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Optimization of inulin production process parameters using response surface methodology

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Abstract

Background: Chicory is one of the major source of inulin. In our study, Box–Behnken model/response surface analysis (RSM) was used for the optimization of spray drying process variables to get the maximum inulin yield from chicory (*Cichorium intybus* L.). For this investigation, the investigational plan utilized three process variables drying temperature (115–125 °C), creep speed (20–24 rpm), and pressure (0.02–0.04 MPa).

Result: The optimal variables established by applying the Box–Behnken model were as follows: drying temperature 119.20 °C, creep speed 21.64 rpm, and pressure 0.03 MPa. The obtained powdered inulin by spray drying was investigated for the yield value, identification, size, and surface morphology of the particle. The inulin obtained from the spray drying process consists of a fine molecule-sized white powder. Instead, the drying methods shows a significant effect on the morphology and internal configuration of the powdered inulin, as the inulin obtained from spray drying was of a widespread and uniform size and shape, with a rough surface on increase in temperature and smoother surface while increasing the creep speed. The findings indicate that the spray drying with optimum parameters resulted in maximum product yield.

Conclusion: The outcomes of the study concluded that the product yield through spray drying technique under optimized condition is optimal as compared to other drying technique. Hence, this technique may be applied at commercial scale for the production of inulin.

Keywords: Inulin, Spray drying process, Optimization, Box–Behnken design/response surface methodology

Background

The chicory, tuberous roots that store inulin, may be a native plant from Europe, Asia, Africa, and South America, but it often cultivates altogether over the planet [1]. Chicory (*Cichorium intybus* L.) may be a biennial plant which comes under the Asteraceae family with many applications within the food industry. The roots of chicory are the principal source of inulin [2]. Inulin could be a fructan which generally composed of 1, 2-[3-1inked D-fructofuranose entities bound by an (od-132) type bond to a terminal glucose moiety. By taxation, inulin primarily made up of linear fructose segments knotted via a β-(2-6) glycosidic bond [3]. Inside the human, inulin may act in parallel with dietary fibers, adding to the general improvement in the gastrointestinal state. Due to these belongings, the food and pharmaceutical sector utilizes inulin for the manufacture of functional foods, nutritional products, in drug delivery system and medications; it may function as a prebiotic which stimulates the growth of the number of useful microorganisms, specifically bifidobacteria which can improve host health and diminishes mucosal inflammation in the colon [4, 5].

The number of the monomeric unit and relative molar mass of inulin mainly depends on the plant used for the isolation. The degree of inulin polymerization (DP) varies between 2 and 70, with inulin having the degree of polymerization less than 10 recognized as oligofructose or

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shorter-chain inulin, and inulin having a degree of polymerization greater than 23 known as polyfructose or longer-chain inulin [6, 7]. Furthermore, inulin as an indigestible carbohydrate can also be utilized as a fat alternative for foodstuffs.

Inulin is generally traded as a powder for easy to pick-up, shipping, and storage [8]. The production technique used to get dried powder inulin is enormously important in defining its grade. At present, freeze drying and spray drying are commonly used techniques for powdered inulin production [9]. The technique lyophilization/freeze drying embraces evaporation and elution and normally needed 2 or 2.5 days for acquiring a spongy and loose product [10]. The freeze drying technique is in demand for packaging and depot; subsequently, the dry powder grips moistness and may be simply oxidated when unveiling to the environment [11, 12]. Additionally, it is a technique with the high cost and thin productivity. Consequently, spray drying was generally used drying technique within the food sector because it is inexpensive and has great pliability [13].

The spray drying technique utilizes a vaporizer to diffuse the fluid into small globules and quickly vaporizes the solution in the dried environment to get a product in powdered form. Ritual yield is frequently accomplished by differing factors like temperature, viscidness, input ratio, or micronizing pressure [14]. The resulting dry powder features particles of uniform size with great sphericity. For example, during a preceding study, Jerusalem artichoke pectin properties were assessed, obtained by employing different methods (vacuum drying, freezing and spray drying), and consequently, the researchers found spray drying is the best among the above used technique [15].

The main objective of our work was, therefore, to optimize the different parameters of the spray drying technique through the execution of response surface methodology/Box–Behnken model to get the maximum production of inulin.

