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Production of 4th range iceberg lettuce enriched with calcium. Evaluation of some quality parameters

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Abstract

This project is part of a study on the process of production of 4th range Iceberg lettuce leaves enriched with Ca using the vacuum impregnation (VI) technique. The objectives of this work were (i) to verify if it is possible that a 250 g portion of impregnated lettuce leaves provides the same quantity of Ca (300 mg) as a 250 mL glass of milk, and (ii) to analyze the effects of VI on some quality parameters of the lettuce leaves: water activity, humidity, soluble solids content, real and apparent densities, pH, optical and mechanical properties, and respiration rate. For the study, three zones of the whole lettuce leaf were differentiated (on a longitudinal axis), because of the different distribution of the vascular system: apical (A), medium (M) and basal (B). Iceberg lettuce leaves with a global content of 169 mg of Ca in 250 g of impregnated product were obtained. A fresher, shinier product is obtained without presenting differences in colour. In average, Ca enrichment doesn't affect significantly the mechanical behaviour. The respiration rates and the respiratory quotient remain almost unchanged in the A area, whereas in the M area the intake of O₂ increases lightly, without change in the production of CO₂, and both respiratory rates increase notably in the B area, with an important decrease in the respiratory quotient, indicating that the operation seems to prevent the fermentative anaerobic routes. The study shows it seems possible to obtain a Ca enriched product, being a possible alternative to dairy products. The industrial application of this process should include the control of the hydric state of the vegetable material, to increase impregnation capacity and reduce its variability.

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Keywords: functional foods; vacuum impregnation; physicochemical properties; mechanical properties; respiration rates.

1. Introduction

Consumers demand for functional foods (FF) is greatly and continuously growing. A wide variety of FF is on market at present, but it does not include “natural” functional foods, manufactured from fruits

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and vegetables, that maintain the cellular structure of the raw material. Vacuum impregnation (VI) is a technique that can introduce physiologically active components into the structure of fruit and vegetables [1]. Ca is an important fortifier in functional foods development because Ca content of diet is critical in most stages of life. Industrial probiotic foods are mainly dairy products, but lactose intolerance and cholesterol content are two drawbacks related to their consumption. Vegetable products play an important role in human diet, partly because of their high fibre content. The aim of this research was to study the technological feasibility of manufacturing 4th range Iceberg lettuce leaves enriched with Ca using the VI technique. The objectives of this work were (i) to verify if it is possible that a 250 g portion of impregnated lettuce leaves provides the same quantity of Ca (300 mg) as a 250 mL glass of milk, which is the 37.5% of the Recommended Daily Intake (RDI) (800 mg/day), and (ii) to analyze the effects of VI and of the enrichment with Ca on some quality parameters of the lettuce leaves: water activity (a_w), humidity (water mass fraction, X_w), soluble solids content (Brix), real and apparent densities (ρ_r , ρ_a), pH, colour (CIEL*a*b* coordinates), mechanical properties (maximum stress and strain, work performed and number of peaks up to maximum stress), and respiration rates.

2. Materials & Methods

Less than 2 days harvested Iceberg lettuces (*Lactuca sativa*) were used. For the study, three zones of the whole lettuce leaf were differentiated (on a longitudinal axis), because of the different distribution of the vascular system: apical (A), medium (M) and basal (B). Vacuum impregnation (VI) experiments were carried out in a specially designed equipment [2]. The vacuum period of the VI process was performed at 500 mbar during 10 minutes, and samples remained immersed into the solution of impregnation (SI) during 10 minutes at atmospheric pressure. Prior to analysis, the SI in excess on samples surface was eliminated using a manual centrifuge. Two SI (SucSI and CaSI) were prepared. SucSI was a sucrose aqueous solution of the same a_w than lettuce leaves, used as a VI reference SI. CaSI (also isotonic with the raw material) was prepared with 62.5 g of Ca lactogluconate (Puracal XPRO, product code 72541, PURAC) per liter of water, and presented the following characteristics: Ca content = 5.4 g Ca/L; a_w = 0.986 ± 0.003 ; Brix = 4.13 ± 0.03 ; density = 1020 ± 4 kg/m³; pH = 7.3 ± 0.1 . Three groups of samples from each zone (A, M, B) were obtained and their properties compared: F (fresh, not submitted to VI), VISuc (VI with SucSI), VICa (VI with CaSI). Carbon dioxide production and oxygen consumption were determined at 4°C, and respiratory quotient calculated. Analytical determinations and calculus were performed as described in previous papers [3, 4, 5]. Numerical values of a^* and b^* were converted into hue angle (h^*), chrome (C^*) and colour differences (ΔE) [5].

