Engineering Aspects of Pulsed Electroplasmolysis of Vegetable Tissues

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Abstract

The increasing interest in pulsed electric field (PEF) treatment lies in its potential for inducing non-thermal electroplasmolysis of cellular materials resulting in increased cell permeability. Electropermeabilization of vegetable tissues increases electrical conductivity, diffusivity, heat and mass transfer coefficients. It decreases compressibility and failure strength of the biological tissues. These effects of electropermeabilization are exploited in developing novel processing operations. PEF treatment has been reported to enhance extraction and dehydration processes in vegetable tissues. The choice of suitable PEF parameters is determined by application, technological, energy consumption and economical considerations. The engineering design aspects of the processes continue to be a challenge to industrial application of the techniques. New processes involving PEF either as stand-alone or in synergistic combination with other processes are envisaged as a result of the increased consumer demand for minimal processed, high quality food products.

Keywords: *Pulsed electric field, electroplasmolysis, electropermeabilization, electroporation, nonthermal method, emerging technology*

Introduction

Fruits and vegetable tissues contain the main nutrients needed for balanced human nutrition. Most food processing technologies often involve different extraction and dehydration operations requiring the separation of liquid and solids from the vegetable tissues (Potter & Hotchkiss, 1995). Plant cell walls normally require disruption (plasmolysis) before the liquid components can be separated and thus some form of pre-treatment is often required (Brennan et al., 1990). Among the different available pre-treatment methods such as freezing, enzymatic treatment, alkaline breakage and others, heating is the most widely used. However, due to thermal degradation of the nutrients and high energy consumption level during heating, new techniques for plasmolysis are being developed.

Electroplasmolysis of fruits and vegetables has been reported since the late forties (Flaumenbaum, 1949; Zagorulko, 1957). It is the increased permeability of biological tissue cells after electric field application. Recently, this phenomenon was also called electropermeabilization (Teissie, 1999; Angersbach et al, 2002). The microstructure of plant and animal tissues changes considerably during electrical treatment as a result of contraction and gaping of cell vacuoles (Gudmundsson and Hafsteinsson, 2001; Fincan and Dejmek, 2002). Since electrical conductivity of intercellular juice is significantly higher than the

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conductivity for plasmatic membranes that covers the cells, up to 95% of the voltage applied across tissue samples drop at the cellular membranes (Zagorulko, 1958; Lebovka *et al.*, 2000b).

Transmembrane potential of a spherical cellular membrane u_m is determined by the following equation (Zimmermann et al, 1976):

$$u_m = 0.75 d_c E \cos\Theta \tag{1}$$

where E is the electric field strength in the whole tissue, d_c is the cell mean diameter, and Θ is the angle between electric field direction and radius-vector on the cell surface. Following Eq. (1), the maximal u_m value is achieved at the cell poles.

Electric field E_m in the cell membrane with thickness d_m is expressed as:

$$E_m = \frac{u_m}{d_m} \tag{2}$$

Considering that the mean size d_c of a plant cell size is between 10^{-5} and 10^{-4} m (Avers, 1986; Jackman & Stanley, 1995) and considering also that the plasmalemma membrane thickness d_m is about 10^{-8} m (Dyson, 1974), it can be deduced that electric field strength in plant cell membranes is increased by about 10^3 - 10^4 times compared to the electric field strength in the media. This is the basis of electroplasmolysis operation.

Since plant cells are typically larger than microbial cells with mean sizes between 10 nm and 1 µm (Rogers et al., 1980), electropermeabilization of vegetable tissues can take place at relatively low electric fields. Gulyi et al. (1994) indicated that electroplasmolysis in sugar beet initiated at 50 V/cm whereas electropermeabilization of microbial cells is reported to occur between 10 and 50 kV/cm (Barbosa-Canovas et al., 2000). The purpose of this paper is to review engineering aspects of food electroplasmolysis and identify current trend of developments in this area.

Mechanism of electroplasmolysis

The mechanism of plant tissue electropermeabilization is not yet well understood. There are different proposals on the mechanisms for membrane rupture under electric fields. These include electromechanical compression (Crowley, 1973; Zimmermann, 1986), electroosmotic stress (Dimitrov and Sowers, 1990), electrochemical changes in cell molecular structure (Sabri et al., 1996), viscoelastic deformation of cell vesicles (Winterhalter & Helfrich, 1988; Lebovka *et al.*, 1991; Sukhorukov *et al.*, 1998), electroporation in cell membrane (Neumann *et al.*, 1992; Prasanna & Panda, 1997; Pawlowski *et al.*, 1998) and other mechanisms (Weaver & Chizmadzhev, 1996). In all the reported applications of electric fields, tissue temperature has been increased only a few degrees indicating that tissue electropermeabilization is largely a nonthermal process. The most widely accepted mechanism of biological membrane electropermeabilization is the formation and growth of pores with subsequent molecular exchange and unbalance of natural osmotic equilibrium between internal and extracellular volumes (Weaver & Chizmadzhev, 1996; Sundaram & Stebe, 1997).