Methods

Materials

Chicory root was obtained from a market of Gwalior, Madhya Pradesh, India. The Botanical Survey of India, Central Regional Centre, Allahabad, under the aegis of the Ministry of Environment, Forest and Climate Change, identified and authenticated the plant specimen. The authentication accession no. is 102272. The supplementary materials utilized in the current research work were of analytical rank and were secured from Hi-Media Laboratories Ltd. (Mumbai, India).

Preparation of chicory extract

The chicory roots were collected; after that, they were clean with tap water and dehydrated at 40 °C within the vacuum oven. The grounded taproots were extracted with hot water (~ 90 °C) under the pressure of 1.5 MPa for 2–3 min. Repeating the extraction twice, the filtrates were mixed and the pH was adjusted to 8.0 by adding $Ca(OH)_2$. The blend was left at ambient temperature for 1 h. Afterwards, the sludge was filtrated. The residues were neutralized to pH 7 at temperature 60–65 °C by oxalic acid. Activated carbon was added then the filtration process was done again. The residue was precipitated by addition of acetone and left to settle down at 2–5 °C. The resulting amorphous substance was filtered, washed with ethanol twice, acetone once, and finally dried at 40 °C [2] (Fig. 1).

Development of inulin powder

Inulin as the dry powder was produced by employing the spray drying technique. Box–Behnken design was employed to optimize the process variables. The level of each process variables/independent factor is low, mid, and high, and their equivalent actual values are presented in Table 1. Three independent factors utilized in the Box–Behnken design were drying temperature (115–125 °C), speed (20–24 rpm), and the applied pressure (0.02–0.04 MPa). Table 2 presents the investigational runs executed according to the Design-Expert Box–Behnken model.

The data from the investigation were examined statistically via version 11.1.0.1 of Design-Expert (Stat Ease Inc., USA). Product yield remained countered to a model of quadrate regression for response surface examination.

As presented in the equation:

$$\begin{aligned}\mathrm{Yield} = \alpha_0 + \alpha_1 A + \alpha_2 B + \alpha_3 C + \alpha_4 AB + \alpha_5 AC \\ + \alpha_6 BC + \alpha_7 A^2 + \alpha_8 B^2 + \alpha_9 C^2\end{aligned}$$

Here, A, B, and C represent temperature, creep speed, and pressure. The α displays values of the analogous regression. The investigation was arbitrary in taking advantage of the influence of undiscovered variation on perceived results due to extrinsic facets. Half liters of clear inulin extract was utilized with a spray-dryer (SPD-D-111) for drying experimentation; the inulin extract was propelled via a spout to the counter-current dehydrating compartment utilizing a roller pump. The extract of the product was heat-up to a defined degree before every feeding (intake temperature is greater than 60 °C, and the outlet temperature is higher than 50 °C). Spray drying process variables for maximum product yield were established with the help of response surface analysis assay.

Characterization and analysis

Determination of inulin yield

The product yield was determined by using the following formula:

