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The Application of Vacuum Impregnation Techniques in Food Industry

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1. Introduction

The interest of food scientists in the field of microstructure is recently exponentially increased. This interest has raised after the recognition of the importance that chemical reactions and physical phenomena occurring at microscopic scale have on safety and quality of foods. This concept was well resumed from Aguilera (2005) which stated that “...the majority of elements that critically participate in transport properties, physical and rheological behavior, textural and sensorial traits of foods are below the 100 μm range”. As example, Torquato (2000) reported that through microscopic observation it is possible to observe that only a portion of cells of the crumb bread solid matrix are connected, even though at macroscopic level it may appear as completely connected; so, the three dimensional spatial distribution of cell crumbs greatly affects the sensorial quality of bread. Before the scientific recognition of the above consideration, food scientists which focused their efforts on the effects of traditional and innovative industrial processes on food quality, analyzed only macroscopic indexes such as color, texture, taste, concentration of several nutritional compounds, etc. without to consider that they essentially are the result of chemical and physical phenomena occurring at microscopic level. However, with the aim to be more precise in the use of the term “microscopic” we may generalize the classification that Mebatsion et al. (2008) reported for the study of fruit microstructure. The authors considered three different spatial scales: 1. The macroscale which refers the food as a whole or a continuum of biological tissues with homogeneous properties; 2. The mesoscale which refers the topology of biological tissues; 3. The microscale which addresses the difference of individual cells in terms of cell walls, cell membranes, internal organelles, etc. Here, we would like focus the attention on the importance to study the foods at mesoscopic scale by considering them as mesoscopic divided material (MDM). On this basis the majority of foods may be defined as “biological systems where an internal surface partitions and fills the space in a very complex way”. At mesoscopic scale the three dimensional architecture of foods may be studied analyzing the relation between void and solid phases where the voids (capillaries, pores) may be partially or completely filled with liquids or gases. The relations between these two phases and their changes during processing is one of the most important factors affecting the safety and the sensorial and/or nutritional quality of foods. Until few years ago only the porosity fraction of foods was reported in literature as experimental index of the internal microstructure but it gives us only low level of information. Instead, a second level of

structure characterization may be reached by analyzing pore dimension, shape, length, surface roughness, tortuosity, connectivity, etc. So, one of the most important future challenge will be the precise characterization of the three dimensional architecture of foods, its changes during food processing and its relation with safety and quality. However, some pioneering researchers have focused their attention on this research field and the first results are reported in literature (Datta, 2007a; Datta, 2007b; Halder et al., 2007). For instance, the term “food matrix engineering (FME)” has been used to define a branch of food engineering that applies the knowledge about food matrix composition, structure and properties with the aim to promote and control adequate changes that improve some sensorial and/or functional properties in the food products as well as their stability (Fito et al., 2003). The authors reported that food gels and emulsions, extruded, deep fried and puffed foods may be considered as current FME. Instead, among the non-traditional processing, one of the most important and newest techniques based on the properties of food microstructure is the vacuum impregnation (VI). With this term are classified some technologies based on the exploitation of void fraction of foods with the aim to introduce, in a controlled way, chemical/organic compounds into the capillaries of biological tissues. VI is based on the application of a partial vacuum pressure which allows to remove native liquids and gases entrapped into the capillaries and to impregnate them with a desired external solution after the restoration of atmospheric pressure. At first, a vacuum osmotic dehydration (VOD) treatment was proposed and studied as method to accelerate water loss and solid gain during the immersion of vegetables into hypertonic solution. More recently, VI treatments have been studied as method to enrich food with nutritional and functional compounds, to introduce some ingredients with the aim to obtain food with innovative sensorial properties as well as to introduce compounds able to inhibit the most important degradation reactions and the microbial growth. This chapter has the aim to analyze and to discuss the application of vacuum impregnation techniques in food industry. At first, the theoretical principles and the mathematical modeling of the phenomena involved during the application of VI will be discussed. Also, the response of biological tissues to vacuum impregnation will be reviewed. In a second section, focusing the attention of the reader on different potential applications in food industry, the effect of process variables on both impregnation level and quality of final products will be analyzed.

2. Theoretical background and structural changes

The main phenomenon on the basis of vacuum impregnation treatment is the hydrodynamic mechanism (HDM) which was well described from Fito & Pastor (1994), Fito (1994) and Chiralt & Fito (2003). The authors, discussing the results of the water diffusion coefficients (De) obtained with both microscopic and macroscopic analysis during osmotic dehydration (OD), reported that the values were in general 100 times greater when measured at microscopic scale; in particular, De values were about $10^{12} \text{ m}^2/\text{s}$ and $10^{10} \text{ m}^2/\text{s}$ when relieved respectively at microscopically and macroscopically. These observations suggested that a fast mass transfer mechanism, in addition with the traditional diffusion, is involved during OD treatments. This mechanism is the hydrodynamic mechanism which is based on pressure gradients generated from the changes of sample volume and/or externally imposed at non-compartmented section such as intercellular spaces, capillaries, pores, etc. These phenomena play a key role in the solid-liquid operation increasing the rate

of several processes during which mass transfer occurs. The action of HDM was well explained from Chiralt & Fito (2003). When food pieces are immersed in an external solution the surface of samples is washed and the solution partially penetrates into the open pores. After, in line with a deformation of cell membranes due to the loss of native liquids and gases, a pressure gradient is generated and HDM is promoted.

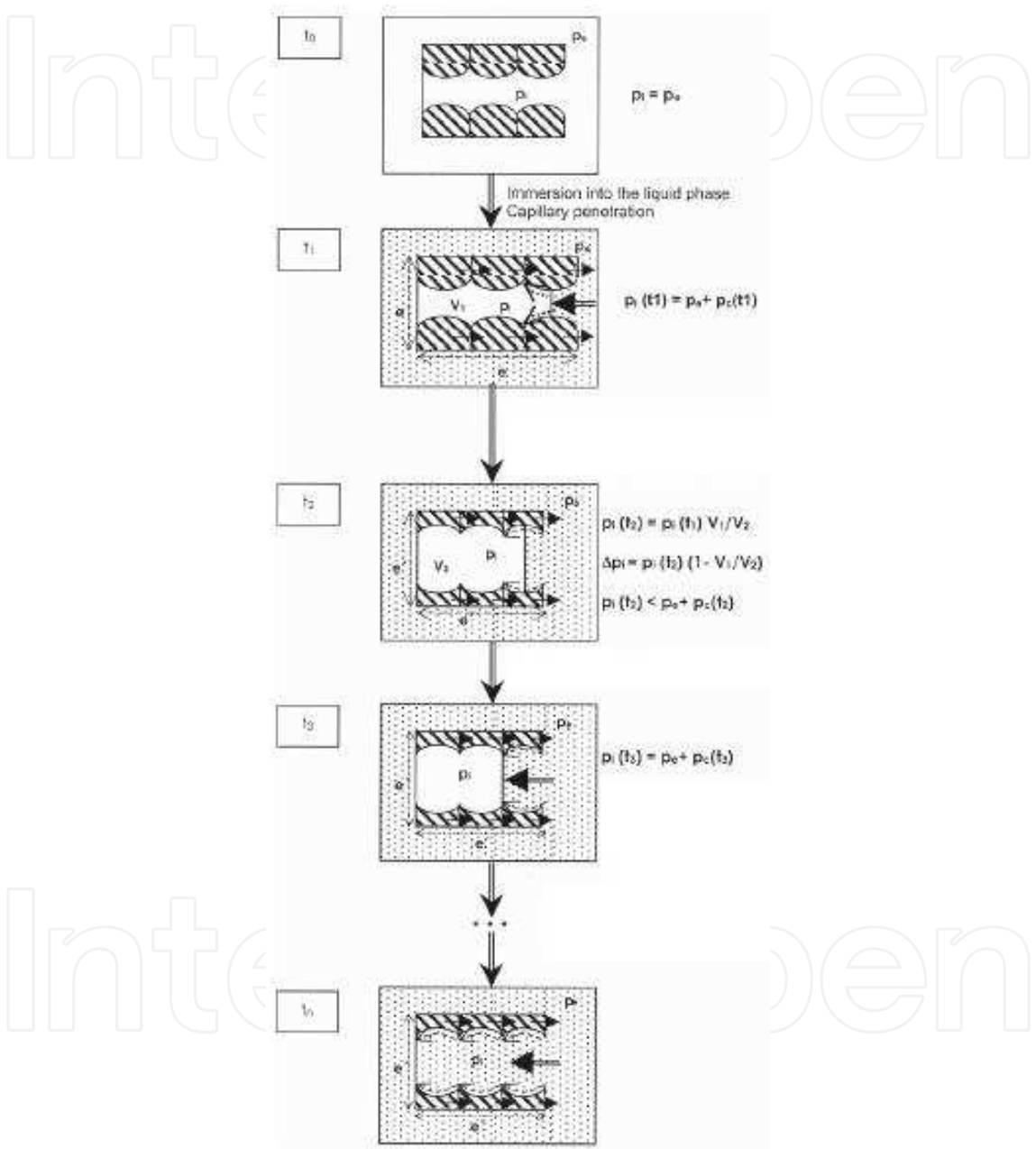


Fig. 1. Schematic representation of hydrodynamic mechanisms due to capillary action and pressure gradients as a consequence of internal volume changes (From Chiralt and Fito, 2003).

In figure 1 a schematic representation of HDM during osmotic dehydration is shown. Before the immersion of vegetable tissues into hypertonic solution the capillary pressure inside the pore is equal to the external (atmospheric) pressure. At t_1 capillary pressure promotes the

initial gain of osmotic solution and the compression of gases inside the pores; so, internal pressure becomes greater than external one. In these conditions gases irreversibly tend to flow out and the cells in contact with hypertonic solution dehydrated due to osmotic pressure gradient. The water loss produces an increase of internal volume as a result of cells shrinkage during dehydration. Moreover, in line with the volume increase the pressure inside the capillary becomes lower to the external one promoting the suction of additional external solution. This process proceeds until the capillary is completely impregnated with the osmotic solution (Barat et al., 1998; Chiralt and Fito, 2003). All these phenomena becomes marked during vacuum impregnation treatments when a vacuum pressure gradient is externally imposed. The treatments are performed through two subsequent steps: 1. The immersion of foods into the solution and the application of a vacuum pressure (p) for a vacuum period (t_1) (also called vacuum time); 2. The restoration of atmospheric pressure maintaining the samples immersed into the solution for a relaxation period (t_2) (also called relaxation time). During VI in addition to HDM a deformation-relaxation phenomena (DRP) simultaneously occurs. HDM and DRP both affect the reaching of an equilibrium situation and their intensities are strictly related with the three dimensional food microstructure and mechanical properties of solid matrix.

Figure 2 schematically shows the phenomena involved during vacuum solid-liquid operations of an ideal pore. At time zero the samples are immersed into the external liquid and the internal pressure of the pore (p_i) is equal to external (atmospheric) pressure (p_e). After, a vacuum pressure (p) is applied in the head space of the system for a time (t_1) promoting a situation in which p_i is greater than p_e . In this condition, the internal gases expand producing the deformation (enlargement) of capillary and the increase of internal volume. Moreover, native liquids and gases partially flow out on the basis of the pressure gradient (step 1-A). At this time hydrodynamic mechanism begins and external liquid partially flows inside the capillary as a consequence of the pressure gradient. These phenomena simultaneously occur until the equilibrium is reached (step 1-B). In the second step the atmospheric pressure in the head space of the system is restored and the samples are maintained into the solution for a relaxation time (t_2). During this period, the generated pressure gradient ($p_i < p_e$) promotes both HDM and solid matrix deformation (compression) which respectively produce capillary impregnation and the reduction of pore volume until a new equilibrium is reached (Fito & Chiralt, 1994; Fito et al., 1996).

2.1 Mathematical modeling of vacuum impregnation and related structural changes

Fito (1994) and Fito et al. (1996) were the first scientists which translate in mathematical language the phenomena involved at mesoscopic scale during VI treatments. The model is based on the analysis of the contributions of both liquid penetration and solid matrix deformation of an ideal pore of volume $Vg_0 = 1$ during each step of VI. From time $t = 0$ to the step 1B a situation expressed by the following equation is obtained:

$$Vg_{1b} = 1 + Xc_1 - Xv_1 \quad (1)$$

Where Vg_{1b} is the pore volume at the step 1-B, Xc_1 is the increment of pore volume due to the expansion of internal gases and Xv_1 is the partial reduction of pore volume due to the initial suction of external liquid as a consequence of HDM.