Electroplasmolysis influences electrical, thermal, diffusion and rheological properties of vegetable tissues. These properties can be used to monitor or evaluate the degree of electroplasmolysis during pulsed electric field (PEF) treatment. Changes in some properties of vegetable tissues as a result of PEF treatment as reported by different authors are shown in Table 1. In general, electropermeabilization increases porosity, electrical conductivity, diffusion, heat and mass transfer coefficients. However, it decreases compressibility and failure stress of plant tissues. Attempts to exploit the influence of PEF and electroplasmolysis on vegetable tissues have increased only within the past decade. Application of PEF to intensify juice pressing (Bazhal & Vorobiev, 2000; Knorr *et al.*, 2001), diffusion (Jemai *et al.*, 2000; Knorr *et al.*, 2001), osmotic dehydration (Rastogi *et al.*, 2000a; Taiwo *et al.*, 2002), and drying (Ade-Omowaye *et al.*, 2001) have been reported in the literature. Some pilot and semi-industrial equipment for electrical treatment of vegetable materials have been developed. However, some unresolved engineering and technological aspects of the treatment systems have limited wide industrial application of the techniques.

Table 1. Vegetable tissue properties estimated for control and PEF plasmolysed samples.

Material	Parameter	Value		Process	Reference
		Control	PEF treatment		
Apple	Electrical conductivity (S/m)	0.003-0.007	0.035-0.070	PEF treatment	Lebovka *et al.* (2001)
Carrot	Electrical conductivity (S/m)	0.03	0.41	PEF treatment	Rastogi *et al.* (1999)
Potato	Electrical conductivity (S/m)	0.06	0.53	PEF treatment	Knorr & Angersbach (1998)
Apple	Porosity (%)	67	75		Bazhal *et al.* (2003b)
Carrot	Liquid diffusion coefficient (m^2/s)	0.98×10^{-9}	1.55×10^{-9}	osmotic dehydration	Rastogi *et al.* (1999)
Sugar beet	Sugar diffusion coefficient (m^2/s)	0.68×10^{-9}	1.2×10^{-9}	diffusion	Gulyi *et al.* (1994)
Paprika	Mass transfer coefficient ($kg/m^2\ s$)	0.043	0.058	drying	Ade-Omowae *et al.* (2001)
Paprika	Constant drying rate ($kg/m^2\ s$)	9.68×10^{-4}	13.02×10^{-4}	drying	Ade-Omowae *et al.* (2001)
Paprika	Heat transfer coefficient ($W/m^2\ s$)	73.13	98.36	drying	Ade-Omowae *et al.* (2001)
Sugar beet	Elastic modulus (MPa)	12.5	6.5	compression test	Matvienko (1996)
Apple	Elastic modulus (MPa)	1.53	0.32	compression test	Bazhal *et al.* (2003b)
Apple	Failure stress (MPa)	1.26	0.53	compression test	Bazhal *et al.* (2003b)

PEF waveforms and system design

Electric field waveform is an important design consideration for a PEF application. Biological cells respond differently to different electric field waveforms. Different types of waveform have been used for electroporation and disruption of biological cells (Zhang *et al.*, 1995; Jemai *et al.*, 2000; Bazhal, 2001). These include DC, AC and different pulse waveforms as illustrated in Fig. 1. Earlier works on electroplasmolysis were by using AC and DC waveforms (Lazarenko *et al.*, 1977, Papchenko *et al.,* 1988a, 1988b, McLellan *et al.*, 1991). The use of these waveforms is restricted by the inherent practical difficulties involved in control of energy input for short duration ranging from microseconds to seconds during treatment. The exponential decay and rectangular pulses are widely used in most PEF applications (Barbosa-Canovas *et al.*, 2000). From the point of view of current polarity, all electric fields used for electroplasmolysis can be classified as either monopolar or bipolar. Gulyi *et al.* (1994) reported that juice purity (percent ratio of sugar in the total dry solids of juice) was increased by 1 - 2% after bipolar electroplasmolysis and by 3 - 5% after monopolar electrical treatment in comparison with traditional thermal plasmolysis. Bazhal & Gulyi (1983) explained this difference as due to polarization of the tissue during monopolar electroplasmolysis, electrical coagulation of proteins and colloids that increased retention of nonsugar substances in the treated tissue. Zhang *et al.* (1995) also indicated that monopolar pulses maximized polar deposit of charged molecules. Therefore the pulse characteristics has effect on the quality of processed product (Kupchik, 1991).

