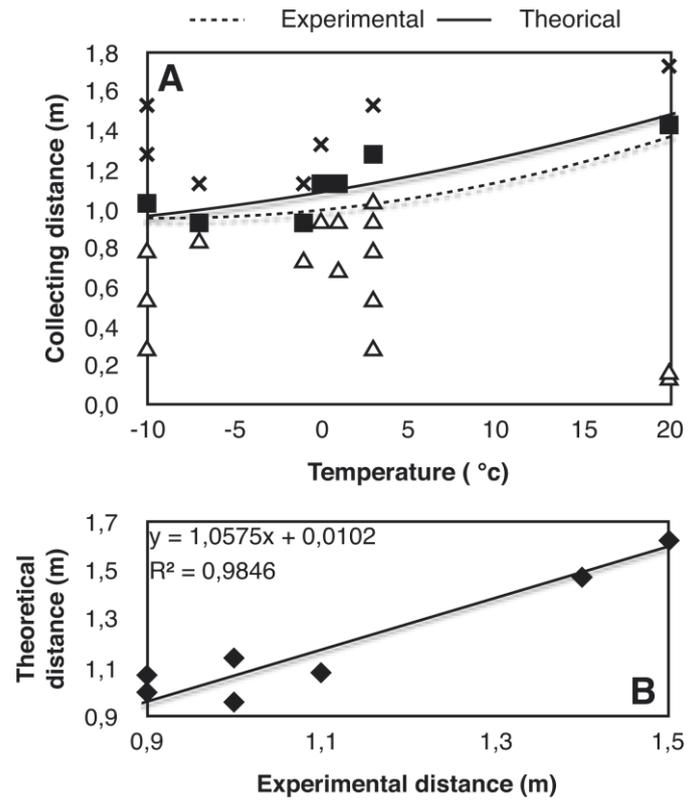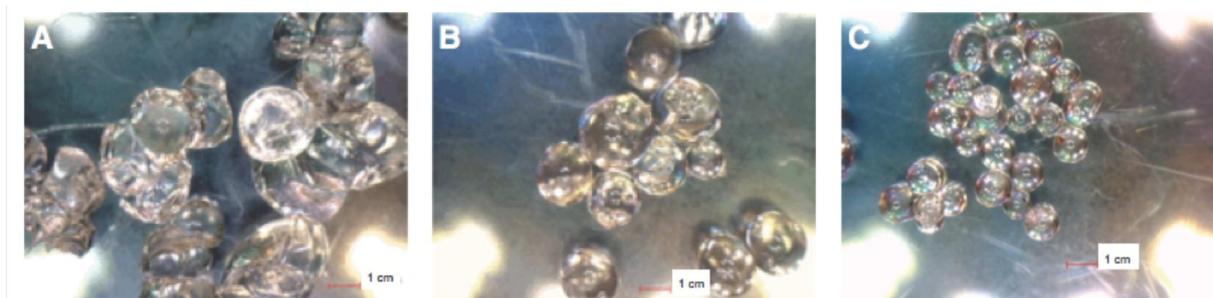
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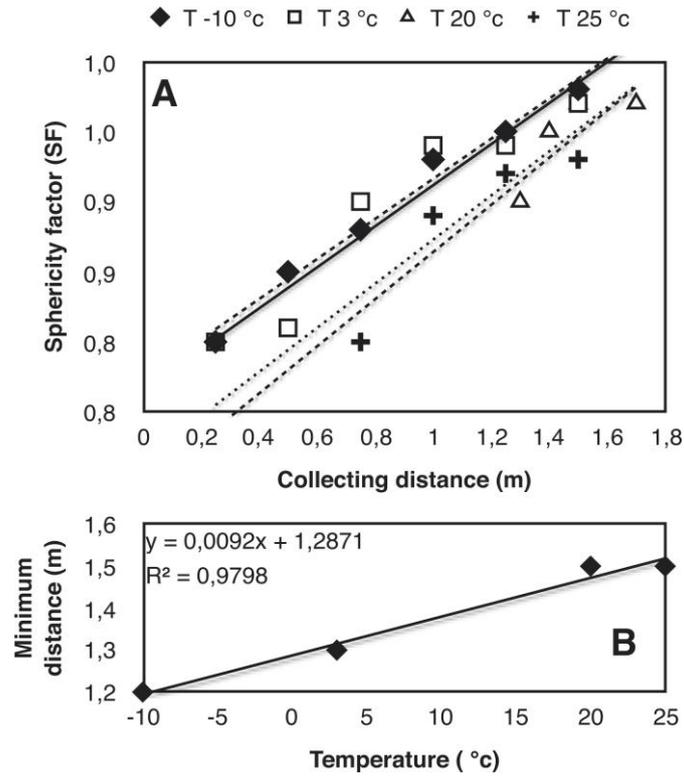
Figure 2