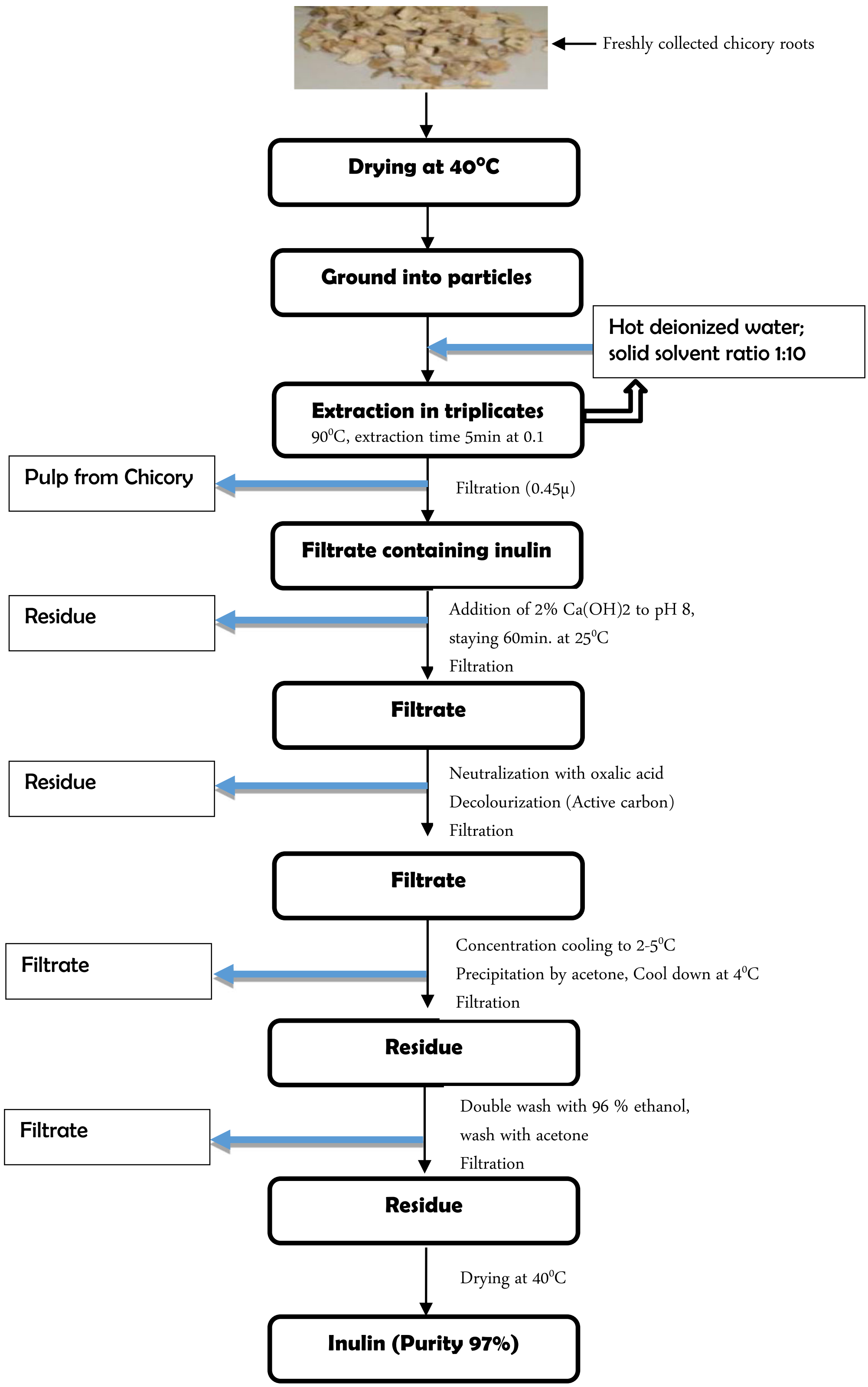

Fig. 1 Schematic inulin extraction process from chicory

Table 1 Independent variables and levels of experimental design

S. no.	Independent variables	Code level		
		Low (− 1)	Mid (0)	High (+ 1)
1.	Heating temperature °C (A)	115	120	125
2.	Creep speed rpm (B)	20	22	24
3.	Pressure MPa (C)	0.02	0.03	0.04

$$Y = \frac{\mathrm{mp}}{\mathrm{M}} \times 100\%$$

where mp (kg) is the powdered inulin mass gained from 6-L extract and *M* is the weight of chicory root utilized in the isolation to get 6-L extract.

Identification of isolated inulin

Fourier transform-infrared spectrometer (FT-IR) was utilized to elucidate and characterize the structure of extracted material from chicory. Inulin dry powder (~ 3 mg) was added with ~ 200 mg FTIR ranks anhydrous KBr and milled into a very fine powder by utilizing pestle and mortar. The disc was packed with the blend. Infrared spectrogram of a disc was taken over a wavenumber scope of 400–4000 cm^{-1} at a resolution of 4 cm^{-1} with a test speed of 1 cm/s using the FT-IR (FT-IR 8400S, Shimadzu, Japan).

Determination of melting point and degree of polymerization

The fusion point of inulin was determined by the capillary method. A very small amount of inulin powder was inserted in a capillary tube. The capillary was inserted in the apparatus and the temperature at which inulin commenced to melt was recorded.

