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Microwave inactivation of red beet (*Beta vulgaris* L. var. conditiva) peroxidase and polyphenoloxidase and the effect of radiation on vegetable tissue quality

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ABSTRACT

The inactivation of polyphenoloxidase (PPO) and peroxidase (POX) in red beet by traditional and microwave (MW) blanching was studied. Microwave heating effects on color and texture were also studied.

Red beet subjected to MW blanching for 5 min at 100–200 W resulted in large weight losses accompanied by a high degree of shrinking. POX was the more heat resistant enzyme. The 90% destruction (*D* value) of the activity of both enzymes could be achieved only at 200 W within the 5 min frame employed for the tests.

When the samples were immersed in water and both the food sample and the water were submitted to MW exposure at 250–450 W or variable power with a maximum at 935 W, shrinking was avoided. The *D* value at 90 °C (reference; $D_{T_{ref}}$) and *z* could be determined after time–temperature corrections, and it was observed that, in general, $D_{T_{ref}}$ values for POX were smaller than for PPO. The microwave treatment (maximum power: 935 W) designed to provide a similar temperature profile to the one observed for traditional blanching (immersion in water at 90 °C), showed the smallest $D_{T_{ref}}$ value for POX inactivation. All treatments reduced elastic characteristics and changed the color of the tissues showing a shift to blue mainly in the case of microwave processes.

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1. Introduction

Preservation treatments in use for fruits and vegetables include traditional technologies (i.e. freezing, dehydration, canning). Other alternative technologies, still under study or just of incipient application (i.e. electric pulses, high pressures, light pulses, microwaves) have begun to be internationally applied on vegetable foods (Geveke and Brunkhorst, 2003;Hayashi, 2002; Tewari and Juneja, 2007). The increasing demand by consumers for minimally processed (fresh-cut) fruits and vegetables has prompted the sale of pre-cut red beet roots as well as other vegetables in trays commercialized in markets or sold in bulk for institutions.

The application of microwave energy on agricultural production includes drying, insect control and seed germination (Venkatesh and Raghavan, 2004). In the food processing area, applications such as tempering, vacuum drying, freeze drying, dehydration, cooking, blanching, baking, roasting, pasteurization, sterilization and extraction are increasingly being perfected (Orsat et al., 2005). Microwave use has been studied for the blanching of vegetable tissues (Andres et al., 2004; Roberts et al., 2004), observing that the technique was useful for enzyme inactivation but more research is needed concerning its effect on nutrients, texture and color of fruits and vegetables.

Red beet (*Beta vulgaris* L. var. conditiva) is a traditional and popular vegetable in many parts of the world. It is especially rich in fibers as well as in sugars but with moderate caloric value. It has an important content of B-vitamins (B1, B2, B3 and B6) as well as folic acid. Red beet roots are consumed either fresh or after thermal processing or fermentation. The soluble and cell wall associated phenolics as well as betalains, the main pigments in red beet responsible for its reddish-purple hue, are bioactive compounds (Schwartz et al., 1980), due to their antioxidant capacity that is beneficial for human health (Kanner et al., 2001; Gliszczynska-Swiglo, 2006).

The enzymatic activity of vegetable tissues is one of the principal causes of food nutritional and organoleptical impairment. In the oxidative degradation of the phenolic compounds there are two relevant enzymes involved whose activity can lead to the production of brown polymers. These enzymes are the polyphenoloxidase (PPO) and peroxidase (POX) (Tomas-Barberán and Espin, 2001). The POX (sysname: donor hydrogen-peroxide oxidoreductase; EC 1.11.1.7) is one of the most heat-stable enzymes and as such it is often used as a marker for the adequacy of the blanching process (Barret and Theerakulkait, 1995;Hemeda and Klein, 1990; Flick et al.,

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1978). Tyrosinase or PPO (sysname: monophenol L-dopa:oxygen oxidoreductase; EC 1.14.18.1; PPO) is the key enzyme in melanin biosynthesis and in the enzymatic browning of fruits and vegetables. The role of PPO in the secondary metabolism of plants still remains unclear, but its implication in betalain biosynthesis has been proposed. PPO is a copper enzyme that catalyzes two different reactions using molecular oxygen: the hydroxylation of monophenols to o-diphenols (monophenolase activity) and the oxidation of the o-diphenols to o-quinones (diphenolase activity; Sanchez-Ferrer et al., 1995). This enzyme is widely distributed in plants, microorganisms, and animals where tyrosinase is responsible for melanization. Many studies (Richard-Forget and Gauillard, 1997; Subramanian et al., 1999) have demonstrated that while the PPO develops an oxidative action on the substrate, it gives origin to H_2O_2. Quinones are formed by oxidation and H_2O_2 is used by POX.

Tomas-Barberán and Espin (2001) suggested that one of the major concerns in the food industry is to prevent the development of enzymatic browning prior to or during the processing of fruit and vegetables, because formed products generate organoleptical alterations that preclude consumer acceptance of the product. The primary goal of blanching is the inactivation of undesirable enzymes present in fruits and vegetables and the reduction of the microbial load; at the same time, it aids in removing tissue gases, stabilizing texture, color, flavor and nutritional quality of products (Williams et al., 1986).The blanching process usually involves exposing plant tissue to some form of heat, for example through the use of steam or hot water (Barret and Theerakulkait, 1995). Inactivation of POX and PPO enzymes is often used to determine when the vegetable has been satisfactorily blanched considering the high resistance of both enzymes to thermal processing (Collins and McCarty, 1969; Vural Gökmen et al., 2005).

