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Characteristics of thin-layer drying of the cyanobacterium *Aphanothece microscopica* Nägeli

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Abstract

This study was undertaken to investigate the dehydration characteristics of the cyanobacterium *Aphanothece microscopica* Nägeli in a convective hot-air dryer. The dehydration characteristics of the biomass were examined at air temperatures of 40, 50 and 60 °C and sample thicknesses of 3, 5 and 7 mm. During the dehydration experiments, air velocity was held stable at 1.5 m/s (parallel flow). The effects of air temperature and sample thickness on the dehydration characteristics and quality parameters (instrumental colour and chlorophyll *a*) of the dehydrated biomass were determined. The transport of water during dehydration was described by Fick's equation and effective diffusivity was between 8.1×10^{-8} and 18.8×10^{-8} $m^2\ s^{-1}$. The experimental dehydration data on biomass obtained were fitted to the Henderson and Pabis model, and the drying rate constant was between 0.005 and 0.024 min^{-1}. The response surface methodology was very efficient in determining product quality, showing the strong effect of temperature on instrumental colour and chlorophyll *a* content. The thicknesses evaluated showed a less significant effect on the final quality.

Keywords: Cyanobacteria; *Aphanothece*; Drying; Chlorophyll *a*; Colour

1. Introduction

Researchers have given considerable attention to methods of preserving microorganisms in order to enable the dehydration of biomass in the formulation of food destined for human consumption, in feed used in fisheries and even in the extraction of biochemical components of industrial interest [1].

The drying operation is commonly used to prolong the shelf life of microbial biomasses. Preservation of cyanobacteria is a difficult process, since the cells are small (3–30 μm diameter) and, moreover, the cultures are usually diluted (less than 0.5 kg m^{-3} of dry biomass in commercial cultures). Drying is thus done by combining mechanical and thermal separation techniques, in such a way so as to reduce the water content of the material to be dried, starting with filtration operations, flocculation and centrifugation [2].

Drying can be described as a method of industrial preservation by which water content and activity of products are decreased by heated air to minimise biochemical, chemical and microbiological deterioration. The objective in drying food products is to reduce of the moisture content to a level which allows safe storage over an extended period [3]. However, during processing, the material may be exposed to temperatures that have an adverse effect on quality, making these products susceptible to colour deterioration. It has been reported that many reactions can affect colour during thermal processing of fruits and their derivatives. Of these, the most common is pigment degradation, especially of carotenoids and chlorophyll [4,5].

Aphanothece microscopica Nägeli is a cyanobacterium that is found in the estuaries of Rio Grande, RS, Brazil throughout the year. Researchers have shown that this cyanobacterium is an important source of protein, and no histological disturbances were observed in Wistar rats fed a diet containing *Aphanothece* [6]. The protein content of *Aphanothece* is higher than those of traditional foods such as meat, eggs and wheat meal [7]. For single-cell protein production, the use of *Aphanothece* in the

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bioconversion of the nitrogen in parboiled rice effluent in to protein has been studied to associate wastewater treatment with the production of single-cell protein [8].

Thus, the study aimed to determine the effects of drying air temperature and tray thickness on the dehydration characteristics of *Aphanothece microscopica* Nägeli and to study the effects of these parameters on the quality characteristics of the dehydrated biomass.

2. Theoretical considerations

The dehydration of biological materials normally follows a falling-rate drying period. The moisture and/or vapour migration during this period is controlled by diffusion. Assuming that the resistance to moisture migration is uniformly distributed within of the homogenous isotropic material, Fick's second law can be derived as follows:

$$\frac{\partial Ml}{\partial t} = \nabla(D_{ef}\nabla Ml) \quad (1)$$

where Ml is the local moisture content, t is the drying time and D_{ef} is the effective diffusivity.

Assuming that the moisture is initially uniformly distributed throughout the sample, that mass transfer is symmetric with respect to the centre, that the surface moisture content of the sample instantaneously reaches equilibrium with the conditions of the surrounding air and that shrinkage is negligible or not taken into consideration, the solution of Eq. (1) for an infinite slab can be defined as follows [9]:

$$\frac{M-M_e}{M_0-M_e} = \frac{8}{\pi^2}\sum_{n=0}^{\infty}\frac{1}{(2n+1)^2}\exp\left(\frac{-(2n+1)^2\pi^2 D_{ef}t}{4L^2}\right) \quad (2)$$

For long dehydration periods, a limiting form of Eq. (2) is obtained for slab geometries by considering only the first term in the series expansion.