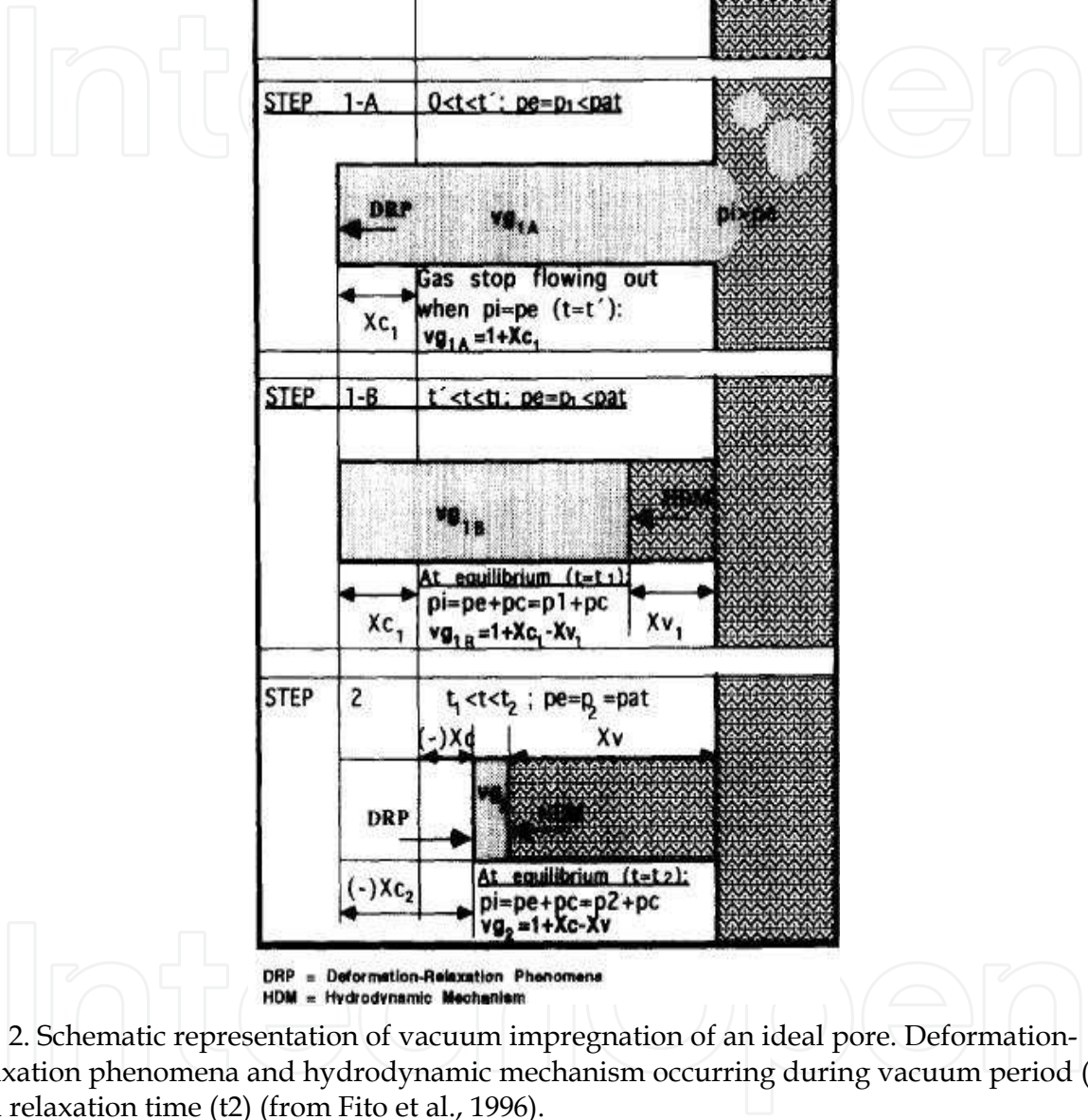


Fig. 2. Schematic representation of vacuum impregnation of an ideal pore. Deformation-relaxation phenomena and hydrodynamic mechanism occurring during vacuum period (t_1) and relaxation time (t_2) (from Fito et al., 1996).

At the end of step 2 the total liquid penetration and matrix deformation may be described respectively by the equations:

$$Xv = Xv_1 + Xv_2 \tag{2}$$

$$Xc = Xc_1 + Xc_2 \tag{3}$$

where Xv_1 and Xv_2 are the volume reduction due to liquid penetration respectively at the end of step 1 and 2; Xc_1 and Xc_2 are the volume pore changes as a result of solid matrix

deformation (enlargement and compression) after the steps 1 and 2. Also, the total volume variation at the end of the process may be described as follow:

$$Vg_2 = 1 + X_c - X_v \quad (4)$$

Equations 2 and 3 may be used to calculate liquid penetration and solid matrix deformation of the total sample taking into account its porosity fraction value (ϵ_e):

$$X = \epsilon_e X_v \quad (5)$$

$$g = \epsilon_e X_c \quad (6)$$

As reported from Fito et al. (1996) when a pressure variation applied in a solid-liquid system and an equilibrium situation is reached, HDM assumes an isothermal compression of gas into the pores. So, the situation reached at the end of step 1-B may be mathematically express as:

$$\frac{V_{g1B}}{V_{g1A}} = \frac{1 + X_c - X_v}{1 + X_c} = \frac{1}{r} \quad (7)$$

Where r is the apparent compression rate (\sim atmospheric pressure/vacuum pressure, dimensionless) (Zhao and Xie, 2004). From equation 7 may be obtained the following:

$$\frac{X_v 1}{1 + X_{c1}} = 1 - \frac{1}{r} \quad (8)$$

Also, by using the equation 5 and 6 it is possible to obtain:

$$X_1 = (\epsilon_e + \gamma_1) \left(1 - \frac{1}{r_1} \right) \quad (9)$$

On these basis the equilibrium situation at the end of step 1-B may be mathematically expressed as:

$$X_1 - \gamma_1 = \epsilon_e \left(1 - \frac{1}{r_1} \right) - \frac{\gamma_1}{r_1} \quad (10)$$

Furthermore, the same considerations may be extended for the phenomena involved from step 1 and step 2. So, between $t = t_1$ and $t = t_2$ the equilibrium situation may be expressed as:

$$X - \gamma = (\epsilon_e + \gamma) \left(1 - \frac{1}{r_2} \right) - \gamma_1 \quad (11)$$

Starting from the above equations it is possible to calculate the porosity value (ϵ_e) at the end of VI process from the value of X , γ and γ_1 by:

$$\epsilon_e = \frac{(X - \gamma)r_2 + \gamma_1}{r_2 - 1} \quad (12)$$

Where X is the volume fraction of sample impregnation by the external liquid at the end of VI treatments (m^3 of liquid/ m^3 of sample at $t=0$), ε_e is the effective porosity, γ_1 is the relative volume deformation at the end of vacuum period (t_1 , m^3 of sample deformation/ m^3 of sample at $t=0$); γ is the volume deformation at the end of process (m^3 of sample deformation/ m^3 of sample at $t=0$). However, although the measure of porosity fraction value is easily obtained from the apparent (ρ_a) and real density (ρ_r) values of the sample (Lewis, 1993; Gras, Vidal-Brotons, Betoret, Chiralt & Fito, 2002), the experimental estimation of X , γ and r are not easy to measure and some modifications of the equipment used for the experiments are necessary (Salvatori et al., 1998). Briefly, the experimental methodology is performed precisely weighting the sample at different steps of the treatment even if it is under vacuum condition (Fito et al., 1996; Salvatori et al., 1998). The authors defined a parameter called magnitude (H):

$$H_t = X_t - \gamma_t = \frac{L_0 - L_t - M_w}{V_0 \rho} \quad (13)$$

Where L is the weight measured in the balance (kg), M_w is the liquid evaporated during the experiment, V is the sample volume (m^3) and ρ is the liquid density (kg/m^3). Instead the subscripts 0 and t refer respectively the time zero and any time t_i corresponding at each step of the process. Practically speaking the magnitude H is an overall index of the contribution of both liquid penetration (X) and solid matrix deformation (γ) on sample volume. A theoretical curve of H value as a function of time during vacuum impregnation is shown in figure 3.

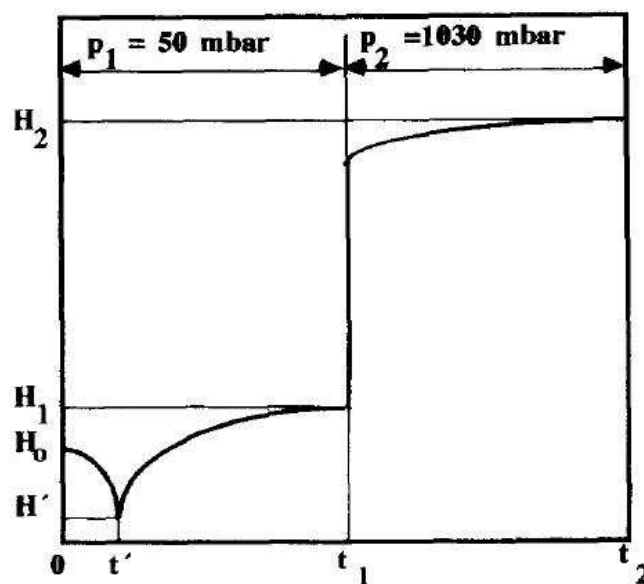


Fig. 3. Theoretical curve of H value as a function of time during vacuum impregnation treatments (From Fito et al., 1998).

From the figure it is possible to observe that as vacuum pressure is applied in the system, H value quickly decreases until H' at $t = t'$. This is because sample volume increases under the action of the gas expansion due to the positive pressure gradient ($p_i > p_e$). During the vacuum period (from $t = t'$ to $t = t_1$) H value slowly increases because native liquids and gases flow out, solid matrix relaxes and external liquid begins to impregnate pores due to

HDM. H_1 is obtained when the equilibrium condition is reached at the end of step 1B. At time $t = t_1$ the restoration of atmospheric pressure ($p = 1030$ mbar) leads a sudden increase of H value as a consequence of liquid penetration. Also, during relaxation time (from $t = t_1$ to $t = t_2$) pore volume reduction due to capillary compression and HDM simultaneously occur. H_2 value is obtained when the new equilibrium situation will be reached. From the above consideration, the crucial importance of microscopic properties of foods such as dimension and shape of samples, their three dimensional architecture, the resistance of biological tissue to gas and liquid flow, solid matrix deformation, etc., is obvious. In particular, these factors affect the kinetics of all phenomena simultaneously involved during VI; so, the quality of foods will be a result of the rates by which each phenomena occur. For instance, if the solid matrix relaxation of food is slow, capillary impregnation could occur without significant deformation. On the other hand a fast deformation could significantly reduce capillary impregnation. Salvatori et al. (1998) studied the time evolution of H value of several vegetables submitted to VI at 50 mbar for different vacuum period (t_1) and relaxation time (t_2). As example, figure 4 shows the results obtained from apple samples (Granny Smith) submitted to VI for a $t_1 = t_2 = 15$ minutes.