According to Chen et al. (1971), microwave energy may be used to blanch vegetable tissues as an alternative to conventional steam and water blanching. Microwave heating takes place in dielectric materials such as in foods due to the polarization effect of electromagnetic radiation at frequencies between 300 MHz and 300 GHz. Two frequency bands are allocated in the USA by the Federal Communications Commissions (FCC) for industrial, scientific, and medical applications. The 915 MHz band is used for industrial heating only, and the 2450 MHz band is used both in the industry and in the domestic microwave oven. Dielectric properties of foods play a critical role in determining the interaction between the electric field and the foods, and they are dependent on composition, temperature and microwave frequency. Hence, there is an important influence of the dielectric properties of the material on the efficiency of electromagnetic energy coupled into the materials, electromagnetic field distribution and conversion of electromagnetic energy into thermal energy within material (Tang, 2005). Matrix must contain dipolar or ionic species to enable heating to occur. The heating occurs via dipolar polarization and conduction.

Several researchers have studied the application of microwave radiation for blanching or enzyme inactivation (Quenzer and Burns, 1981; Kermasha et al., 1993a,b; Ramaswamy and Fakhouri, 1998; Begum and Brewer, 2001; Brewer and Begum, 2003; Roberts et al., 2004; Lin and Brewer, 2005; Lin and Ramaswamy, 2011) showing its effectiveness and suitability. From the results of Ramaswamy and Fakhouri (1998) and Ramesh et al. (2002) it is possible to observe that vegetable tissue blanched with microwave radiation retained better its nutritional value. Begum and Brewer (1996) reported that microwave blanched asparagus kept its nutritional value, taste and texture, as well as and often better than, asparagus blanched using traditional methods. In many studies reported, the time–temperature and time-power history of the samples was not properly monitored.

Venkatesh and Raghavan (2004) emphasized in their studies the microwave limitations that include the fact that, as materials are being processed, they often undergo physical and structural transformations that affect their dielectric properties. Thus, the ability of microwaves to generate thermal energy varies during processing. Sharp transformations in the ability of the microwave to generate heat can affect process success. An inherent problem associated with microwave heating is the non-uniformity of heating caused by an uneven spatial distribution of electromagnetic field inside the cavity (Vadivambal and Jayas, 2007). The success of microwave blanching depends mostly on raw material quality, timing and wattage of the process.

Absorption of microwave energy by plant tissue results in chemical and physical changes. The intermolecular friction from microwave heating may cause internal cell pressure leading to rupture resulting in a loss of cell content and organization (Quenzer and Burns, 1981). For that reason it is necessary to perform specific studies for different tissues, trying to state a systematic background of information that allows to find the optimum conditions (i.e. time, power level) that permit to attain the goal of the process with minor nutritional and organoleptical damage.

The aim of this work was to evaluate the characteristics of PPO and POX inactivation in red beet by traditional and microwave blanching. The evaluation of color and texture of the samples was also performed.

2. Materials and methods

2.1. Chemicals

They were of analytical grade unless stated.

2.2. Sample preparation and blanching process

Red beet (*B. vulgaris* L. var. conditiva) roots harvested in Argentina were purchased at the local market. They were carefully cleaned, peeled and cut into 10 mm-thick slices perpendicular to their longitudinal axis. Cylindrical samples with a diameter of 15 mm were then obtained from each slice at about 7–10 mm distance from the periphery, by using a cork borer. This sample geometry was chosen by considering its adequacy for mechanical assays.

2.3. Microwave blanching

Batch microwave processing was carried out in a microwave system ETHOS Plus (Milestone Srl, Sorisole, Italy) with a magnetron of 2450 MHz. The microwave used the ATC-400 system for continuous monitoring and control of the internal temperature. The optical sensor used was fitted in a teflon coated ceramic thermowell. The samples were distributed in six glass vessels which were placed inside a polypropylene container of cylindrical shape having a diameter of 27.9 cm diameter and a height of 19.7 cm (Fig. 1). A 360° alternate movement was programed for the container in the microwave cavity to avoid bending of the sensor con-

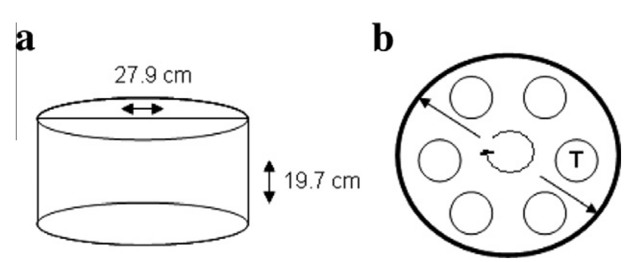

Fig. 1. Scheme of the cylindrical polyprolylene container used into the microwave system (a) and of its upper view (b) which shows the localization of the six vessels. T: means the initial localization of the temperature sensor.

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