Then, Eq. (2) can be written in the following form:

$$\frac{M-M_e}{M_0-M_e} = \left(\frac{8}{\pi^2}\right)\exp\left(-\frac{\pi^2 D_{ef}t}{4L^2}\right) \quad (3)$$

where M is the moisture content at any time, M_e is the equilibrium moisture content, M_0 is the initial moisture content, L is the half-thickness of the slab in the sample and n is a positive integer.

Thin-layer drying models that describe the drying behaviour of biological materials fall into three categories, namely, theoretical, semi-theoretical and empirical. The semi-theoretical models are generally derived by simplifying general series solutions of Fick's second-law or modifying simplified models and are valid within the ranges of temperature, relative humidity, airflow rate and moisture content for which they were developed [10]. The Henderson and Pabis model [11] is a frequently used thin-layer drying model. The first term of Eq. (2) is known as the Henderson and Pabis model and is written as

$$\frac{M-M_e}{M_0-M_e} = A\,\exp(-kt) \quad (4)$$

where A is the drying coefficient, k is the dehydration rate constant and t is the drying time. This model has been used to determine the drying behaviour of several products [12–15].

3. Material and methods

3.1. Biomass

Aphanothece microscopica Nägeli culture stocks were spread and maintained in standard BGN medium (Bauer & Gauer Medium) [16]. The conditions used were 30 °C, a light of 2 Klux and constant stirring in the growth chamber for a photoperiod of 12 h dark/light. The experiments were set up in a batch cylindrical reactor with 600 L of culture in parboiled rice effluent. They were operated at 30 °C in the absence of light, a C/N ratio of 50, a pH adjusted and kept at 8.0, constant aeration of 1 VVM allowing oxygen saturation and inoculum of 100 mg L^{-1} of *Aphanothece microscopica* Nägeli in the exponential growth phase.

3.2. Harvesting

The biomass was separated from the rice parboiling effluent by centrifugation, as indicated by Molina Grima et al. [2].

3.3. Biomass drying

The drying experiments were performed in a tray dryer, in to which air was blown at a constant speed of 1.5 m/s in a parallel flow at 40, 50 and 60 °C and thicknesses of 3, 5 and 7 mm.

3.4. Product quality

The response surface methodology (RSM) was used to study product quality with two experimental factors (temperature and thickness of tray). A three-level factorial design was employed to study the relationship between the drying conditions (independent variables) and the colour and chlorophyll *a* content (response variables). Experimental design and statistical analysis were performed using Statistica 7.0 software (Statsoft, USA). The ranges and levels of experimental variables investigated are presented in Table 1.

For a two-factor system, the model equation is

$$Y = \beta_0 + \beta_1\cdot x_1 + \beta_2\cdot x_2 + \beta_{11}\cdot x_1^2 + \beta_{22}\cdot x_2^2 + \beta_{12}x_1\cdot x_2 + \beta_{12}x_1^2x_2 + \beta_{12}x_1x_2^2 + \beta_{12}x_1^2x_2^2 \quad (5)$$

Table 1
Values of independent variables at different levels of the 3^2 factorial design

Independent variable	Symbol	Level		
		−1	0	1
Temperature (°C)	x_1	40	50	60
Thickness (mm)	x_2	3	5	7

where Y is the predicted response, β_0 is the intercept, β_1 and β_2 are the linear coefficients, β_{11} and β_{22} are the squared coefficients and β_{12} is the interaction coefficients.

Sample colour was measured with a MINOLTA (CR-300, MINOLTA Corporation, Ramsey, NJ, USA). The colour values were expressed as L (whiteness or brightness/darkness), a (redness/greenness) and b (yellowness/blueness). The hue angle (Eq. (6)) was calculated from the $L-a-b$ values

$$\text{hue angle} = \tan^{-1}\frac{b}{a} \quad (6)$$

Chlorophyll a concentration in the biomass was meansured spectrophotometrically by the method described by Paranhos [17].

4. Results and discussion

The curves which show the drying behaviour of biomass of the cyanobacterium *Aphanothece microscopica* Nägeli under different drying conditions are shown in Figs. 1–3.

It is possible to verify distinct phases for all the conditions evaluated. The first corresponds to the sharper variation in moisture with time, referring to the constant rate period and the first stage of the falling rate drying period. After a transition, a second phase, which corresponds to a less sharp variation in moisture, characterized by the second stage of the falling rate drying period, is observed.