With the help of the following equation, the degree of polymerization of isolated chicory inulin was determined:

Degree of polymerization = $C_{\mathrm{fructose}}/C_{\mathrm{glucose}} + 1$

where C concentration, %

Study of particle size distribution and surface morphology of isolated powdered inulin

The product's size and surface morphology was calculated using a scanning electron microscope (SEM) article JSM 5600 JEOL, Japan. The assessment of the particles size was done by assessing them on an electronic paquimeter utilizing the picture system [16]. The equivalent mean diameter of every particle was believed as the mean within two perpendicular measurements. For all investigation, the subsequent assessment was taken: mean equivalent, maximal, and minimal diameters.

Results

The extraction yield of this investigation by applying hot water extraction (conventional method) was determined as ~ 11.96%.

Table 2 Test design and results of response surface analysis (each test was performed in triplicate)

Run	Coded variable			Actual variable			Response
	X	*Y*	*Z*	*A*	*B*	*C*	Yield %
1	0	+1	−1	120	24	0.02	10.97 ± 0.06
2	− 1	0	−1	115	22	0.02	11.35 ± 0.09
3	+ 1	−1	0	125	20	0.03	11.05 ± 0.11
4	− 1	0	+1	115	22	0.04	09.77 ± 0.04
5	0	+1	+1	120	24	0.04	09.35 ± 0.03
6	0	0	0	120	22	0.03	12.03 ± 0.14
7	0	0	0	120	22	0.03	12.16 ± 0.15
8	0	0	0	120	22	0.03	11.98 ± 0.14
9	+ 1	0	−1	125	22	0.02	10.50 ± 0.07
10	0	−1	+1	120	20	0.04	09.95 ± 0.02
11	+ 1	0	+1	125	22	0.04	09.55 ± 0.05
12	+ 1	+1	0	125	24	0.03	10.33 ± 0.07
13	− 1	−1	0	115	20	0.03	10.20 ± 0.06
14	− 1	+1	0	115	24	0.03	10.66 ± 0.07
15	0	−1	−1	120	20	0.02	10.48 ± 0.08
16	0	0	0	120	22	0.03	12.16 ± 0.18
17	0	0	0	120	22	0.03	12.98 ± 0.22

X&A,Y&B, Z&C indicate the temperature (°C) creep speed and applied pressure respectively

Optimization of spray drying technique with the help of response surface analysis

Table 2 presents the experimental plan for the spray drying process and consequently the results obtained by using Box–Behnken design to optimize inulin isolation. Likewise, the analysis of variance (ANOVA) investigation is shown in Table 3.

12.97 model F value indicates the model is significant. p values under 0.050 specify system levels are significant. Our study shows C, A^2, B^2, and C^2 are significant levels. p values higher than 0.100 specify the levels are insignificant. 0.67 F value of lack of fit implies that in relation to the pure error, the lack of fit is insignificant and the insignificant lack of fit is best.

Table 2 investigation findings are fitted with regression, and the final equation for securing the coded variables through statistics scanning is presented in Eq. (1). The true variable regression is set out in Eq. (2).

$$Y = 12.26 - 0.0687 \times X - 0.0462 \times Y - 0.5850 \times Z - 0.2950 \times XY + 0.1575 \times XZ - 0.2725 \times YZ - 0.7985 \times X^2 - 0.9035 \times Y^2 - 1.17Z^2 \quad (1)$$

$$Y = -639.15525 + 8.20635 \times A + 13.86412 \times B + 565.85000 \times C - 0.029500 \times AB + 3.15000 \times AC - 13.62500BC - 0.031940 \times A2 - 0.225875 \times B2 - 11710.00000 \times C2 \quad (2)$$

The comprehensive p value of the model is 0.0014 which is less than 0.01, indicating regression equation is significant, and p value for lack of fit is 0.6132 which is greater than 0.05, representing insignificant lack of fit. It represents equation fitting is comparatively acceptable. Regression study (Table 3) revealed A^2, B^2, and C^2 show a noteworthy impact on product yield. Likewise, the influence of the variables on yield showing significance as per the order: drying temperature < creep speed < applied pressure.