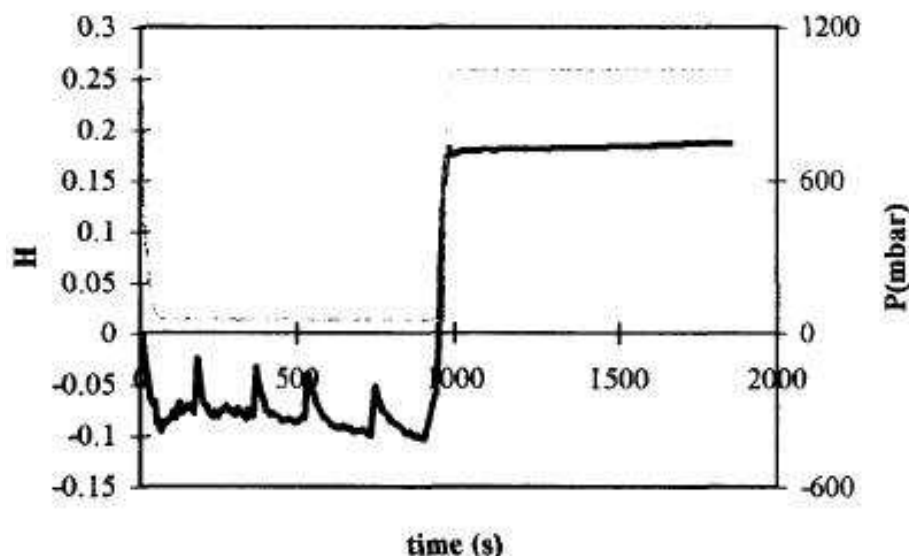


Fig. 4. Experimental curve of apple sample submitted to VI treatment at 50 mbar for a $t_1 = t_2 = 15$ minutes (From Salvatori et al., 1998).

As expected, H value decreased quickly after the application of vacuum pressure in line with pores expansion. However, in disagreement with the behavior of an ideal pore reported in figure 3, at the end of vacuum period (~ 1000 s), H value was negative ($H_1 < 0$) stating that pore volume deformation still was greater than liquid penetration due to the enlargement of pores. In particular, the authors reported that at the end of vacuum period the X_1 and γ_1 values (an average of all experiments performed in different operative conditions) respectively were -4.2% and 1.7% . The negative value of the liquid penetration at the end of vacuum period was explained on the basis of native liquid release from the pores under the action of the negative pressure gradient ($p_e < p_i$). The peaks values observed in figure 1 are in line with the variability of both tissue structure characteristics and the surrounding fluid properties (Salvatori et al., 1998). After, the restoring of atmospheric pressure led to a significant increase of H value due to the compression of pores ($\gamma < 0$) and

the suction of external liquid ($X > 0$). Indeed at the end of the process the authors reported a value of X and γ of 15% and -0.6% respectively. However, foods may show significant different trends as a function of their unique microstructure properties which are very different from the behavior of an ideal pore. For instance, mango and peach samples as well as oranges showed a positive solid matrix deformation (enlargement of the pores) also after the restoration of atmospheric pressure (Salvatori et al., 1998; Fito et al., 2001). Gras et al. (2002) reported that carrot, diced zucchini and beetroot, submitted to VI treatment in a sucrose isotonic solution, showed a pore volume increase ($\gamma > 0$) also at the end of process coupled with significant impregnation values of 16%, 20% and 7% respectively. These behaviors were explained with an overall situation in which the rate of liquid impregnation being very fast in comparison with the solid matrix deformation allowed to keep a residual capillary expansion.

2.2 Gas, liquid and solid matrix volume changes during VI

With the aim to have a complete theoretical analysis of the phenomena involved during VI it is important to consider the gases, liquid and solid matrix volume changes occurring during vacuum and relaxation times. Barat et al. (2001) proposed an experimental approach to study the volume changes during VI assuming food constituted of three phases: solid matrix (SM), liquid phase (LP) and gas phase (GP). On this basis the authors reported that total volume changes which occur during vacuum impregnation treatment may be mathematically express as:

$$\Delta V = \Delta V^{LP} + \Delta V^{GP} + \Delta V^{SM} \quad (14)$$

Moreover, since the variation of solid matrix volume may be assumed equal to zero equation 14 may be reduced at:

$$\Delta V = \Delta V^{LP} + \Delta V^{GP} \quad (15)$$

Here, ΔV may be obtained by pycnometer method of fresh and impregnated food whereas the volume changes of liquid phase (ΔV^{LP}) may be estimated by the following equation (Barat et al., 2001; Atares, Chiralt & Gonzales-Martinez, 2008):

$$\Delta V^L = \left\{ \left[m^t (x_w^t - x_s^t) \right] / r_{LP}^t - \left[m^0 (x_w^0 - x_s^0) \right] / r_{LP}^0 \right\} / V^0 \quad (16)$$

Where m is the mass of sample (g), x_w and x_s are respectively the moisture and solid content of sample (water or solid, g/g of fresh vegetable), ρ_{LP} is the density of liquid phase (g/mL) and V is the volume of samples (mL). Moreover, the superscripts t and 0 refer respectively to each time treatment (t_i) and fresh vegetable.

The liquid phase density values may be estimated by the follow equation (Atares et al., 2008):

$$r_{LP} = (230z_s^2 + 339z_s + 1000) / 1000 \quad (17)$$

where z_s is the solid content of sample liquid phase (g solid/g liquid phase). This method has been proved to give precise results during osmotic dehydration process (Barat et al., 2000; Barat et al., 2001). In particular, Barat et al. (2001) plotted the ΔV as a function of ΔV^{LP}

of apple (Granny Smith) samples submitted to traditional osmotic dehydration (OD) and pulsed vacuum osmotic dehydration (PVOD) performed applying a pressure of 180 mbar for a $t_1 = 5$ minutes.

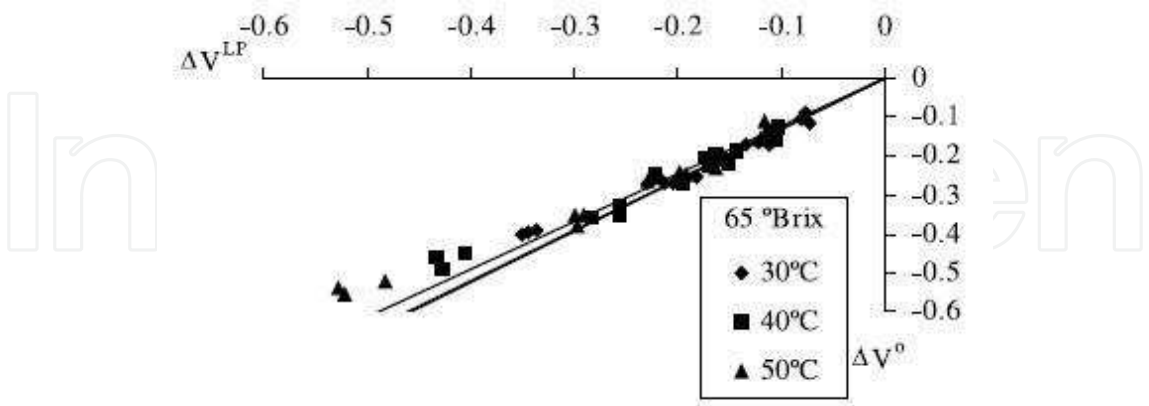


Fig. 5. Total volume changes as a function of liquid phase volume changes of apple samples submitted to OD and PVOD in different operative conditions.

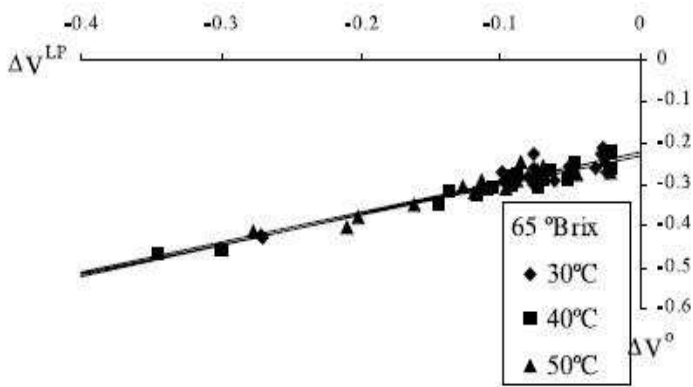


Fig. 6. Total volume changes as a function of liquid phase volume changes of apple samples submitted to OD and PVOD in different operative conditions.

Samples submitted to OD showed a linear trend with no significant intercept value stating that no significant total volume variation was observed before liquid penetration occurring under the action of HDM. By linear regression of experimental data it is possible to estimate the relative contribution of ΔV^{LP} and ΔV^{GP} by using the following equation:

$$\Delta V = s_1 \Delta V^{LP} \tag{18}$$

$$\frac{\Delta V^{GP}}{\Delta V^0} = 1 - \frac{1}{s_1} \tag{19}$$

In comparison with OD treatments, it is worth noting (Figure 6) that samples submitted to PVOD showed a significant intercept with an average value of 0.20. This may be considered as the result of the initial vacuum pulse which promotes the pores expansion coupled with the removal of native liquid and gases and the action of HDM. In this case the linear regression of experimental data shown in figure 6 assumes the following form:

$$\Delta V = i_2 + s_2 \Delta V^{LP} \quad (20)$$

Also, in the case of PVOD the general form of the equation 15 was modified with the following:

$$\Delta V = \Delta V^{LP} + \Delta V^{LP-VI} + \Delta V^{CR} \quad (21)$$

Where ΔV^{LP-VI} and ΔV^{CR} are the volume change due to liquid impregnation and volume changes due to compression-relaxation of solid matrix respectively. In equation 21 the two additional terms substituted ΔV^{GP} . Also, it is important to note that these two terms explain the intercept value of equation 20 and their sum is close to the porosity of food (for apples sample about the 0.22). In their conclusion the authors stated that osmotic dehydration in porous fruits may be explained in terms of LP and GP changes in line with the flow of gases and external liquid and the changes of pore volume as a consequence of the enlargement and compression of pores.

3. Process variables

As previous reported, vacuum impregnation is a technique that allows to introduce several chemical compounds and/or ingredients in the void phase of foods. The process is performed by applying a vacuum pressure in the head space of the system for a time t_1 and then by restoring atmospheric pressure for a relaxation time t_2 . The numerous phenomena involved during the process are affected by several variables which may be classified as external and internal of foods. In the first class, vacuum pressure (p), time length of vacuum period (t_1), time length of relaxation time (t_2), viscosity of external solution, temperature, concentration of solution, product/solution mass ratio and size and shape of the samples may be introduced. Instead, the three dimensional architecture of food and the mechanical properties of the biological tissues may be considered as internal variables. However, it is worth nothing that the term "three dimensional architecture" refers to an ensemble of microscopic and mesoscopic characteristics such as porosity, size and shape of pores, connectivity, tortuosity, capillary curvatures, etc. which greatly affect the vacuum impregnation treatment. In this paragraph the effects of each variable above reported, considered both singularly and as synergistic effects will be discussed.

3.1 External variables

Among the external variables, vacuum pressure may be considered as the most important because it represents the force that produces the pressure gradient between the void phase of food and the atmosphere surrounding the external liquid. Briefly, vacuum pressure is the variable by which all phenomena previously reported may occur. In general, a vacuum pressure ranged between 50 and 600 mbar is reported in literature (Fito et al., 1996; Rastogi et al., 1996; Salvatori et al., 1998; Barat et al., 2001; Fito et al., 2001a; Fito et al., 2001b; Giraldo et al., 2003; Mujica-Paz et al., 2003; Zhao & Xie, 2004; Silva Paes et al., 2007; Corzo et al., 2007; Derossi et al., 2010; Derossi et al. 2011). Also, vacuum level is generally considered directly related to an increase of impregnation level (X) as a consequence of a higher release of native liquids and gases coupled with a greater HDM and DRP. Mujica-Paz et al. (2003a) studied the effect of vacuum pressure in a range of 135-674 mbar on the volume of pores impregnated from an isotonic solution of several fruits. The authors showed that for apple, peach, papaya and melon samples a greater impregnation was observed in line with an increase of vacuum

pressure; instead, for mango, papaya and namey X values increased with the increasing of vacuum level until a maximum after that impregnation slightly decreased. Mujica-Paz et al. (2003b) studied the effect of vacuum pressure (135-674 mbar) on the weight reduction (WR), water loss (WL) and solid gain (SG) of apple, melon and mango slices kept in a hypertonic solution for a $t_1 = t_2 = 10$ minutes. For all experiments, melon and mango samples showed a positive WR, stating that fruits lost a significant fraction of their weight; instead, apples showed a negative WR values, stating a weight gain. In particular, for melon samples the authors reported a direct correlation between vacuum pressure and WR probably because as lower the vacuum pressure as greater the capillary impregnation, which decreased the weight reduction of the samples caused by both the osmotic dehydration and the removal of native liquids from the pores. Indeed the negative values observed for apple samples were explained on the basis of their high porosity fraction (~27.3%). In this conditions, the high free volume of apples increased the impregnation level more than water loss leading to an increase of weight of samples (figure 7a and 7b). Also, similar results were observed for water loss which assumed positive values for mango and melon and negative values in certain operative condition for apple samples (figures 8a and 8b).