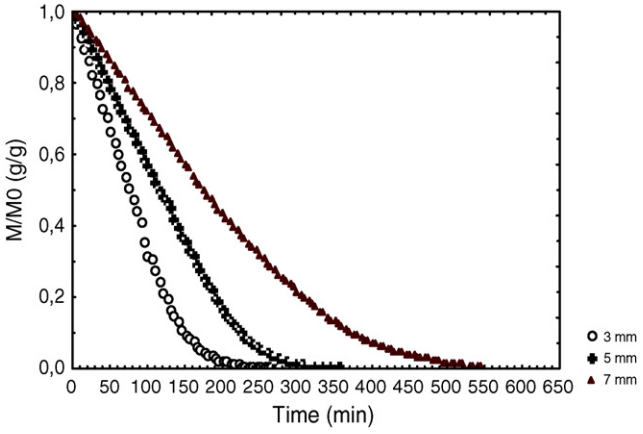

Fig. 1. Drying curves for *Aphanothece microscopica Nägeli* (40 °C).

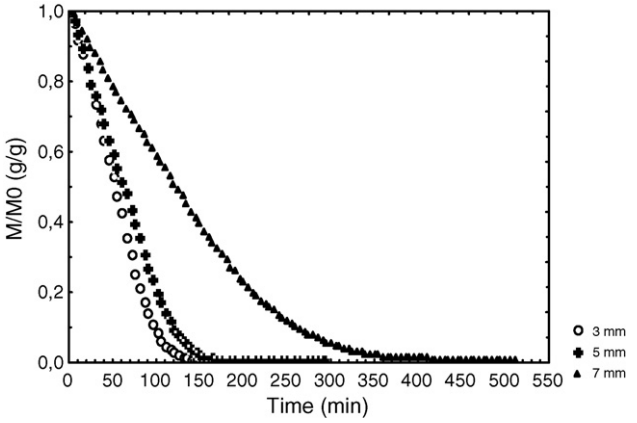

Fig. 2. Drying curves for *Aphanothece microscopica Nägeli* (50 °C).

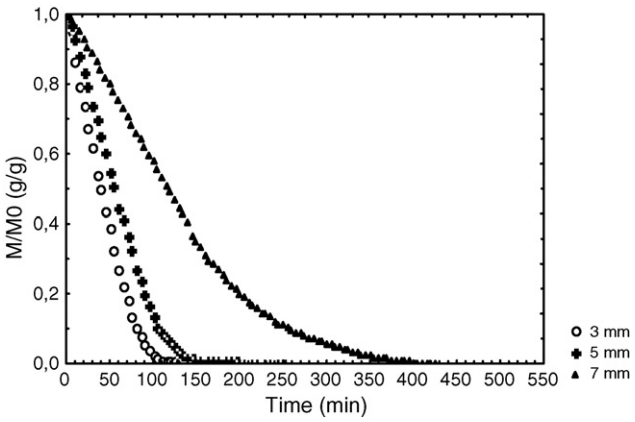

Fig. 3. Drying curves for *Aphanothece microscopica Nägeli* (60 °C).

Table 2 shows a characterisation of the biomass drying of the cyanobacterium *Aphanothece microscopica* Nägeli in a thin layer, with a starting moisture of 94.5% for the different drying conditions.

These data show that the constant rate drying period (t_C), is relevant to the total drying time (above 20% for all the drying conditions). According to Chirife [18], the constant rate drying period is important in situations where the initial moisture content of the material is high.

The values of critical moisture were located in the band between 5.3 and 6.5 for the different drying conditions evaluated. Critical moisture is the moisture content in the end of the constant rate drying period. At these values of moisture, movement of the liquid to the surface of the solid becomes insufficient to replace the liquid, which is being evaporated.