Figure 2 presents the clear vision of the contour plot, the outline of the 3D response surface, and distinctly presenting the influence of the collaboration of the independent variables on the yield value. Figure 2a–c presents the consequences of independent variables on the response. When the heating temperature goes 110 °C to 120 °C, the value of yield initially increases and afterwards decreases. This fact is often accredited an improved drying technique when the heating temperature was elevated, though it may cause decay of inulin moiety. Also, the rise in creep speed and applied pressure shows an equivalent drift concerning variation in product yield.

Identification of isolated inulin

The isolated inulin sample FT-IR was interpreted and checked with the standard IR spectrum shown in Fig. 3. Full information on the FT-IR spectra of chicory-isolated inulin was shown in Table 4. The peaks at 1131 cm^{-1}, 1087 cm^{-1}, and 1031 cm^{-1} are representative for ($C-C$, $C-O$, $C-O-C$) stretching bands in the fructo-furanose ring. 2-keto in fructo-furanose contains band at 874 cm^{-1} and 821 cm^{-1}, which are signals for existence of β-(2→1) glycosidic bonds. Our investigation is very much similar to the reported bands for inulin by Grube and Olennikov [17, 18].

Table 3 ANOVA for quadratic model. Response 1: yield

Basis	Sum of squares	Degree of freedom	Mean square	*F* value	*p* value	Significance
Quadratic model	16.78	9	1.86	12.97	0.0014	Significant
A-Temperature	0.0378	1	0.0378	0.2630	0.6239	
B-Creep speed	0.0171	1	0.0171	0.1190	0.7402	
C-Pressure	2.74	1	2.74	19.04	0.0033	
AB	0.3481	1	0.3481	2.42	0.1637	
AC	0.0992	1	0.0992	0.6901	0.4336	
BC	0.2970	1	0.2970	2.07	0.1938	
A^2	2.68	1	2.68	18.67	0.0035	
B^2	3.44	1	3.44	23.90	0.0018	
C^2	5.77	1	5.77	40.15	0.0004	
Residual	1.01	7	0.1438			
Lack of fit	0.3369	3	0.1123	0.6707	0.6132	Not significant
Pure error	0.6697	4	0.1674			
Cor total	17.79	16				