3. Results & Discussion

Weight percentage of each group of lettuce leaves pieces was determined in 250 g bags of commercial 4th range Iceberg lettuce: A, $50 \pm 4\%$; M, $35 \pm 3\%$; B, $15 \pm 5\%$.

Fresh samples from A, M and B zones (Table 1) showed no differences in a_w (0.992 ± 0.003), X_w (0.950-0.953), ρ_a (0.954-0.957), and ρ_r (1.018-1.019). pH increased from B (5.8 ± 0.2) to M (6.1 ± 0.2) and A (6.4 ± 0.2). "Natural" porosity was quite homogeneous (A: $6.4 \pm 0.4\%$; M: $6.1 \pm 0.8\%$; B: $5.9 \pm 0.2\%$). Nevertheless, previous works [3] demonstrated that this parameter does not provide a good estimate of the capacity of these type of vegetal tissues to get impregnated by VI, because the vascular faeces that are almost full of sap are assessed as part of the not porous tissues, but they play an important role in the course of the VI operation, since they are capable of emptying of sap and to be filled with external SI.

Table 1. Some physicochemical properties of fresh Iceberg lettuce leaves samples.

	Zone A	Zone M	Zone B
X _w (water mass fraction)	0.950 ± 0.004 ^{a b}	0.952 ± 0.003 ^{a b}	0.953 ± 0.003 ^{a b}
a _w (water activity)	0.992 ± 0.003 ^{a b}	0.992 ± 0.003 ^{a b}	0.992 ± 0.003 ^{a b}
pH	6.4 ± 0.2 ^{a d}	6.1 ± 0.2 ^{b d}	5.8 ± 0.2 ^{c d}
ρ _a (g/cm ³) (apparent density)	0.954 ± 0.003 ^{a c}	0.956 ± 0.002 ^{ab c}	0.957 ± 0.003 ^{b c}
ρ _r (g/cm ³) (real density)	1.019 ± 0.002 ^{a c}	1.018 ± 0.008 ^{ab c}	1.018 ± 0.002 ^{b c}
ε (%) (porosity)	6.4 ± 0.4 ^{a c}	6.1 ± 0.8 ^{ab c}	5.9 ± 0.19 ^{b c}

In each row, different superscripts denote significant differences due to the treatment (F or VI) between samples of the same zone, and different subscripts denote significant differences between samples of different zones, for a given treatment.

The operation of VI with CaSI led to obtaining Iceberg lettuce with an average content of 170±50 mg of Ca in 250 g of impregnated product (200±60, 160±15 and 110±20 mg of Ca, for A, M and B zones, respectively), that is to say the 21% of the RDI. It is probable that the use of more concentrated solutions of Ca will allow reaching the objective.

VI with Suc (Table 2) or with Ca (Table 3) did not significantly affect physicochemical properties of lettuce leaves samples. The differences in porosity between zones continued existing. This suggests a similar degree of sap emptying and SI filling of the vascular faeces in the whole leaf, and/or similar interactions between Ca and the pectins of the vegetable walls, so that the viscoelastic response of the lettuce samples under the pressure gradients of the VI operation is not significantly modified.

Table 2. Some physicochemical properties of Iceberg lettuce leaves samples submitted to VI with sucrose solution

	Zone A	Zone M	Zone B
X _w (water mass fraction)	0.91 ± 0.014 ^{a b}	0.942 ± 0.002 ^{a b}	0.945 ± 0.003 ^{a b}
a _w (water activity)	0.992 ± 0.003 ^{a b}	0.986 ± 0.003 ^{a b}	0.992 ± 0.003 ^{a b}
pH	6.3 ± 0.2 ^{a d}	5.9 ± 0.2 ^{b d}	5.7 ± 0.2 ^{c d}
ρ _a (g/cm ³) (apparent density)	0.955 ± 0.002 ^{a c}	0.956 ± 0.002 ^{ab c}	0.956 ± 0.002 ^{b c}
ρ _r (g/cm ³) (real density)	1.033 ± 0.006 ^{a c}	1.022 ± 0.002 ^{ab c}	1.0121 ± 0.002 ^{b c}
ε (%) (porosity)	7.6 ± 0.5 ^{a c}	6.5 ± 0.10 ^{ab c}	6.3 ± 0.13 ^{b c}

In each row, different superscripts denote significant differences due to the treatment (F or VI) between samples of the same zone, and different subscripts denote significant differences between samples of different zones, for a given treatment.