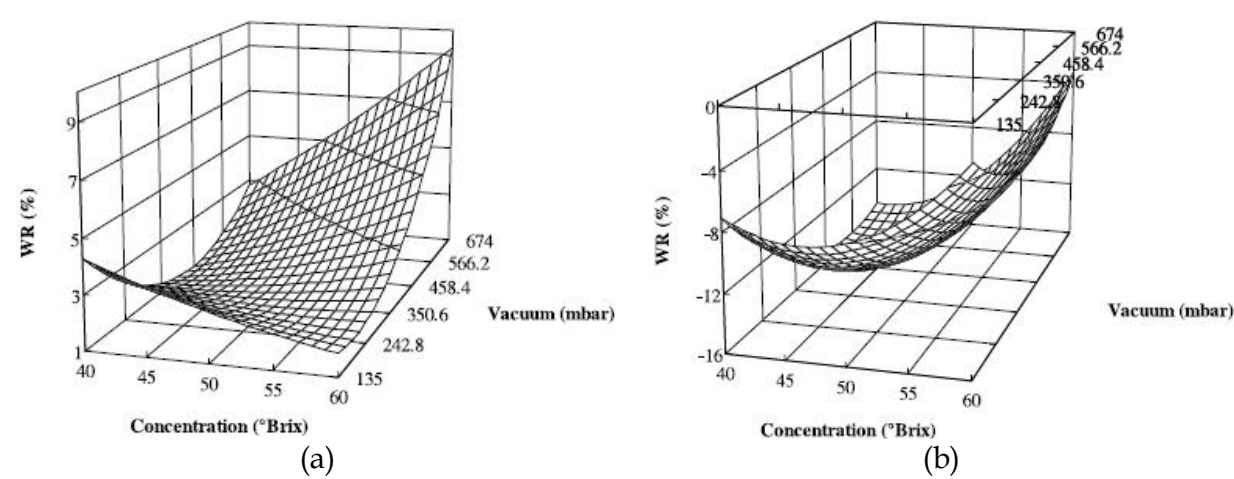


Fig. 7. Effect of vacuum pressure and osmotic solution concentration on weight reduction of melon and apple samples (from Mujica-Paz et al., 2003).

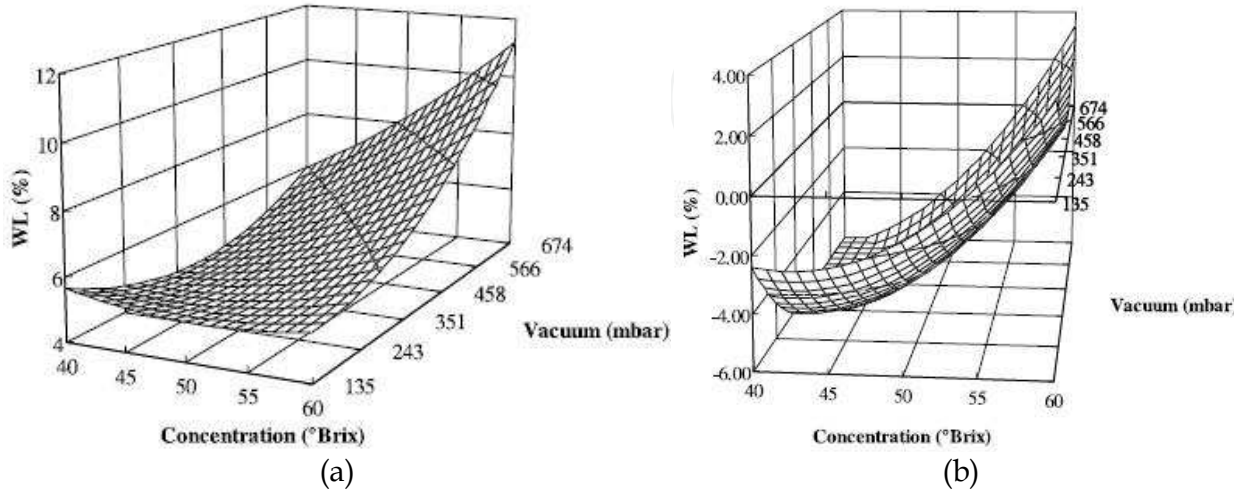


Fig. 8. Effects of vacuum pressure and osmotic solution concentration on water loss of mango and apples samples (from Mujica-Paz et al., 2003).

Derossi et al. (2010), studying the application of a vacuum acidification treatment on pepper slices, reported that pH ratio values (RpH) were lower when a pressure of 200 mbar was used in comparison with the results obtained applying a pressure of 400 mbar. Furthermore, some authors studied the effect of a decrease of vacuum pressure on the acidification of zucchini slices, showing that RpH values were directly correlated with vacuum pressure (Derossi et al., 2011). Nevertheless, the same authors did not observe a statistically significant variation of the porosity of the impregnated samples when vacuum level increased from 400 to 200 mbar. However, Derossi et al. (2010; 2011) showed that the use of a vacuum pressure significantly improved the rate of acidification in comparison with a traditional acidifying-dipping at atmospheric pressure. The authors attributed this result to the increase of acid-solution contact area due to the capillary impregnation. Hofmeister et al. (2005) studied the visual aspect of Mina cheese samples submitted to vacuum impregnation in different operative conditions. The authors reported a greater impregnation level when a pressure of 85.3 kPa was applied in comparison with the experiments performed at 80 kPa. In agreement with the theoretical principles this result was assumed as a consequence of a greater removal of air from the pores of cheese samples. Moreover, Andres et al. (2001) studied the effect of vacuum level on apple samples showing that impregnation level was affected by the applied vacuum pressure.

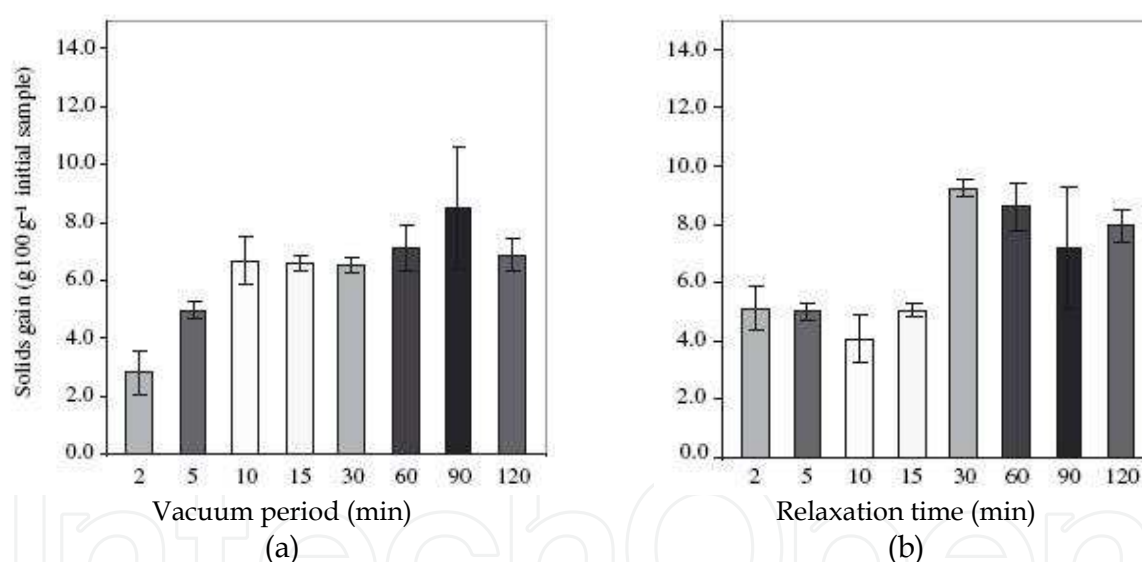


Fig. 9. Solid gain values of apple cylinders submitted to vacuum osmotic dehydration at 40 mbar as a function of vacuum and relaxation times (from Silva Paes et al (2007).

Vacuum period and relaxation times represent two important variables affecting the results of the application of VI. Vacuum period (t_1) refers to the time during which food microstructure tends to reach an equilibrium situation after the application of the vacuum pulse. As previously reported, several phenomena such as deformation (enlargement) of capillaries, the expulsion of gases and native liquids from the pores and the partial impregnation of pores, simultaneously occur during t_1 . Instead, relaxation time (t_2) is the period during which food structure moves toward an equilibrium situation after the restoration of atmospheric pressure. During t_2 , capillary impregnation (by HDM) and deformation (compression) of pores occur under the action of a negative pressure gradient.

In general an increase of impregnation level of foods as a function of the time length of both t_1 and t_2 should be expected. Nevertheless due to the complex equilibrium occurring among the several phenomena involved the interpretation of the results is very difficult. Fito et al. (1996) and Salvatori et al. (1998) studied the response of several fruits (apple, mango, strawberry, kiwi, peach, banana, apricot) on both the X and γ values when submitted to vacuum impregnation at 50 mbar for a t_1 ranged between 5 and 15 minutes and a relaxation time ranged between 5 and 20 minutes. In both cases the authors did not observe significant differences of volume impregnated and solid matrix deformation as a function of t_1 and t_2 variation. Derossi et al. (2010) did not show a significant difference of total mass variation of pepper slices submitted to vacuum acidification at 200 mbar for a vacuum time of 2 and 5 minutes. On the contrary the results reported from Silva Paes et al (2007) stated a great influence of vacuum period and relaxation time on water gain (WG), solid gain (SG) and weight reduction (WR) of apple cylinders submitted to vacuum osmotic dehydration. As example figures 9a and 9b respectively show the solid gain of apple samples as a function of vacuum period and relaxation time.

It is worth nothing that SG values were directly correlated with an increase of t_1 in the range of 2 and 10 minutes. Instead, when a greater vacuum time was applied, no significant differences were observed. The authors suggested that when a short t_1 was applied, the removal of gases from the pores was not complete; so, their residues could have hindered the osmotic solution penetration. When vacuum period increased from 10 to 120 minutes, the complete removal of gases made the increase of t_1 unable to influence solid gain. In figure 9b it is possible to observe an increase of solid gain until a relaxation time of 30 minutes after that SG values are approximately constant. The authors hypothesized that 30 minutes is the time necessary to reach the equilibrium situation after which no differences were observed. Mujica-Paz et al. (2003a), studying the application of vacuum treatment to mango, papaya, peach and melon samples, showed that vacuum time directly affected the pore volume impregnated from external solution in a range of 3-25 minutes. Instead, the authors reported that vacuum time did not influence the X values for banana and apple samples. In the same way Salvatori (1997) reported that t_1 in a range of 5 and 15 minutes did not affect the impregnation level for mango and peach samples. Guillemín et al. (2008) studied the effect of sodium chloride concentration, sodium alginate concentration and vacuum time on weight reduction, chloride ions concentrations and pectinmethylesterase activity (PMEa) of apple cubes. The results showed that the effect of vacuum period was less significant and the more difficult to explain. Hofmeister et al. (2005) reported that an increase of vacuum time significantly affected the impregnation level of Mina cheese submitted to intermittent vacuum impregnation at 85.3 kPa. Chiralt et al. (2001), reviewing the scientific literature concerning the application of vacuum impregnation in salting process, reported that for meat fillets as longer the vacuum time as smaller the weight loss, in accordance with a greater pore volume impregnated from brine solution. Derossi et al. (2010) showed a significant positive effect of vacuum time on the pH reduction of pepper slices submitted to VI at a pressure of 200 mbar; instead a not clear behavior was observed for the experiments performed at 400 mbar. Moreover, in all cases the authors showed a positive effect of the increase of relaxation time on acidification level of the samples in a range of 10 and 30 minutes. Hironaka et al. (2011), studying the enrichment of whole potato with ascorbic acid through a vacuum impregnation treatment showed that an increase of

vacuum time ranged between 0 and 60 minutes was effective in the improvement of ascorbic acid concentration of different varieties of whole potatoes. Apart the three above variables which are much strictly related to the vacuum process, the ensemble of the external solution characteristics with particular attention to its viscosity and temperature significantly affect the quality of vacuum impregnated foods. As reported from Barat et al. (2001), which studied the structural changes of apple tissues during vacuum osmotic dehydration, the impregnation level may be inhibited from a high viscosity of the hypertonic solution. The authors speculated that the relaxation of samples could occur when sample is taken out from the osmotic solution leading to an impregnation (regain) of gas if a liquid with very high viscosity is used. Also, the authors correlated the volume changes caused by capillary impregnation, ΔV^{LP-VI} , and the volume changes due to compression-relaxation phenomena, ΔV^{CR} , as a function of viscosity and temperature of the osmotic solution (figure 10).