The equilibrium moisture obtained ranged from 5% to 13% for the different drying conditions. These values are regarded as particularly important in the drying operation, since they represent the limit value for the reduction in moisture in the material under a given atmospheric condition. Desmorieux and Decaen [19] report that an equilibrium moisture between 7.0% and 7.5% is the considered the industrial reference in drying cyanobacteria *Spirulina,* which means that only at a temperature of 60 °C is the

Table 2
Characteristics of thin-layer drying of *Aphanothece microscopica Nägeli*

Drying condition	t_C (min)	M_C (g/g)	M_{trans} (g/g)	T (min)	M_e (g/g)
40 °C – 3 mm	85	5.8	0.16	275	0.130
40 °C – 5 mm	150	5.3	0.14	355	0.110
40 °C – 7 mm	280	5.8	0.18	605	0.110
50 °C – 3 mm	55	6.0	0.10	210	0.090
50 °C – 5 mm	65	6.1	0.08	290	0.084
50 °C – 7 mm	110	6.4	0.12	510	0.104
60 °C – 3 mm	50	6.5	0.08	200	0.078
60 °C – 5 mm	60	5.8	0.05	250	0.050
60 °C – 7 mm	145	6.1	0.05	425	0.051

t_C: critical time; M_C: critical moisture; M_{trans}: transition moisture; T_{total}: drying time; M_e: equilibrium moisture.

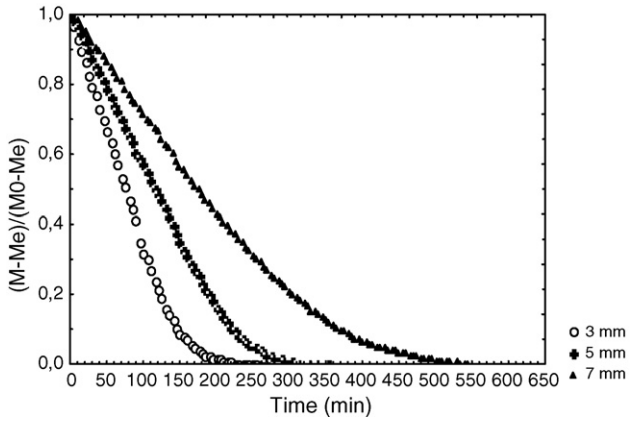

Fig. 4. Curves for dimensionless moisture content (40 °C).

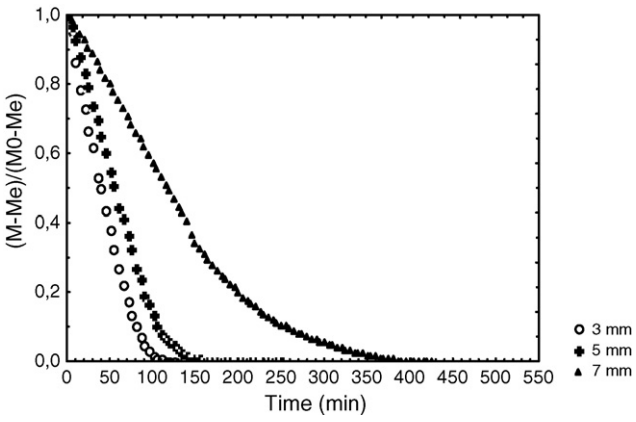

Fig. 6. Curves for dimensionless moisture content (60 °C).

range of commercial moistures for microalgae drying obtained under the conditions evaluated.

The transition, values for moisture under all the drying conditions evaluated, indicate that the first stage of drying is the mainly responsible for the removal of moisture from the biomass of *Aphanothece microscopica* Nägeli under experimental conditions, which proves that the diffusion of liquid water is the physical mechanism that governs the movement of moisture in the biomass of the cyanobacterium *Aphanothece microscopica* Nägeli under the experimental conditions, as suggested by King [20].

The dimensionless moisture content curves (Figs. 4–6), were used for the calculation of drying rate constant.

Table 3 shows the results of a non-linear regression analysis of fitting the semi-theoretical model to the experimental data and the evaluation criteria used to compare the statistical validity of the fit and the values of average effective diffusivity of moisture.

Table 2 shows the good fit of the model for estimation of drying rate constant, which is proved by the values of the correlation coefficient obtained by analysis of the non-linear regression, established from the curves The values obtained for drying rate constants at the different temperatures and thicknesses indicate the effect of these factors with an the increase in the drying constant as the temperature rises and the thickness of the tray decreases. Krokida et al. [21] report that the drying rate constant has a strong correlation with the air conditions and size of the material. These authors claim that the effect of factors such as temperature, relative humidity, air velocity and size of material on the drying rate constant has been widely studied for several products.