A, *B*, and *C* are the temperature, creep speed, and pressure respectively

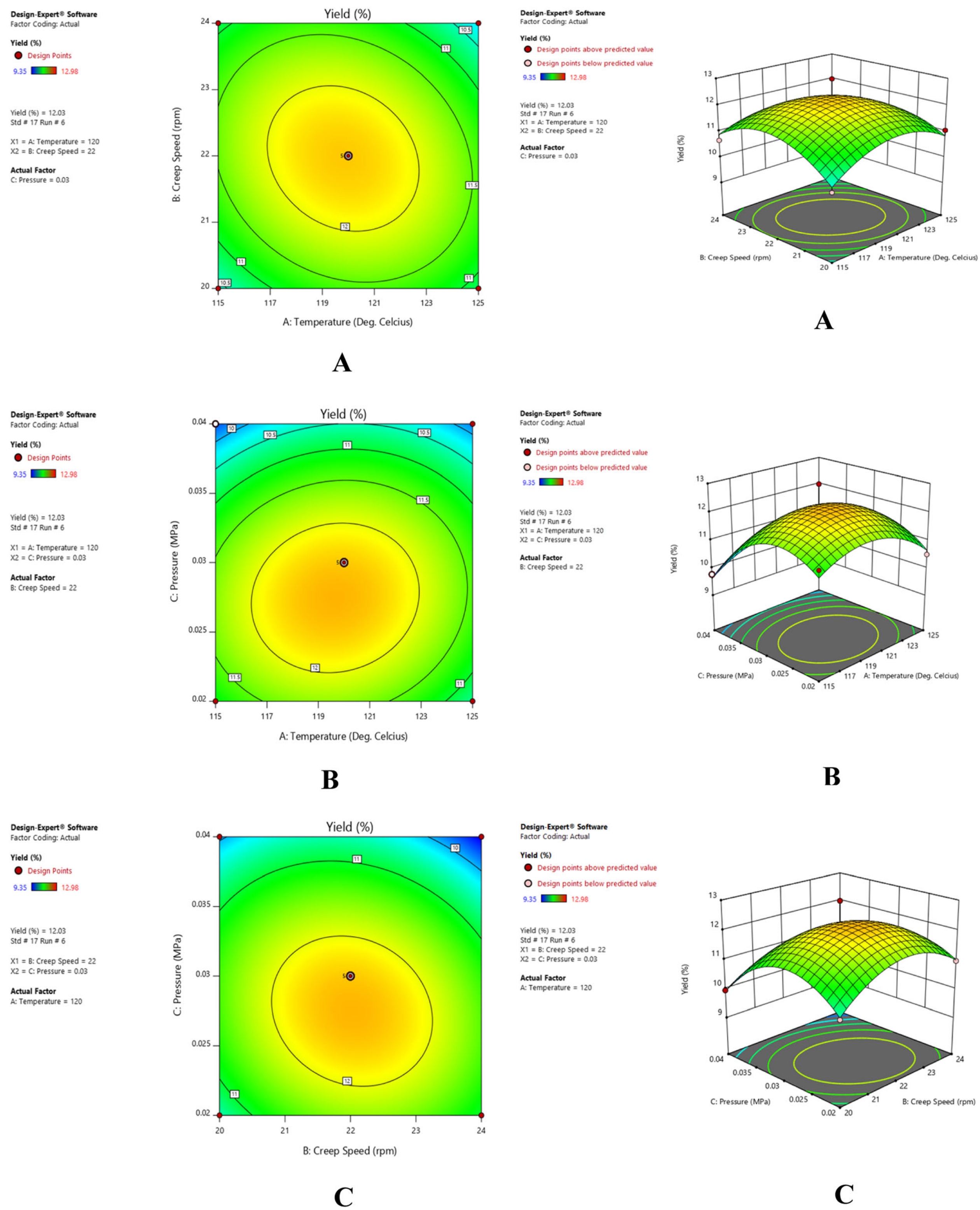

Fig. 2 Result of interactions of independent variables on inulin yield

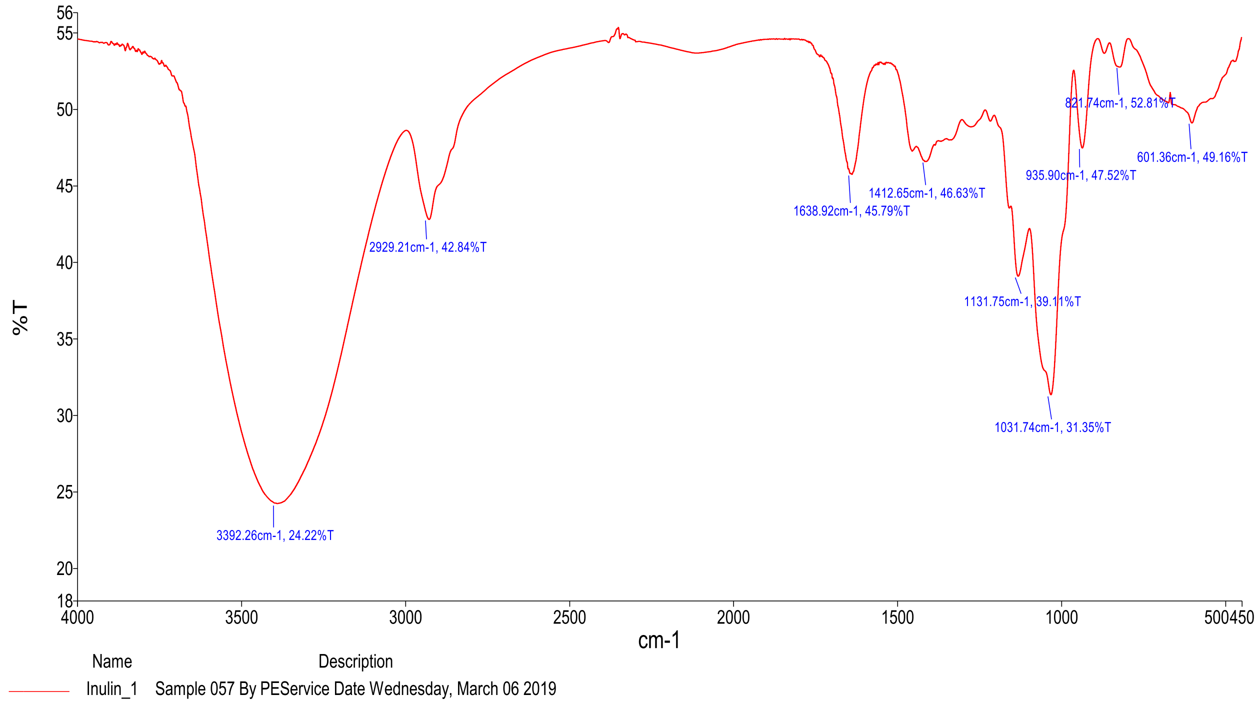

Fig. 3 FT–IR spectra of inulin isolated from chicory root

Determination of melting point and degree of polymerization

The fusion point of isolated powdered inulin was found in the range of 163–164 °C which was very close to the standard reported by Nadezhda and Panteley [2]. The determined degree of polymerization of extracted inulin was 26. This value is nearby to the degree of polymerization 23 stated by Praznik and Beck for inulin obtained from chicory roots [19].

Table 4 Interpretation of FT- IR spectra of isolated inulin

Wave number cm^{-1}	IR bands cm^{-1}	Interpretation
3200–3400	3361	ν_{O-H} (OH); intermolecular H-bonds
2933–2981	2931	$\nu_{C-H}{}^{as}$(CH2)
2850–2904	2880	$\nu_{C-H}{}^{s}$(CH2)
1664–1634	1635	Absorption of water
1416–1430	1430	$\delta_{C-H}{}^{s}(CH_2)$
1335 –1336	1336	β_{O-H}(OH)
1225–1235	1220	β_{O-H} (OH)
1125–1162	1135	$\nu_{C-O-C}{}^{as}$ (C–O–C)
1015–1060	1031	ν_{C-O} (C–O) C–O
985–996	987	ν_{C-O} (C–O)
930	935	α-D-Glcp residue in chain
892–895 874	873 873	Anomeric bendings δ(C1–H), ring vibration (2-ketofuranose)
818	820	2-ketose

Study of particle size distribution and surface morphology of isolated powdered inulin