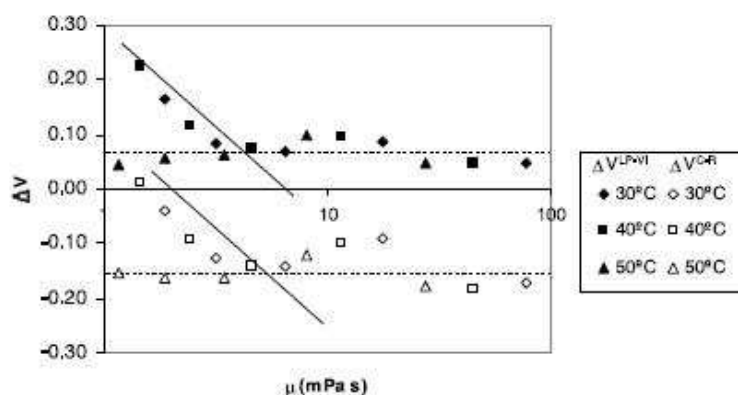


Fig. 10. Relationship between viscosity and temperature of osmotic solution and volume changes due to impregnation and compression-relaxation phenomena of apple samples submitted to vacuum osmotic dehydration (from Barat et al., 2001).

It is worth noting that for the experiments carried out at 40°C and 50°C the increase of viscosity did not show effects on impregnation and deformation. Instead, for samples treated at 30°C and for a viscosity of osmotic solution ranged between 0.5 and 3.5 mPa it is possible to observe that as viscosity increased ΔV^{LP-VI} and ΔV^{CR} respectively decreased and raised. This behavior may be explained with a slow flow (because hindered from the high viscosity) of the osmotic solution into the pores; in this way more time for the relaxation process of the solid matrix is available which becomes the major contribution to the total volume changes of food. In line with this consideration, Barat et al. (2001) stated that: “when viscosity is very low, impregnation tends to the theoretical value of a non-deformable porous matrix, and the deformation tends to zero”. Mujica-Paz et al. (2003), studying the effects of osmotic solution concentration on water loss of apple, melon and mango samples, reported similar results. In general they showed that WL values increased in line with an increase of hypertonic solution concentration due to the greater osmotic dehydration; nevertheless, apple samples showed negative value of WL (a gain of water) for solution concentration lower than 50° Brix. In this condition the penetration of osmotic solution probably was the most important phenomenon which produced an increase of water content; also, it is important to take into account that the experiments performed from the authors were constituted from a vacuum period and a relaxation time

of 10 minutes that represent a very short osmotic treatments to generate a high dehydration. However, for the experiments performed with a solution concentration $> 50^\circ\text{Brix}$, WL values were positive because the OS has difficulty to penetrate into the pores and the pressure gradient promoted the outflow of water from the apple microstructure. Similar effect of the osmotic solution concentration was observed from other authors (Martinez-Monzo et al., 1998; Silva Paes et al., 2007).

The temperature of solution directly affects the mass transfer of process like osmotic dehydration, acidification, brining, etc., but also affects the viscosity of external liquid and the viscoelastic properties of solid matrix of foods. The latter properties which are one of the most important internal variables will be reviewed in the following paragraph; in general, as foods are soft as higher is the contribution of relaxation phenomena on the total volume changes which, in turn reduces the impregnation level.

3.2 Internal variables

Vacuum impregnation is a technique strictly dependent from the characteristic of the food structure both at mesoscopic and microscopic scale (Fito et al., 1996; Salvatori et al., 1998; Barat et al., 2001; Fito et al., 2001; Chiralt & Fito, 2003; Giraldo et al., 2003; Gras et al., 2003; Mujica-Paz et al., 2003; Zhao & Xie, 2004; Derossi et al., 2010; Derossi et al., 2011). At first, the porosity fraction of biological tissue is the most important parameter for the application of VI because it presents the void space potentially available for the influx of the external solution. In general, fruits and vegetables show a higher porosity fraction in comparison with meats, fishes and cheeses making them more suitable for the use of VI techniques. In table 1 the porosity fraction values of several foods are reported.

In general, as greater is the porosity fraction as greater is the impregnation level. Fito et al. (1996) showed that for apple, banana, apricot, strawberry and mushroom samples the impregnation level was in the same order of the effective porosity values of fresh vegetables. Gras et al. (2003) studied the calcium fortification of eggplants, carrots and oyster mushroom by VI treatments. The authors reported the maximum X values for eggplants and oyster mushrooms which showed, in comparison with carrots, the greater intercellular spaces respectively of $54\% \pm 1$ and $41\% \pm 2$. Fito et al. (1996), studying the HDM and DRP on apple, banana, apricot, strawberry and mushroom samples submitted to VI, showed that the impregnation levels were directly correlated with the porosity values of the vegetables.

However, the porosity value is a not sufficient index to completely characterize and to predict the behavior of food during vacuum impregnation. As reported from Zhao & Xie (2004), during VI three main phenomena are involved: gas outflows, deformation-relaxation of solid matrix and the liquid influx. Since these phenomena simultaneously occur, the result of VI is a consequence of the equilibrium among their kinetics which, in turn, are affected by (Fito et al. 1996):

- Tissue structure (pores and size distribution)
- Relaxation time of the solid matrix, a function of the viscoelastic properties of the material;
- The rate of HDM, a function of porosity, size and shape of capillaries, their connectivity, the viscosity of solution;
- Size and shape of samples.

Food (variety, type)	Porosity fraction (ϵ , %)	Reference
Apple (Granny Smith)	23.8±1.0	Salvatori et al. (1998)
Apple (Golden Delisious)	27.3±1.1	Mujica-Paz et al. (2003b)
Apple (Gala)	18.3	Silvia Paes et al. (2007)
Mango (Tommy Atkins)	9.9±1.3	Salvatori et al. (1998)
Strawberry (Chandler)	6.3±1.6	Salvatori et al. (1998)
Kiwi fruit (Hayward)	2.3±0.8	Salvatori et al. (1998)
Pear (Passa Crassana)	3.4±0.5	Salvatori et al. (1997)
Plum (President)	2±0.2	Salvatori et al. (1997)
Peach (Miraflores)	2.6±0.5	Salvatori et al. (1998)
Peach (Criollo)	4.6±0.2	Mujica-Paz et al. (2003a)
Apricot (Bulida)	2.2 ± 0.2	Salvatori et al. (1997)
Pienapple (Espanola Roja)	3.7±1.3	Salvatori et al. (1997)
Banana (Giant Cavendish)	~ 9	Fito et al. (1996)
Banana (Macho)	1.6±0.3	Mujica-Paz et al. (2003a)
Orange peel (Valencia Late)	21±0.04	Chafer et al. (2000)
Mandarina peel (Satsuma)	25±0.11	Chafer et al. (2001)
Eggplant (Saroya)	64.1±2	Gras et al. (2001)
Zucchini (Blanco Grise)	4.4±0.9	Gras et al. (2001)
Zucchini	8.39±2.22	Derossi et al. (2011)
Mushroom (Albidus)	35,9±1.9	Gras et al. (2001)
Oyster mushroom	16.1±4	Gras et al. (2001)
Mango (Manila)	15.2±0.1	Mujica-Paz et al. (2003b)
Melon (Reticulado)	13.3±0.6	Mujica-Paz et al. (2003b)
Carrot (Nantesa)	13.7±2	Gras et al. (2001)
Beetroot	4.3±1.3	Gras et al. (2001)
Cherry	~ 30	Vursavus et al (2006)
Persimonn (Rojo Brillante)	4.9±0.8	Igual et al. (2008)
Cheese (Manchego type)	~ 3	Chiralt et al. (2001)

Table 1. Porosity fraction values of several foods

Moreover, further internal variables such as the tortuosity of the internal pathway and the effect of temperature on the mechanical properties of solid matrix greatly affect the phenomena above reported. Furthermore, a synergistic (positive or negative) effects of the internal variables were reported from several authors. Among the above variables the mechanical properties of biological tissues are one of the most important, because HDM is based on the pressure gradient generated by both vacuum pressure externally imposed and the deformation-relaxation of solid matrix. Salvatori et al. (1998) studied the effects of VI on pore volume impregnated from external (osmotic) solution and on deformation phenomena of several fruits. Strawberries, even tough showed a greater porosity fraction (6.3%) in comparison with kiwi fruit and peach, reported a negligible impregnation level ($X = 0.2$). It was hypothesized that the kinetic of liquid penetration was longer than the relaxation time promoting the deformation (compression) of pores rather than impregnation phenomena. These differences in the rate of HDM and DRP phenomena could be attributed to the microscopic properties of the strawberry tissues such as high tortuosity of the internal

pathways and/or size and shape of pores which hindered the influx of the external solution. On the other hand strawberries being a soft material could be characterized from a fast compression rate which reduced the liquid penetration. In agreement with these hypothesis the authors showed a high deformation (compression) index ($\gamma = -4.0$). Also, in the same paper it was reported that mango and peach samples showed positive γ values at the end of relaxation time, stating that an enlargement of the capillaries was still observed when the equilibrium was reached. In this case a high rigidity of the vegetable tissues could have reduced the rate of compression phenomena and increased the liquid penetration. However, among the studied fruits, strawberries and kiwi fruits were those with a lower impregnated pore volume (respectively of 0.2% and 0.89%) and between them kiwi fruits showed the lowest porosity fraction of $\sim 2.3\%$. Mujica-Paz et al. (2003a) studied the effect of vacuum pressure on the pore volume impregnated from an osmotic solution. Figures 11a and 11b respectively show the prediction of X values of apple and others fruit samples as a function of the factor $(1 - 1/r)$. Since r is defined as the ratio between atmospheric pressure and work pressure the x -axis reports an index of pressure gradient intensity.

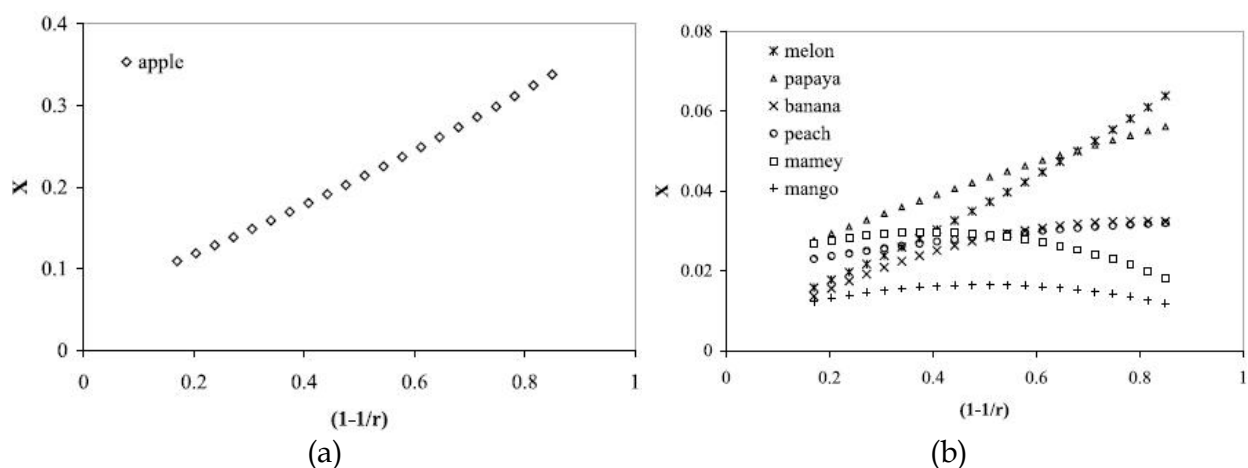


Fig. 11. Prediction of impregnation level value (X) as a function of $(1 - 1/r)$ for some fruit samples submitted to VI for a vacuum time of 10 minutes and relaxation time of 25 minutes (From Mujica-Paz et al., 2002)