Table 3. Some physicochemical properties of Iceberg lettuce leaves samples submitted to VI with Ca solution

	Zone A	Zone M	Zone B
X _w (water mass fraction)	0.939 ± 0.004 ^{a b}	0.945 ± 0.003 ^{a b}	0.953 ± 0.003 ^{a b}
a _w (water activity)	0.996 ± 0.003 ^{a b}	0.991 ± 0.003 ^{a b}	0.991 ± 0.003 ^{a b}
pH	6.1 ± 0.2 ^{a d}	5.9 ± 0.2 ^{b d}	5.8 ± 0.2 ^{c d}
ρ _a (g/cm ³) (apparent density)	0.954 ± 0.003 ^{a c}	0.955 ± 0.002 ^{ab c}	0.956 ± 0.003 ^{b c}
ρ _r (g/cm ³) (real density)	1.0123 ± 0.002 ^{a c}	1.021 ± 0.008 ^{ab c}	1.018 ± 0.002 ^{b c}
ε (%) (porosity)	6.7 ± 0.2 ^{a c}	6.4 ± 0.11 ^{ab c}	6.1 ± 0.11 ^{b c}

In each row, different superscripts denote significant differences due to the treatment (F or VI) between samples of the same zone, and different subscripts denote significant differences between samples of different zones, for a given treatment.

Ca enrichment of the vegetable doesn't affect significantly its mechanical behaviour (Table 4), though a light increase of the maximum strength and of the shear work was detected for B samples, probably due to the interaction of Ca with the wall pectins and cellular membranes that causes a bigger adhesion between the cells.

Table 4. Mechanical properties

Zone	Treatment (Sample)	Max. strain	Max. stress	F/D	Work up to F	Number of peaks
		D (mm)	F (N)	(N/mm)	(Nmm)	up to F
A	F	2.3 ± 0.4 ^{a c}	3.4 ± 0.8 ^{a c}	1.6 ± 0.3 ^{a c}		
	VISuc	3.0 ± 0.7 ^{a c}	3.1 ± 0.4 ^{a c}	1.5 ± 0.2 ^{a c}		
	VICa ¹	2.2 ± 0.5 ^{a c}	3.2 ± 0.5 ^{a c}	1.6 ± 0.2 ^{a c}		
M	F	2.6 ± 0.7 ^{a c}	4.3 ± 0.9 ^{a c}	1.8 ± 0.4 ^{a c}		
	VISuc	2.4 ± 0.5 ^{a c}	4.3 ± 0.8 ^{a c}	1.8 ± 0.4 ^{a c}		
	VICa ²	2.3 ± 0.4 ^{a c}	4.2 ± 0.9 ^{a c}	2.0 ± 0.6 ^{a c}		
B	F	9 ± 1.3 ^{b c}	28 ± 6 ^{b c}	4 ± 2 ^{b c}	100 ± 17 ^c	8 ± 2 ^c
	VISuc	7 ± 1.6 ^{b c}	33 ± 3 ^{b c}	5 ± 2 ^{b c}	100 ± 20 ^c	4 ± 2 ^c
	VICa ³	8.0 ± 0.9 ^{b c}	35 ± 4 ^{b c}	4 ± 1.5 ^{b c}	130 ± 17 ^c	6 ± 2 ^c

In each column, different superscripts denote significant differences due to the treatments between samples of the same zone, and different subscripts denote significant differences between samples of different zones, for a given treatment.

¹: 80 mg Ca/100 g sample. ²: 62 mg Ca/100 g sample. ³: 35 mg Ca/100 g sample.

A fresher, shinier product is obtained without presenting significant differences in colour (Table 5).