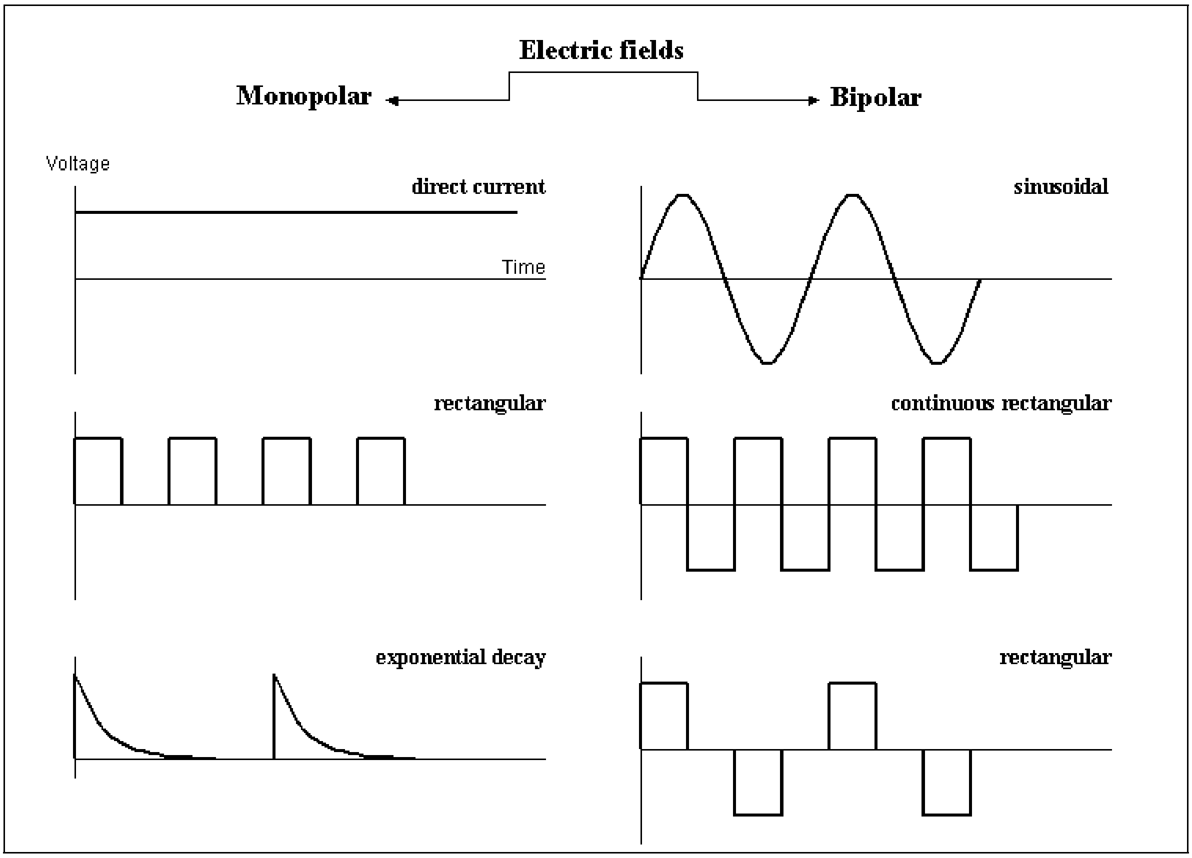

Fig. 1. Different types of electric field waveforms.

Design of PEF systems depends on desired pulse characteristics and applications (Gongora-Nieto *et al.*, 2000). Generally, a PEF system includes a high voltage DC generator to supply electrical energy; bank of capacitors to store the energy generated by the generator; a high voltage switch to deliver energy to electrodes and a treatment cell to hold and contain samples. The generation of electric pulses across a treatment cell requires rapid (in the order of nanoseconds) charging and subsequent discharging of electrical energy stored in the capacitors. Exponential decay pulses are easier to generate due to the simplicity of their circuits involving a bank of capacitors connected in series with a charging resistor (Barbosa-Canovas *et al.*, 2000). The energy stored in the capacitor is then rapidly switched across a treatment chamber. The exponential waveforms are generated independent of the electrical resistance of the load. Rectangular pulse waveforms are more difficult to generate and normally require a pulse-forming network consisting of an array of capacitors, inductors and switching device. Effective generation of rectangular or square pulses depend not only on the generator but also on the material to be treated and circuit impedance (Grahl & Markl, 1996; Angersbach *et al.*, 2002). Rectangular type pulses can be transformed to exponential decay type if the characteristic time for electrical discharge through a sample is less than the time constant for charging the capacitors (Grahl & Markl, 1996). High electrical power accumulation in the capacitors is needed to support rectangular shape of the applied pulses. This may present engineering design difficulties in some applications.