Mango, papaya, namey and peach samples showed a linear increase of X , stating that their vegetable tissues are subject to low deformation phenomena. Instead, banana, mango, namey, and peach showed a linear increase of X values until certain limits (about 400 mbar), after which impregnation level decreased. The authors supposed that these operative conditions could have produced a high vegetable tissue deformation (compression), which reduced the void phase available for liquid penetration. It was concluded that these fruits suffered of a great deformation when submitted to VI treatments in particular operative conditions. In their conclusion the authors highlighted the importance of the internal variables such as the number and the diameter of pores as well as the mechanical properties of solid matrix. Moreover, the spatial distribution of cells and their characteristics as well as the kind of fluid (liquid or gas) present in the intercellular space must be considered for the precise setting of vacuum impregnation treatments. Fito et al. (2001) deeply studied the vacuum impregnation and osmotic dehydration in matrix engineering. Among the obtained results, the authors reported that although orange peel showed a porosity fraction lower

than eggplants, the fruit may be considered more suitable for VI treatments. This is because in orange peel the cells in flavedo zone are densely packed but the albedo zone shows large shape cells with a rupture of cell junctions that occurs during fruit ripening. This peculiar cell space distribution gives sponge-like properties with a high impregnation and swelling capacity. Taking into account the effect of the size of samples, it can be state that as thinner the food piece as greater the impregnation level because a greater void space is exposed to the surface. Gras et al. (2003) reported that carrot slices submitted to VI treatments showed an impregnation level of 5.8 ± 0.3 , 4.6 ± 0.7 and 3.4 ± 0.3 respectively for samples with a diameter of 10 mm, 15 mm and 25 mm. Derossi et al. (2011), studying the effect of a pulsed vacuum acidification treatment on zucchini slices in different operative conditions, showed that no significant differences were observed when the experiments were performed at 200 and 400 mbar in terms of pH reduction. It was hypothesized that the variability of vegetable microstructure coupled with the small diameter of the samples (~ 1.5 cm) favored the capillary impregnation reducing the effect of an increase of vacuum level. In accordance with this result the authors showed that no differences were observed in terms of porosity value reduction when fresh zucchini slices were submitted to VI at 200 mbar and 400 mbar. However the interaction of all these variables act simultaneously and could interact within them producing unexpected and/or complicated result. For instance, Chiralt et al. (1999) stated that samples with narrow pores treated with viscous liquid tend to show more deformation than impregnation effect.

4. Industrial applications

Back in the past, when HDM was proposed as an additional mass transfer mechanism occurring during osmotic dehydration, the first application of VI techniques had the aim to increase the rate of osmotic dehydration through the impregnation of capillaries with hypertonic solutions. Since then, vacuum osmotic dehydration (VOD) was subject to a great number of scientific papers; in addition to VOD, the application of an initial vacuum pulse followed by the restoration of atmospheric pressure and the maintaining of food immersed into the osmotic solution for a long time was proposed as pulsed vacuum osmotic dehydration (PVOD). In the last 10 years a great number of industrial applications have been studied (Javeri et al., 1991; Ponappa et al., 1993; Rastogi et al., 1996; Fito et al., 2001a; Fito et al., 2001b; Gonzalez-Martinez et al., 2002; Betoret et al., 2003; Giraldo et al., 2003; Gras et al., 2003; Mujica-Paz et al., 2003a ;Mujica-Paz et al., 2003b; Zhao & Xie, 2004; Hofmeister, et al., 2005; Corzo et al., 2007; Silva Paes et al., 2007; Igual et al., 2008; Cruz et al., 2009). In general it is possible to consider two principal aims: 1. The increase of mass transfer; 2. The introduction of chemical and biological compounds in food microstructure with the aim to improve their quality. On these basis, VI may be used as pre-treatment before drying or freezing, as innovative treatment inserted into a more complex processing with several industrial applications. In figure 12 is schematically reported some potential application of VI in food industry (Zhao & Xie, 2004).

4.1 VI as method to increase the rate of mass transfer of food during food processing

As known, several industrial processes, carried out with the aim to extend the shelf life or to improve/change their nutritional, functional or sensorial properties, are based on mass transfer toward foods or *vice versa*. If the purpose is to increase the rate of mass transfer

between food and an external solution as in the cases of osmotic dehydration acidification treatments, vacuum impregnation significantly increases the rate of the processes due to HDM and the increase of liquid-product contact area. A large literature concerning the effect of VI on the rate of osmotic dehydration recognized the effectiveness of VI (Shi & Fito, 1994; Fito et al., 1996; Rastogi et al., 1996; Tapia et al., 1999; Cunha et al., 2001; Moreno et al., 2004). Shi & Fito (1994) reported that water loss of apricot samples occurred much faster when vacuum was applied during the experiments. Shi et al. (1995) showed that osmotic dehydration under vacuum promotes a faster water diffusion when apricots, pineapples and strawberries were treated. Rastogi et al. (1996), studying the kinetics of osmotic dehydration of apple and coconut samples showed that the estimated rates were significantly higher in vacuum conditions (235 mbar). Fito et al. (2001b) stated that VI promotes the effective diffusion in the fruit liquid phase when impregnated with low viscosity solution.

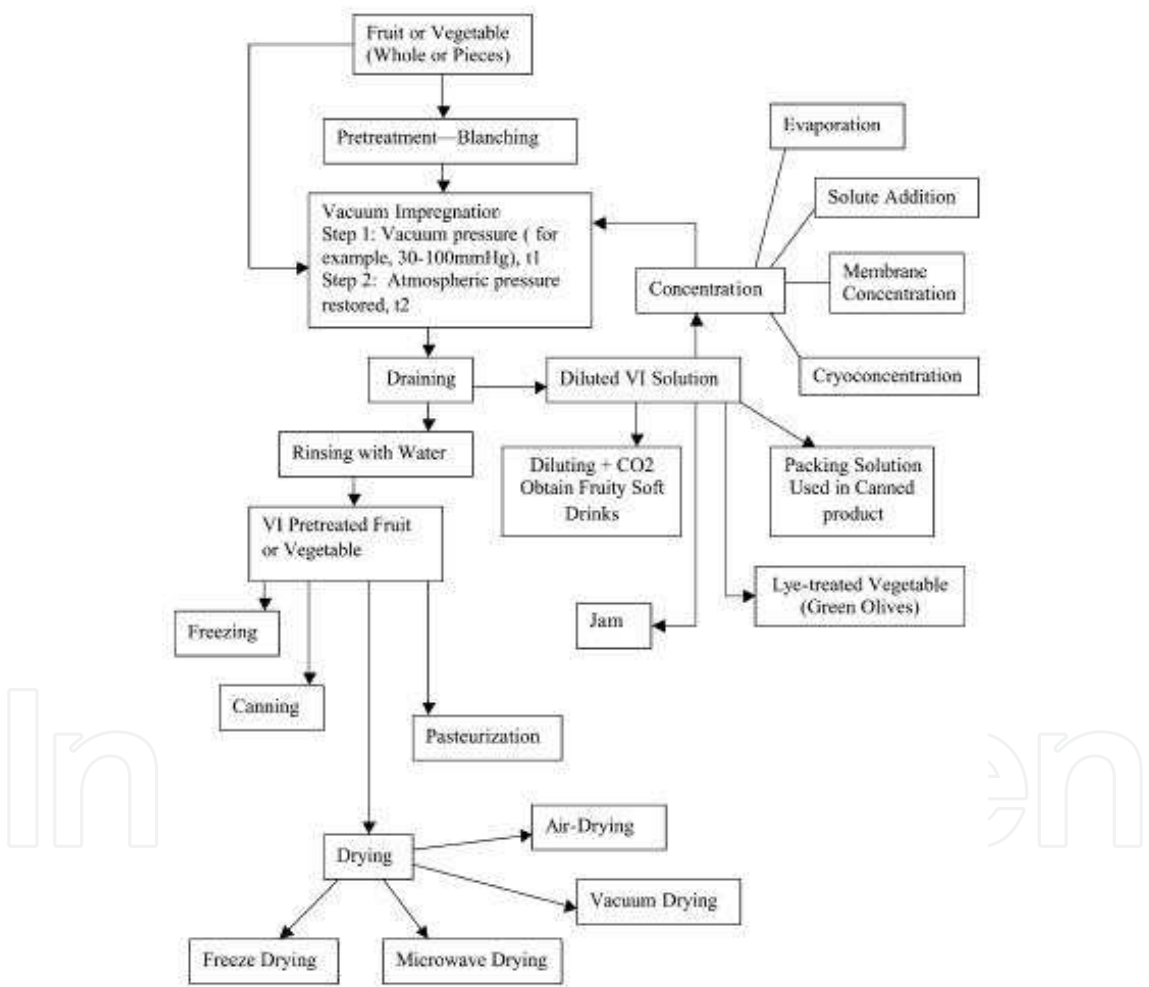


Fig. 12. Potential application of vacuum impregnation techniques in food industry (from Zhao & Xie, 2004)

Figure 13 shows the estimated diffusion coefficient values (D_e) of different type of fruits submitted to traditional osmotic dehydration and pulsed osmotic dehydration. It is possible to observe higher D_e values in the case of VI for vegetables which have high porosity values stating the effectiveness of the treatment as a consequence of the increase of mass transfer. Fito

et al. (1994) found that water loss and solid gain obtained with a PVOD treatment performed at 70 mbar with a t_1 of 5 minutes were greater than those obtained through a traditional OD. Mujica-Paz et al. (2003a) performed osmotic dehydration treatments under vacuum (135-674 mbar, $t_1=t_2=10$ min) and at osmotic pressure measuring water ($t = 20$ min) activity depression (Da_w , %) for apple, melon and mango samples. The authors showed that in all cases the experiment carried out at sub-atmospheric conditions allowed to obtain the greater Da_w values stating that water loss and solid gain (the two main phenomena involved in the water activity depression) were faster than at atmospheric pressure. Giraldo et al. (2003) studied the kinetic of OD and PVOD treatments of mango samples performed with different osmotic solutions concentrations (35 – 65 °Bx). The authors, using a simplified Fickian equation, estimated De values of 5.9×10^{-10} m²/s, 6.4×10^{-10} m²/s and 9.7×10^{-10} m²/s for PVOD performed with osmotic solution concentration respectively of 65°Bx, 55°Bx and 35°Bx; in the same conditions were estimated De values of 1.8×10^{-10} m²/s, 5.9×10^{-10} m²/s and 5.9×10^{-10} m²/s for OD experiments. These results stated that in PVOD diffusion in the tissues is promoted, due to the removal of gas in the intercellular space and the introduction of hypertonic solution. Moreover, the application of VI as substituted of traditional brining of fishes, meats and cheeses or acidifying-dipping showed to be effective for the increase of mass transfer. Gonzales-Martinez et al. (2002) studied three type of brining methods applied to Machengo type cheese: at atmospheric pressure (BI), at vacuum pressure (VI) and with an initial vacuum pulse of 30 minutes (PVI). The authors estimated diffusion coefficients by using diffusional mass transfer mechanism. They obtained in the upper part of the samples De values of 4.4×10^{-10} m²/s, 6.1×10^{-10} m²/s and 9.5×10^{-10} m²/s respectively for BI (traditional), PVI and VI treatments. Also, similar results were obtained for the lower part of the samples. In their conclusion the authors stated that BI in vacuum condition greatly improved the kinetics of salting process as a consequence of hydrodynamic mechanism. Derossi et al. (2010), studying the application of an innovative vacuum acidification treatment (VA) on pepper slices, reported RpH values of 0.968, 0.929 and 0.894 for a traditional acidifying-dipping, a VA performed at 400 mbar ($t_1 = 5$ min, $t_2 = 30$ min) and a VA performed at 200 mbar ($t_1=5$ min, $t_2=30$ min) respectively. In agreement with the theoretical principles of VI the authors concluded that VA is able to increase the rate of pH reduction as a consequence of the capillary impregnation which improve the acid solution-product contact area.