Table 5. Colour parameters (L*, a*, b*, h* and C*) and colour differences (ΔE)

Zone	Zone A		Zone M		Zone B	
	F-A	VICa-A	F-M	VICa-M	F-B	VICa-B
L*	68 ± 5 ^{a d}	66 ± 3 ^{a d}	80 ± 6 ^{b d}	83 ± 4 ^{b d}	56 ± 4 ^{c d}	56 ± 4 ^{c d}
a*	-11 ± 1,4 ^{a c}	-11 ± 1,7 ^{a c}	-10 ± 2 ^{a c}	-13 ± 3 ^{a c}	-1,8 ± 0,4 ^{b c}	-1,3 ± 0,4 ^{b c}
b*	35 ± 4 ^{a c}	37 ± 5 ^{a c}	30 ± 6 ^{a c}	50 ± 4 ^{a c}	7 ± 1,6 ^{b c}	5 ± 1,1 ^{b c}
hue angle h*	108 ± 6 ^{a b}	108 ± 6 ^{a b}	110 ± 10 ^{a b}	104 ± 6 ^{a b}	105 ± 9 ^{a b}	106 ± 10 ^{a b}
chrome C*	37 ± 5 ^{a c}	39 ± 6 ^{a c}	32 ± 7 ^{a c}	52 ± 4 ^{a c}	7 ± 2 ^{b c}	5 ± 1,4 ^{b c}
colour difference ΔE	3 ± 2		2 ± 3		2,3 ± 0,5	

In each row, different superscripts denote significant differences due to the treatment (F or VI) between samples of the same zone, and different subscripts denote significant differences between samples of different zones, for a given treatment.

VI with CaSI doesn't modify the respiration rates nor the respiratory quotient of the zone A lettuce samples (Table 6). For zone M samples, the intake of O₂ increases lightly, without change in the production of CO₂, therefore slightly decreasing the respiratory quotient. For zone B samples, both respiratory rates increase notably, with an important decrease in the respiratory quotient, indicating that the reaction seems to prevent the fermentative anaerobic routes.

Table 6. Respiration rates at 4°C.

Zone	Treatment (Sample)	O ₂ consumption (mL O ₂ kg ⁻¹ h ⁻¹)	CO ₂ production (mL CO ₂ kg ⁻¹ h ⁻¹)	RQ (mL CO ₂ / mL O ₂)
A	F	54 ± 1 ^a	54 ± 1 ^a	1.0 ± 0.2 ^a
	VISuc	53 ± 1 ^a	53 ± 1 ^a	1.0 ± 0.2 ^a
	VICa ¹	54 ± 1 ^a	54 ± 1 ^a	1.0 ± 0.2 ^a
M	F	56 ± 1 ^a	56 ± 1 ^a	1.0 ± 0.2 ^a
	VISuc	55 ± 1 ^a	55 ± 1 ^a	1.0 ± 0.2 ^a
	VICa ²	61 ± 7 ^a	56 ± 1 ^a	0.9 ± 0.12 ^a
B	F	50 ± 1 ^a	50 ± 1 ^a	1.0 ± 0.2 ^a
	VISuc	49 ± 1 ^a	49 ± 1 ^a	1.0 ± 0.2 ^a
	VICa ³	106 ± 2 ^a	71 ± 1 ^a	0.7 ± 0.12 ^a

In each column, different superscripts denote significant differences due to the treatments between samples of the same zone, and different subscripts denote significant differences between samples of different zones, for a given treatment.

4. Conclusions

Following what was said before, you obtain an Iceberg lettuce with a global content of 169 mg of Ca in 250 g of impregnated product (200, 156 and 106 mg, for tip, middle and bottom, respectively), that is to say the 21% of the RDI. It is probable that the use of more concentrated solutions of Ca will allow reaching the objective. A fresher, shinier product is obtained without presenting differences in color. Ca enrichment of the vegetable doesn't affect significantly its mechanical behavior (shear stress-deformation test and puncture test), though a light increase of the maximum strength and of the shear work was detected in the bottom area of the leaf, probably due to the interaction of Ca with the wall pectins and cellular membranes that causes a bigger adhesion between the cells. The operation of VI with Ca lactogluconate doesn't modify the respiration rates nor the respiratory quotient of the lettuce sample's tip area. In the middle area, the intake of O₂ increases lightly, without change in the production of CO₂, therefore slightly decreasing the respiratory quotient. In the bottom area, both respiratory rates increase notably, with an important decrease in the respiratory quotient, indicating that the reaction seems to prevent the fermentative anaerobic routes.

The study shows it seems possible to obtain a Ca enriched product that provides the quantity of said mineral as a glass of milk, being a possible alternative to dairy products. The industrial application of this process should include the control of the hydric state of the vegetable material, with abundant vascular tissue, to increase impregnation capacity and reduce its variability.

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