Figure 3

Figure 4
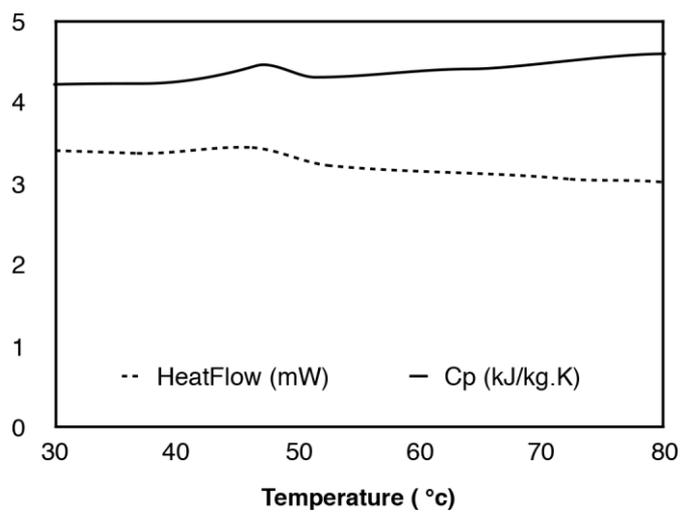
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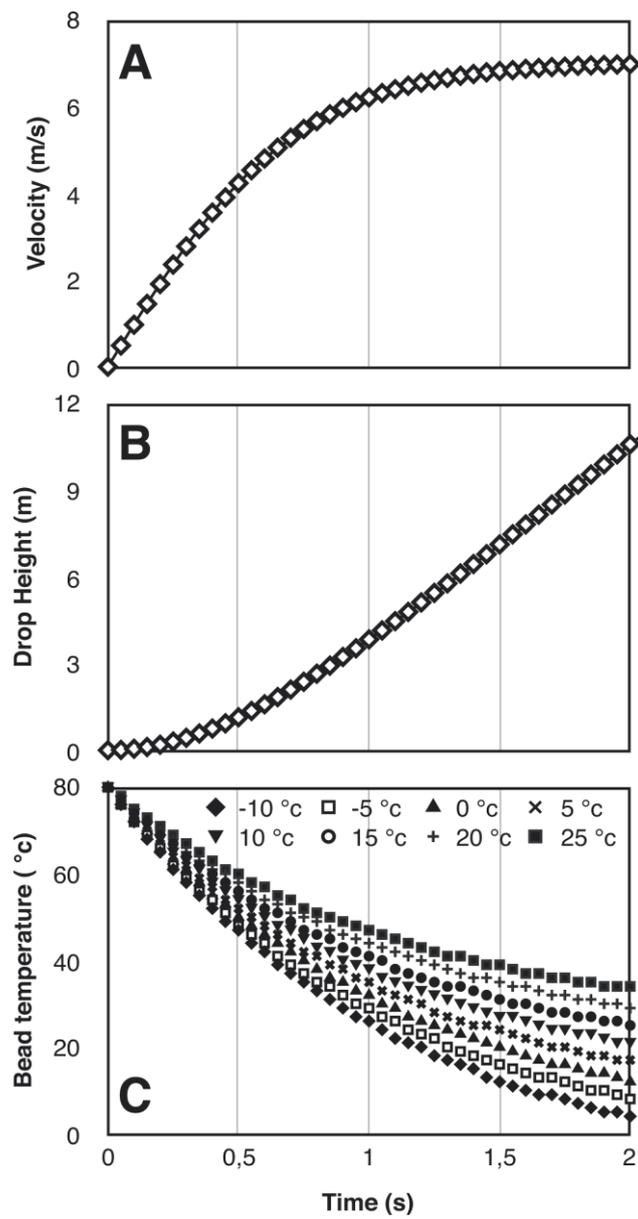
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