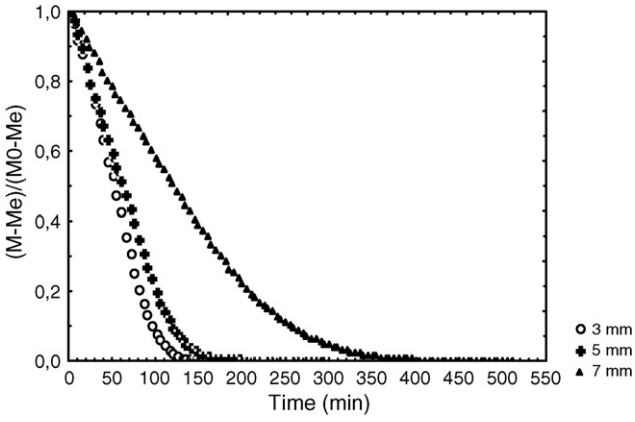

Fig. 5. Curves for dimensionless moisture content (50 °C).

Determination of the transport properties of biological materials must be taken into account in drying these products, so that the effect of drying conditions on the rate of diffusion of water in the biomass of *Aphanothece microscopica* Nägeli is clear.

Colour and chlorophyll *a* content were tested after each drying condition. Results are shown in Table 4.

Statistical analysis (Table 5) of the results showed that, in the range studied, temperature has a strong linear and quadratic effect on chlorophyll *a* content and hue angle at a significance level of 95%.

The response surface curves and the contour plot showing (Figs. 7–10) maximum chlorophyll *a* content and hue angle were obtained at a lower temperatures (40 °C).

Sarada et al. [1] mentions the effect of temperature on pigment stability and colour in dryed cyanobacterium *Spirulina*, indicating that there was a high degree of degradation in temperatures above 45 °C. Chlorophyll *a* is a photosynthetic pigment present in all species cyanobacteria [22]. However, this pigment is highly susceptible to degradation during thermal processing and storage. The conversion of chlorophyll in pheophytins and others derivatives results in a change from bright green to dull olive green or olive yellow, which is ultimately perceived by the

Table 3
Parameter estimation, coefficient correlation and effective diffusivity for the experimental data

Drying condition	A	k (min^{-1})	R	D_{ef}(m^2 s^{-1})
40 °C – 3 mm	1.134	0.013	0.98	9.30×10^{-8}
40 °C – 5 mm	1.146	0.008	0.98	8.10×10^{-8}
40 °C – 7 mm	1.146	0.005	0.99	9.90×10^{-8}
50 °C – 3 mm	1.165	0.019	0.98	15.3×10^{-8}
50 °C – 5 mm	1.249	0.016	0.99	16.2×10^{-8}
50 °C – 7 mm	1.133	0.007	0.99	15.9×10^{-8}
60 °C – 3 mm	1.116	0.024	0.99	17.7×10^{-8}
60 °C – 5 mm	1.153	0.018	0.98	18.2×10^{-8}
60 °C – 7 mm	1.138	0.008	0.99	18.8×10^{-8}

Table 4
Experimental design and results of the 3^2 factorial design

Run	x_1	x_2	Chlorophyll *a*	Hue angle
1	0	1	320.7	103.5
2	1	0	281.7	101.7
3	−1	−1	480.3	110.6
4	−1	0	689.8	108.9
5	1	1	414.4	91.2
6	0	−1	199.0	87.9
7	1	−1	296.1	101.9
8	−1	1	604.3	106.5
9	0	0	284.0	103.2
10[a]	0	1	321.0	103.6
11[a]	1	0	281.3	102.1
12[a]	−1	−1	481.5	111.0
13[a]	−1	0	690.3	109.3
14[a]	1	1	414.0	91.4
15[a]	0	−1	200.5	87.8
16[a]	1	−1	297.4	101.7
17[a]	−1	1	604.2	107.0
18[a]	0	0	283.8	102.7

[a] Replicate.

consumer as a loss of quality [23]. The reduction in hue angle corresponds to a decrease in the intensity of greenness and an increase in yellowness. The reduction in hue angle in this study agrees with the results reported by several authors that inadequate thermal processing caused deterioration of the chlorophyll pigments that resulted in a colour change from green to olive green, and eventually to yellow [24,25].