SEM pictures showing powder product samples obtained from trials clearly illustrate that the high pace of the siphon shows irregular surfaces of the inulin particles. In the same way, the SEM image displays the high temperature result in particles with the smoother surface as depicted in Fig. 4a (run 1) and b (run 3). The mean diameter of all drying situations, as well as extents of maxima and minima of the diameters, is presented in Table 5.

Discussion

The method for isolation of inulin from chicory roots was simple to execute. The isolated inulin was characterized as an odorless and tasteless cremish-white powder with melting point 163–164 °C which was very close to the reported melting point by Nadezheda and Panteley [2]. The determined degree of polymerization of isolated product was 26. This value is nearby to the degree of polymerization 23 stated by Praznik and Beck for inulin obtained from chicory roots [19] and 25 for high-performance inulin extracted from chicory witloof [20]. The FT-IR spectrum of isolated inulin show very much similar to the reported bands for inulin by Grube and Olennikov [17, 18]. The result findings from IR spectrum demonstrate that the isolated material from roots of chicory is inulin-type fructan.

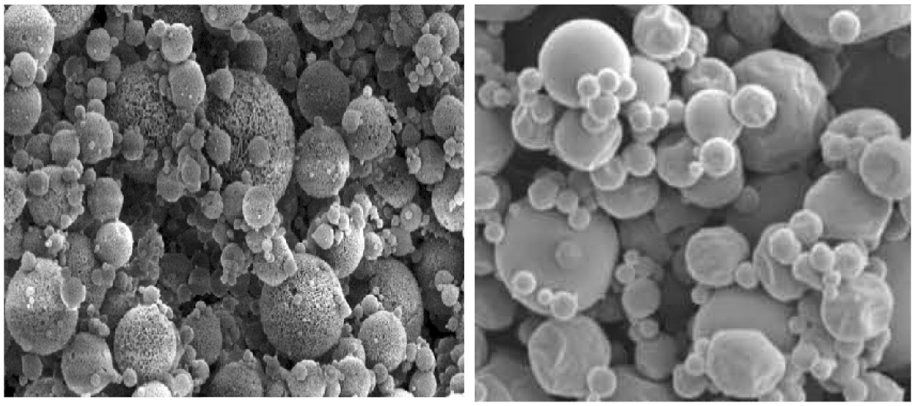

Fig. 4 SEM images of inulin powder. **a** Run 1 presenting the rough surface and **b** run 3 presenting the smooth surface of the particles