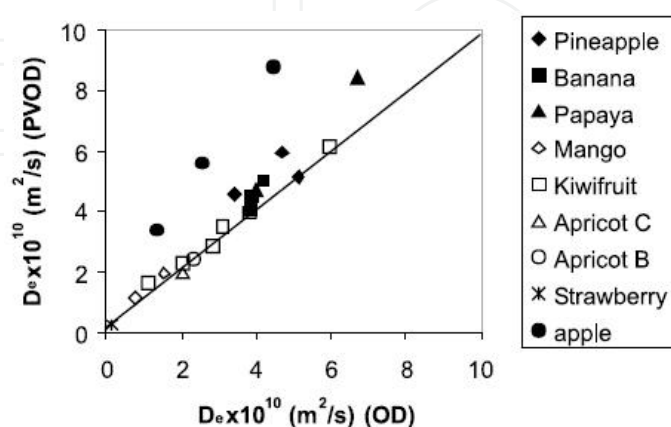


Fig. 13. Comparison of De values obtained from traditional osmotic dehydration (OD) and from pulsed vacuum osmotic dehydration (PVOD). Results obtained from several fruits (From Fito et al., 2001b)

Also Derossi et al. (2001) studied the kinetics of pulsed vacuum acidification treatments (PVA) of zucchini slices. The authors reported that an increase of vacuum did not affect the rate of pH reduction but in all cases the treatments were faster than acidification treatments performed at atmospheric pressure.

4.2 VI as method to improve food quality

As previously reported vacuum impregnation may be considered a technique by which is possible to introduce, in a controlled way, an external liquid in the void phase of foods. The term “external liquid” refers to a liquid obtained by dissolving in water any added chemical components. So, it is clear that VI may be used to introduce several compounds such as anti-browning agents, firming agents, nutritional compounds, functional ingredients, anti-microbial agents, anti-freezing, enzymes, etc. with several aims such as to prolong shelf life, to enrich fresh food with nutritional and/or functional substances, to obtain innovative food formulations as pre-treatments before drying or freezing, etc.

As well known, drying process performed with hot air, microwave, etc., is characterized by high energy consumption hence, high expenses; in this way, the possibility to reduce the initial amount of water in fresh foods (free water) by different techniques would allow to significantly reduce the cost of the process. Several papers showed the effectiveness of a traditional osmotic dehydration before air drying in the reduction of energy consumption. Instead, through a vacuum impregnation treatment applied before air drying two goals may be simultaneously obtained: 1. The reduction of energy costs; 2. The introduction of solutes such as antimicrobial, antibrowning and antioxidant to improve the final quality of dried foods (Sapers et al., 1990; Torreggiani, 1995; Barat et al., 2001b). Fito et al. (2001b) reported an improvement of the drying behavior of several fruits and vegetables submitted to VI pretreatments. Several authors reported that the stability of pigments was enhanced without the use of common compounds for color preservation when VI is applied before drying (Maltini et al., 1991; Torreggiani, 1995). Prothon et al. (2001) reported that water diffusivity of samples was increased when a pre-VI treatment was carried out. On the contrary, the same authors, studying the rehydration capacity of the samples, showed that this property was greater for non-treated samples, probably because both impregnation and DRP reduced the pores in which water flows during rehydration.

Among the stabilization processes, freezing is one of the most important because the use of low temperatures allows to better retain nutritional compounds in comparison with others traditional techniques such as dehydration, pasteurization/sterilization treatments, etc. However the formation of ice crystals leads to several physical damage and drip loss during thawing. Moreover, fluctuation of temperature along the cold chain may promote recrystallization phenomena leading to changes in size and shape of ice as well as their orientation (Zhao & Xie, 2004; Cruz et al., 2009). Vacuum impregnation was shown to be a useful method to incorporate in void phase of foods cryoprotectants and cryostabilizers, such as hypertonic sugar solution, antifreeze protein (AFP), high methoxyl pectin, etc. Martinez-Monzo et al. (1998) showed that as higher the concentration of sugar solution used during VI treatments as lower the freezable water content in food which reduces the drip loss during thawing. The same authors studied the potential application of VI treatment to introduce concentrate grape musts and pectin solution with the aim to

decrease the damages of freezing on apple samples. It was observed that the both aqueous solutions were effective in the reduction of drip loss during thawing. In particular, grape must was able to reduce the freezable water content; instead pectin solution increased glassy transition temperature of liquid phase. Cruz et al. (2009) carried out several experiments to evaluate the potential application of vacuum impregnation with antifreeze protein (AFP) with the aim to preserve the quality of watercress leaves. Results stated that samples treated with AFP showed small ice crystals in comparison with water impregnated samples, due to the ability of the proteins to bound water molecules modifying the natural ice crystal formation. Figures 14a, 14b and 14c show the results of wilting test on watercress leaves. It can be observed that samples impregnated with AFP showed a similar turgidity to the raw samples and higher than the control (water vacuum impregnated samples). This is in agreement with the formation of small ice crystals that during the thawing at room temperature led a reduction of cellular damage. Moreover, it is worth noting that this behavior could produce a better retention of nutrients traditionally lost during thawing. Ralfs et al. (2003) showed that carrot tissues vacuum impregnated with AFP had a higher mechanical strength in comparison with samples submitted to VI with ultrapure water.

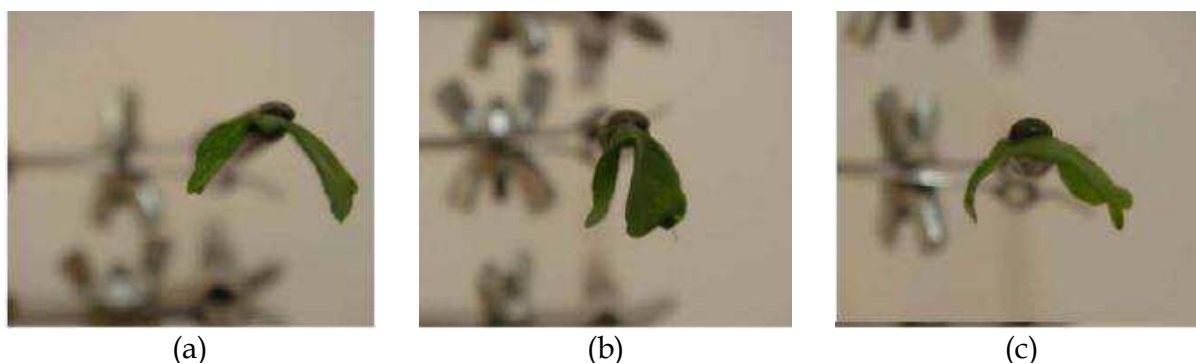


Fig. 14. Wilting test of watercress leaves. (a) raw samples; (b) vacuum impregnated with water; (c) vacuum impregnated with AFP.

Xie and Zhao (2003) showed that vacuum impregnation of strawberries and marionberries with cryoprotectant solutions (HCFS and high methyl pectin) enriched with 7.5% of calcium gluconal highly enhanced the texture and the reduction of drip loss on frozen-thawed samples. In particular the authors reported an increase of compression force in a range of 50-100% and a reduction of drip loss of 20-50% in comparison with untreated samples. According to literature, the authors attributed these results to the reduction of freezable water content. Furthermore, it is worth noting that the reduction of water content as a consequence of VI treatment reduces energy consumption during freezing.

In the last years, on the basis of the consumer interest on the relation between the assumption of correct diet and its health benefits, the possibility to obtain foods with high nutritional and functional properties exponentially increased the efforts of food scientists and industries in this research field. In this way, vacuum impregnation is a useful technique to fill the void phase of foods with nutritional and functional ingredients. However, the scientific results concerning this application of VI are still few. Fito et al. (2001a) were the first scientists that evaluated the feasibility of VI to obtain innovative fresh functional foods (FFF). During their experiments the authors studied the impregnation of

several vegetables with calcium and iron salt solution taking into account the solubility of these salts in water. In particular, the experiments had the aim to estimate the possibility to obtain fortified samples containing the 25% of the recommended daily intake (RDI) for each specific salts. As example, Figure 15 reports the weight of fruits that contain the 25% of RDI as a function of calcium lactate concentration.

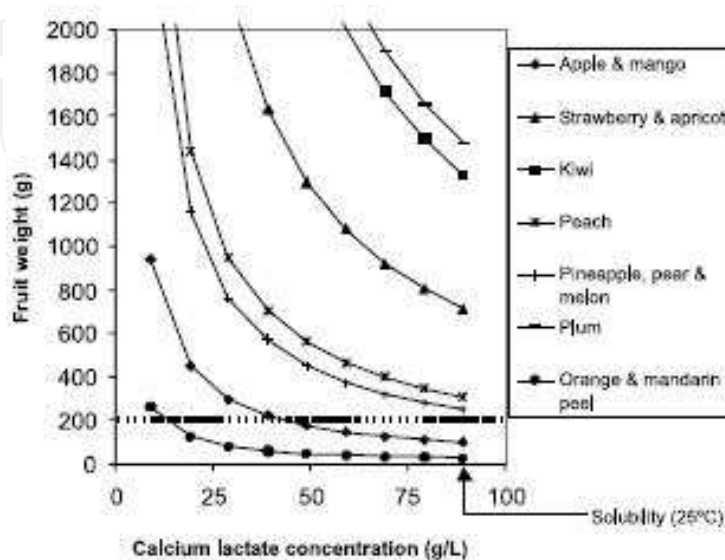


Fig. 15. Weight of some fruits containing the 25% of RDI of calcium salts.

The trends shown in the figure were estimated taking into account the porosity fraction of each type of fruit and by assuming the void phase completely impregnated with the external solution. For instance, by using a calcium lactate solution with a concentration of 50 g/L, less than 100g of orange peel should be consumed to assume the 25% of RDI. In the same way, about 600g and 1200g of peach and strawberry or apricot should be, respectively, assumed. Gras et al. (2003) studied the application of VI with the purpose to obtain eggplant, carrot and oyster mushroom fortified with calcium salts. The authors reported that eggplants and oyster mushroom may be considered as more appropriate to obtain FFF due to their high porosity in comparison with carrots. Hirinoka et al. (2011) studied the application of VI for the enrichment of whole potato with ascorbic acid. Also the authors compared the AA content of samples submitted to VI with the untreated one after a steam cooking treatment over boiling water for 25 minutes and during storage at 5°C for two weeks. Figures 16a, 16b, 16c and 16c show the visual aspect of potato samples submitted to VI and immersed at atmospheric pressure in red ink solution. The figures clearly show that the immersion of potato in solution at atmospheric pressure did not allow to impregnate capillaries of potatoes and the same results were obtained with a VI without apply any restoration time ($t_2 = 0$). Instead after a restoration time of 3 h a significant impregnation was observed.

As expected ascorbic acid content significantly decrease (about 42%) after steam cooking. Nevertheless, VI-cooked samples had an acid ascorbic concentration 22 time higher than samples only submitted to steam cooking (raw-cooking). Also, the VI-cooking samples showed an AA content of ~ 100 mg/100g which is twice of the FAO value (45mg/100g) and close to the RDA value. Furthermore, in another series of experiments Xie & Zhao (2003)

used VI to enrich apple, strawberry and marionberry with calcium and zinc. The experiments performed with high corn syrup solution enriched with calcium and zinc showed that a 15-20% of RDI of calcium more than 40% RDI of zinc could be obtained in 200g of impregnated apple fresh-cut samples.