The results of the second-order response surface model in the form of analysis of variance (ANOVA) are given in Table 6. Fischer's F-test was used to assess the significance of the regression model. With the regression equation R^2-values of 0.9995% and 0.998% were obtained for the chorophyll *a* content and hue angle, respectively. These values ensured a satisfactory adjustment of the quadratic model to the experimental data and

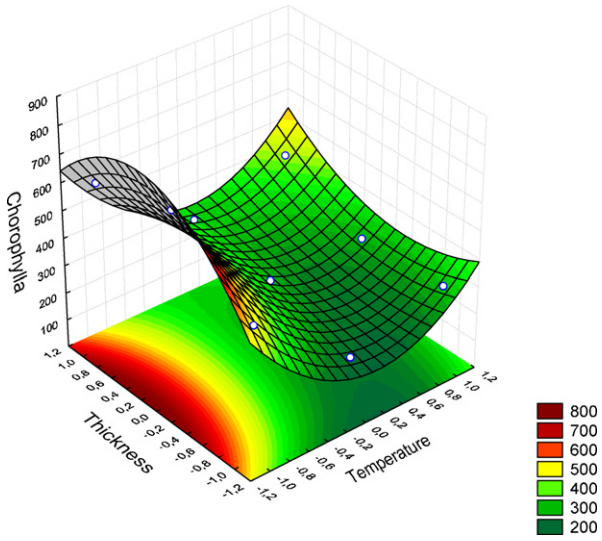

Fig. 7. Response surface curves for chlorophyll *a* content.

indicated that 99% of the variability in the responses could be explained by the model. The following regression equations were obtained (coefficients with a significance level less than 95% were not included).

$$Y = 396.5 - 130.3x_1 + 60.5x_2 - 96.5x_1^2 + 16.3x_2^2 - 1.4x_1 \cdot x_2 - 55.5x_1 \cdot x_2^2 - 3.1x_1^2x_2^2 \quad (7)$$

were Y is the chlorophyll *a* content (μg/g), x_1 is the temperature and x_2 is the thickness (coded values).

$$Y = 101,8 - 5.2x_1 + 0.1x_2 - 2.7x_1^2 + 2.1x_2^2 - 1.6x_1 \cdot x_2 + 1.2x_1 \cdot x_2^2 + 5.7x_1^2 \cdot x_2 + 1.0x_1^2 \cdot x_2^2 \quad (8)$$

were Y is the hue angle (°), x_1 is the temperature and x_2: is the thickness (coded values).

Table 5
Model coefficients estimated by multiple linear regression

Factor	Effect	S.E	*t*(9)	*p*	Coefficient	−95%	+95%
Chlorophyll *a*							
Mean	396.5	0.08	4814.5	0.000	396.5	396.3	396.7
x_1L	−260.7	0.20	−1292.3	0.000	−130.3	−130.5	−130.1
x_1Q	−193.1	0.17	−1105.2	0.000	−96.5	−96.7	−96.3
x_2L	121.1	0.20	600.5	0.000	60.5	60.3	60.8
x_2Q	32.6	0.17	186.6	0.000	16.3	16.1	16.5
x_1L by x_2L	−2.9	0.24	−11.8	0.000	−1.4	−1.7	−1.1
x_1L by x_2Q	−110.5	0.21	−516.5	0.000	−55.2	−55.5	−55.0
x_1Q by x_2L	0.12	0.21	0.5	0.570	0.06	−0.1	0.3
x_1Q by x_2Q	−6.2	0.18	−33.5	0.000	−3.1	−3.3	−2.8
Hue angle							
Mean	101.8	0.05	1832.8	0.000	101.8	101.7	101.9
x_1L	−10.5	0.13	−77.5	0.000	−5.2	−5.4	−5.1
x_1Q	−5.5	0.11	−46.8	0.000	−2.7	−2.8	−2.6
x_2L	0.3	0.13	2.6	0.024	0.1	0.02	0.3
x_2Q	4.2	0.11	36.3	0.000	2.1	2.0	2.2
x_1L by x_2L	−3.2	0.16	−19.3	0.000	−1.6	−1.8	−1.4
x_1L by x_2Q	2.5	0.14	17.4	0.000	1.2	1.09	1.4
x_1Q by x_2L	11.5	0.14	79.8	0.000	5.7	5.59	5.9
x_1Q by x_2Q	2.1	0.12	17.5	0.000	1.0	0.95	1.2