As per the investigational statistics and design study, the maximum product yield might be accomplished under the ideal conditions: drying temperature of 119.20 °C, the creeping speed of 21.64 rpm, and the applied pressure of 0.03 MPa, resulting in a product yield of 11.96%. To check the accessibility and accuracy of the regression study acquired within the Box–Behnken model for the response surface analysis, the above said ideal process variables were utilized for the confirmation trial. For the process accessibility, the drying temperature was fixed at 120 °C, the creeping speed was 22 rpm, the applied pressure was 0.03 MPa, and 3 analyses were executed in similar. In these confirmation trials, the product yield was 12.02 ± 0.52%, which is nearly same to the projected yield, and also specifies that the equation aligns well with the actual condition.

For all the levels of independent variables used in the trial, inulin particulate revealed sphericity, with a high deviation in diameter and with a petite trend to hitch together (agglomeration).

Table 5 Inulin powder average equivalent diameter and maximal and minimal diameters obtained at different Runs

Run	Diameter $_{Avg}$ (nm)	Diameter $_{max}$ (nm)	Diameter $_{min}$ (nm)
1	345.8 ± 32	766.5 ± 42	42.5 ± 0.5
2	452.6 ± 22	725.6 ± 54	38.3 ± 0.5
3	458.5 ± 30	765.2 ± 60	35.6 ± 0.8
4	366.2 ± 26	802.8 ± 75	40.2 ± 1.0
5	389.4 ± 18	785.6 ± 66	33.5 ± 0.7
6	335.8 ± 25	792.5 ± 58	48.6 ± 2.0
7	310.6 ± 34	822.4 ± 64	30.2 ± 0.6
8	454.3 ± 20	775.3 ± 52	45.5 ± 0.5
9	502.7 ± 44	820.6 ± 58	48.8 ± 0.5
10	485.5 ± 38	832.5 ± 55	36.4 ± 1.0
11	382.2 ± 36	750.4 ± 63	46.3 ± 1.0
12	335.8 ± 28	796.5 ± 68	55.3 ± 0.4
13	436.5 ± 33	836.7 ± 63	50.1 ± 2.0
14	442.6 ± 27	828.6 ± 53	52.5 ± 3.0
15	465.4 ± 35	840.8 ± 68	55.8 ± 2.0
16	422.6 ± 42	862.4 ± 80	48.6 ± 0.8
17	368.3 ± 34	874.8 ± 75	56.5 ± 1.0

Conclusion

Inulin dry powder was produced via the spray drying process. The outcomes indicated that the product yield initially rises when independent variables were increased, and then afterwards declined. By employing Box–Behnken design for response surface analysis, the maximum product yield was 11.96%, found within the optimum level of drying temperature 119.20 °C, the creeping speed of 21.64 rpm, and the applied pressure of 0.03 MPa. The examination of the product morphology in an SEM revealed that on raising the temperature and decreasing the creep speed, powdered inulin sample offered spherical shape with smoother surfaces. Based on our research findings, we can accomplish that the spray drying technique may be utilized for the inulin production at commercial scale.

Abbreviations

RSM: Response surface methodology; Rpm: Rotation per minute; ANOVA: Analysis of variance; DP: Degree of polymerization; $Ca(OH)_2$: Calcium hydroxide; SEM: Scanning electron microscope; FT–IR: Fourier transform-infrared; Diameter $_{Avg}$: Average diameter; Diameter $_{max}$: Maximum diameter; Diameter $_{min}$: Minimum diameter

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Authors' contributions

"WA" designed and optimized the study and developed the methodology. "WA" performed the experiments and collection and interpretation of data. "WA" wrote the manuscript. "NG" contributed to the manuscript revision and provided supervision. "WA" and "NG" read and approved the final manuscript.

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Availability of data and materials
The datasets of research were collected from experiments and analysis of variables during current study. These datasets are available from the corresponding author on reasonable request.

Ethics approval and consent to participate
Not applicable.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

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