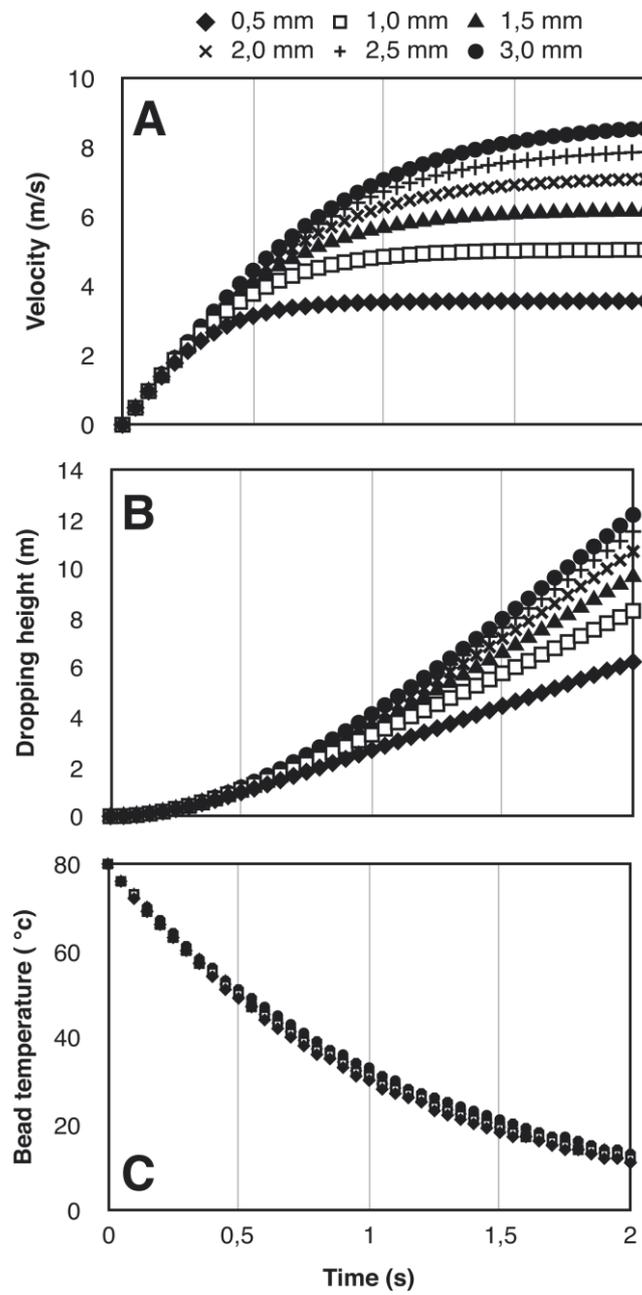
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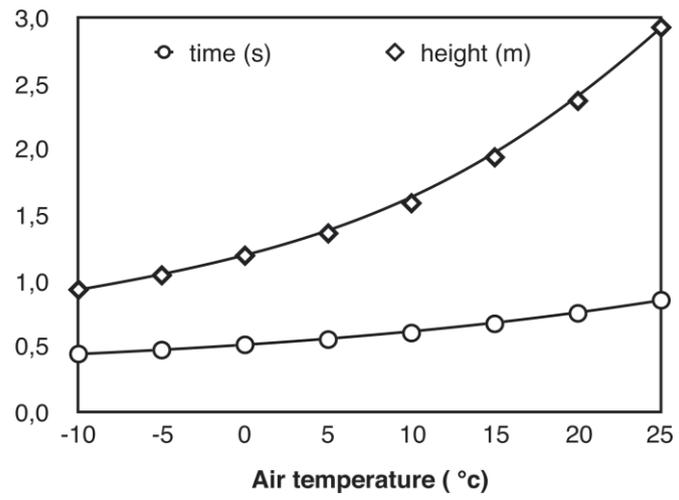
Figure 8

Figure 9

Figure 10