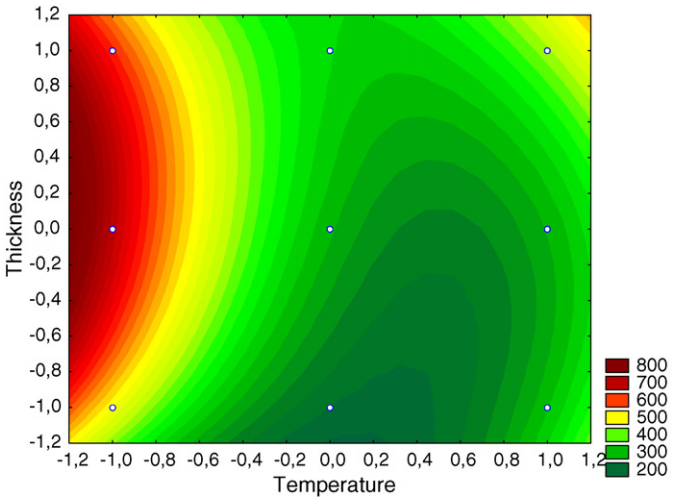

Fig. 8. Contour diagrams for chlorophyll *a* content.

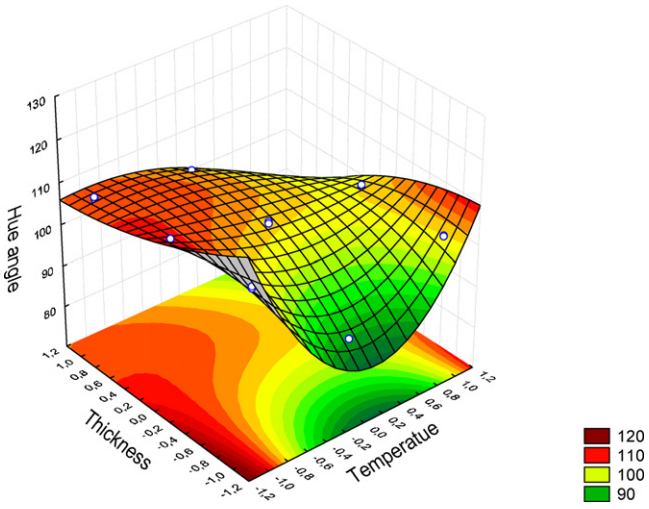

Fig. 9. Response surface curves for hue angle.

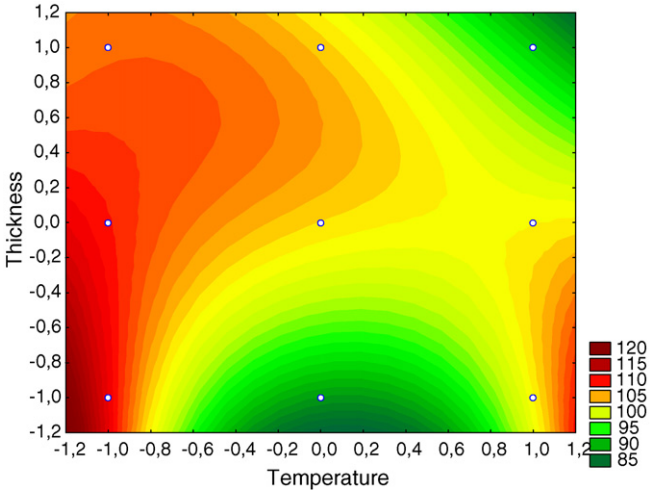

Fig. 10. Contour diagrams for hue angle.

Table 6
Analysis of variance (ANOVA) for the quadratic model

Source variation	Sum of squares	Degrees of freedom	Mean square	F_{value}
Chlorophyll *a*				
Regression	434091.9	8	54261.48	444766.2[a]
Residual	1.1	9	0.12	
Total	434093.0	17		$R^2 = 0.999$
Hue angle				
Regression	551.08	8	117.39	2347.8[a]
Residual	388.58	9	0.05	
Total	388.08	17		$R^2 = 0.998$

[a] Statistical significance ($\alpha = 0.05$).

5. Conclusion

The predominant stage concerning to the migration of moisture in drying biomass of cianobacteria *Aphanothece microscopica* Nägeli in thin layer was the first step of variation of moisture in function of time, which corresponds to the constant rate period and the first stage of falling rate. The values of constants of drying obtained, are situated among 0.005 and 0.024 min^{-1} and among 8.1×10^{-8} a 18.8×10^{-8} $m^2\ s^{-1}$ for the constant of drying and average effective diffusivity of moisture respectively. Response surface methodology was very efficient to determine product quality, showed a strong influence of temperature in colour intrumental and chlorophyll *a* content. The thickness evaluated showed less significance in the final quality.

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