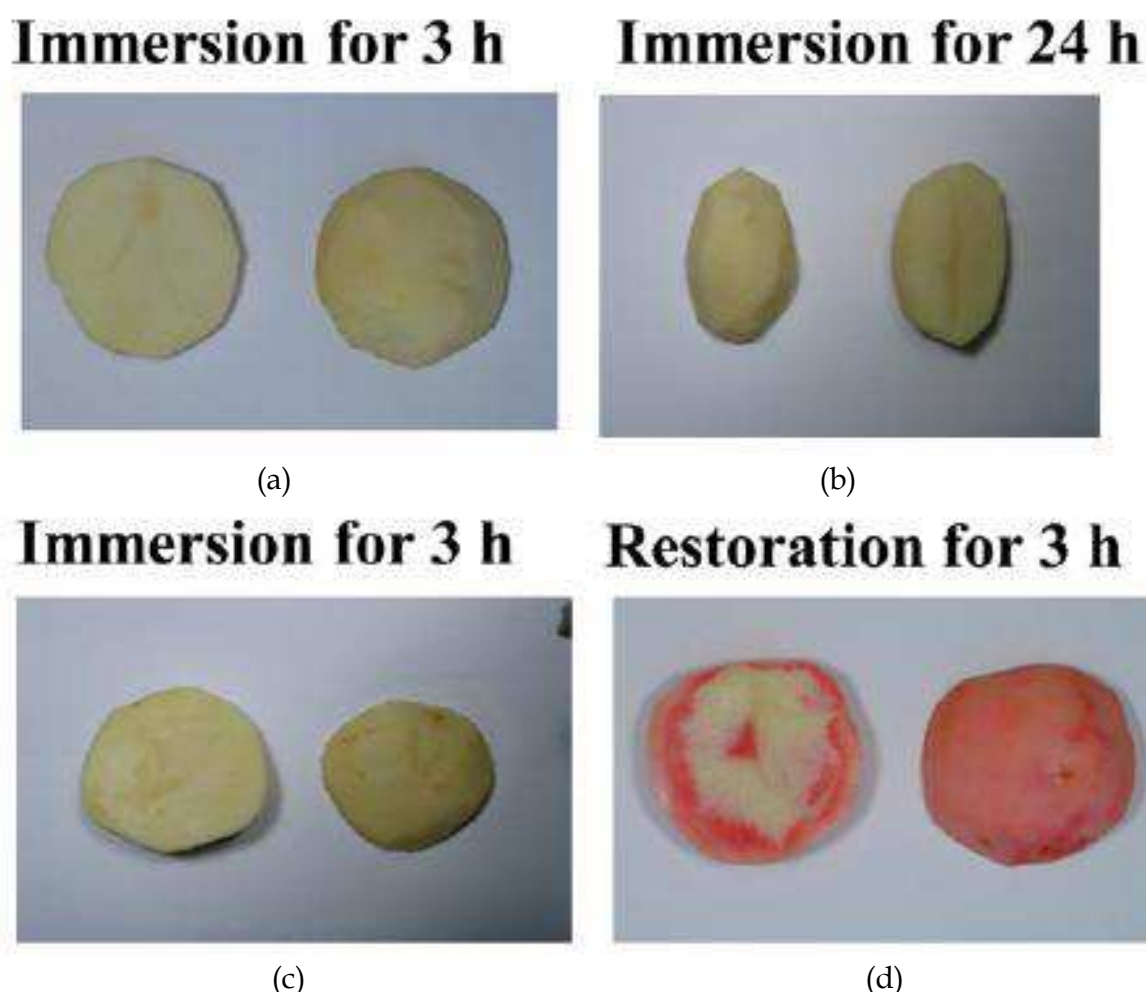


Fig. 16. Potato samples immersed in red ink solution without vacuum (a,b), after a vacuum time of 3 h (without restoration time) and after a restoration time of 3 h (From Hironaka et al., 2011).

Figure 17 reports the ascorbic acid content of whole potato submitted to VI and cooked over boiling water for 25 minutes and the controls (un-VI samples cooked).

Vacuum impregnation could be a method to produce a numerous series of innovative probiotic foods. For instance, Betoret et al. (2003) studied the use of VI to obtain probiotic enriched dried fruits. The authors performed VI treatments on apple samples by using apple juice and whole milk containing respectively *Saccharomyces cerevisiae* and *Lactobacillus casei* (spp. *Rhamnosus*) with a concentration of 10^7 – 10^8 cfu/ml. Results allowed to state that, combining VI and low temperature air dehydration, it was possible to obtain dried apples with a microbial content of 10^6 – 10^7 cfu/g. However, despite the wide number of the potential industrial application, shelf life extension is one of the most important. So, due to its unique advantage vacuum impregnation may be considered a

useful methods to introduce inhibitors for microbial growth and/or chemical degradation reactions; nevertheless, the scientific literature concerning the application of VI in this field of research is still poor. Tapia et al. (1999) used a complex solution containing sucrose (40°Bx), phosphoric acid (0.6% w/w), potassium sorbate (100 ppm) and calcium lactate (0.2%) to increase the shelf life of melon samples. Results showed that foods packed in glass jars and covered with syrup maintained a good acceptance for 15 days at 25°C. Welty-Chanes et al. (1998), studying the feasibility of VI for the production of minimally processed oranges reported that the samples were microbiologically stable and showed good sensorial properties for 50 days when stored at temperature lower than 25°C. Derossi et al. (2010) and Derossi et al. (2011) proposed an innovative vacuum acidification (VA) and pulsed vacuum acidification (PVA) to improve the pH reduction of vegetable, with the aim to assure the inhibition of the out-grow of *Clostridium botulinum* spores in the production of canned food. The results stated the possibility to obtain a fast reduction of pH without the use of high temperature of acid solution as in the case of acidifying-blanching. However, the authors reported the effect of VI on visual aspect of vegetable that need to be considered for the industrial application, because the compression-deformation phenomena could reduce the consumer acceptability. Guillemain et al. (2008) showed the effectiveness of VI for the introduction of pectinmethylesterase which enhances fruit firmness.

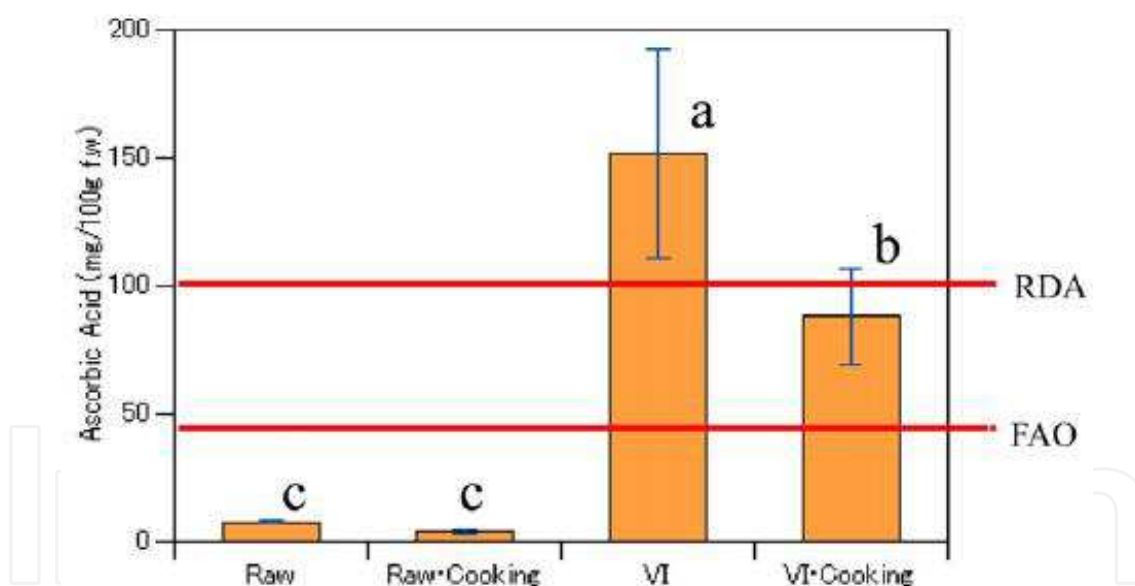


Fig. 17. Effect of steam cooking on ascorbic acid content of whole potato submitted to vacuum impregnation. VI solution: 10% AA, $p = 70$ cm Hg, $t_1=1$ h, $t_2= 3$ h)

5. Conclusion

Although vacuum impregnation was for the first time proposed at least 20 years ago, it may be still considered an emerging technology with high potential applications. Due to its unique characteristics, VI is the first food processing based on the exploitation of three dimensional food microstructure. It is performed by immersing food in an external solution and applying a vacuum pressure (p) for a time (t_1). Then, the restoration of atmospheric

pressure maintaining the foods into the solution for a relaxation time (t_2) allows to complete the process. During these steps three main phenomena occurs: the out-flow of native liquid and gases from the pores; the influx of external solution inside capillaries; deformation-relaxation of solid matrix. The influx of external liquid occurs under the action of a pressure gradient between the pores and the pressure externally imposed; this is known as hydrodynamic mechanisms (HDM). However, on the basis of its nature, VI is a very complex treatment and its results are affected from several external and internal variables. The former are the operative conditions above reported coupled with the temperature and viscosity of external solution. The latter are characterized from the microscopic and mesoscopic properties of food architecture such as length and diameter of pores, their shapes, the tortuosity of internal pathways, the mechanical (viscoelastic) properties of biological tissues, the high or low presence of gas and/or liquid inside capillaries, etc. VI has shown to be very effective in a wide number of industrial applications. The impregnation, causing a significant increase of the external solution/product contact area, is an important method to increase the mass transfer of several solid-liquid operation such as osmotic dehydration, acidification, brining of fish and meat products, etc. VI may be used as pretreatment before drying or freezing, improving the quality of final product and reducing cost operations due to the removal of native liquid (water) from the pores. Furthermore, the possibility to introduce, in a controlled way, an external solution enriched with any type of components catch light on a high number of publications. Indeed, VI has been used to extend shelf life, to produce fresh fortified food (FFF), to enrich food with nutritional/functional ingredients, to reduce the freezing damage, to obtain foods with innovative sensorial properties, to reduce oxidative reaction, to reduce browning, etc. Furthermore, from an engineering point of view some advantages may be considered: 1. it is a fast process (usually it is completed in few minutes); it needs low energy costs; it is performed at room temperature; the external solution may be reused many times. Nevertheless, the applications of VI at industrial scale are still poor. This problem may be attributed to the lack of industrial plants in which it is possible to precise control the operative conditions during the two steps of the process. Also, some technical problems need to be solved. For instance, as reported from Zhao & Xie (2004), the complete immersion of foods into the external solution is a challenge for the correct application of VI. Often, fruits and vegetables tend to float due to their low density in comparison with external solution as in the case of osmotic solution. The current VI is applied by stirring solution with the aim to keep food pieces inside solution with the drawback of an increase of energy costs and possible damages of foods. Furthermore, the lack of information for industries on the advantage of these techniques reduces its application at industrial scale.

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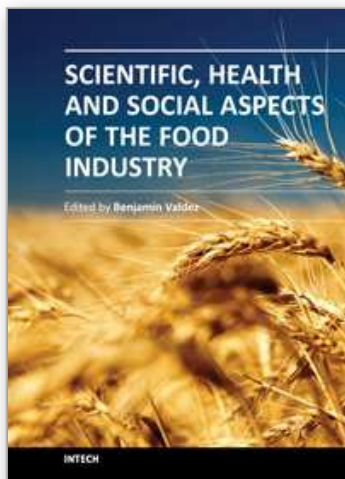
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Scientific, Health and Social Aspects of the Food Industry

Edited by Dr. Benjamin Valdez

ISBN 978-953-307-916-5

Hard cover, 488 pages

Publisher InTech

Published online 01, February, 2012

Published in print edition February, 2012

This book presents the wisdom, knowledge and expertise of the food industry that ensures the supply of food to maintain the health, comfort, and wellbeing of humankind. The global food industry has the largest market: the world population of seven billion people. The book pioneers life-saving innovations and assists in the fight against world hunger and food shortages that threaten human essentials such as water and energy supply. Floods, droughts, fires, storms, climate change, global warming and greenhouse gas emissions can be devastating, altering the environment and, ultimately, the production of foods. Experts from industry and academia, as well as food producers, designers of food processing equipment, and corrosion practitioners have written special chapters for this rich compendium based on their encyclopedic knowledge and practical experience. This is a multi-authored book. The writers, who come from diverse areas of food science and technology, enrich this volume by presenting different approaches and orientations.

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A. Derossi, T. De Pilli and C. Severini (2012). The Application of Vacuum Impregnation Techniques in Food Industry, Scientific, Health and Social Aspects of the Food Industry, Dr. Benjamin Valdez (Ed.), ISBN: 978-953-307-916-5, InTech, Available from: <http://www.intechopen.com/books/scientific-health-and-social-aspects-of-the-food-industry/the-application-of-vacuum-impregnation-techniques-in-food-industry>

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