Rectangular pulses are more efficient for plasmolysis compared to either exponential or oscillatory pulses (Knorr *et al.*, 1994; Qin *et al.*, 1996). This was attributed to the wider pulse width at higher electric fields obtained using square pulses. Square pulses maintain high electric fields with very high rise and fall times whereas exponential pulses have long tail of relatively low voltage. Thus electrical energy is dissipated more effectively with rectangular pulses than with exponential ones. Bipolar pulses have been reported to be more efficient than monopolar waveforms for cellular rupture (Kinosita *et al.*, 1992; Bazhal, 2001). The effectiveness of bipolar pulses is attributed to elimination of the asymmetric properties of cells under bi-polar pulses (Saulis & Venslauskas, 1988). Monopolar pulses should be used when maintenance of juice quality is the prime factor. However, the maximal effectiveness of electropermeabilization can be achieved by the use of bipolar pulses. Thus, the choice of electric field pulse waveform parameters is determined by intended application, technological and economical requirements.

A number of devices have been developed for electroplasmolysis of fruits and vegetables. These electroplasmolyzers are either batch or continuous operation units. Intensity of electrical treatment is controlled by the rate of material passage through the continuous plasmolyzer or by the exposition time under the electric field applied to material in a batch unit. Roller electroplasmolyzers (consisting of rotating electrodes) have been used to achieve crushing, slicing, grinding, rolling and simultaneous electroplasmolysis of vegetable tissues (Chebanu e*t al.*, 1980a, 1980b, Lazarenko *et al.,* 1977; Kogan, 1958). Electrical treatment may also be applied as materials are transported either between sets of roller electrodes or between roller and stator electrodes (Papchenko *et al.*, 1988a, Gulyi *et al.*, 1994, Vorobiev *et al.*, 2000). There have been attempts to combine pressure with electroplasmolysis. This apparently has positive potential application in the juice industry. Different systems that combined pressure and electrical treatment of materials have been reported. Scheglov *et al.* (1969) used a screw press-electroplasmolyzer consisting of a thick-walled cylinder housing an inner rotating screw with a gradually decreasing pitch. The central part of the screw and cylinder served as coaxial electrodes to electrically treat materials as they traverse through the screw press. Other combined pressure and electroplasmolysis designs include plate press electroplasmolyzers in which the material is pressed between 2 plate electrodes (Matov and Reshetko, 1968; Lazarenko et al., 1977; Lebovka et al., 2000a, 2001). Overall juice expression from a vegetable tissue is enhanced when electric field was applied after an initial pressing stage (Bazhal *et al.,* 2001). Synergistic effect between pressure and PEF treatment during juice expression has been reported (Bazhal & Vorobiev, 2000).

Energy consideration

There is an energy saturation threshold, beyond which further energy input has no influence on the PEF application and thus reduce the treatment efficiency (Lebar *et al.*, 1998; Wouters *et al.*, 2001). Maximum plasmolysis rate was reported at energy input of 3-5 kJ/kg for apple tissue (Bazhal & Vorobiev, 2000), 14-16 kJ/kg for potato samples (Knorr & Angersbach, 1998) and 60-70 kJ/kg for sugar beet samples (Bazhal, 1998). In general, the maximum energy input efficiency was observed during the first phase of electrical treatment when degree of plasmolysis was less than 50% (Bazhal, 1998; Bazhal *et al.*, 2003a). Since specific heat of apple is about 3.85 kJ/(kg K) (Batty & Folkman, 1983) and 3.5 kJ/(kg K) for sugar beet (Gulyi *et al.*, 1994), the increase in temperature during PEF treatment may be estimated to be in the range of 20°C.

Energy input density during one electrical pulse application is given by:

$$q_i = s\bar{\sigma}_i t_i E^2 \qquad (3)$$

where $\bar{\sigma}_i$ is the average electrical conductivity of the sample during a pulse application, t_i is a pulse width and s is a factor determined by the pulse shape; for rectangular and exponential pulses s=1 and s=0.5, respectively (Zhang *et al.*, 1995).

Energy consumption during PEF treatment with n pulses is determined using the equation:

$$Q = nq_i \approx ns\bar{\sigma}t_i E^2 = s\bar{\sigma}tE^2 \qquad (4)$$

where $\bar{\sigma}$ is the average electrical conductivity of the sample during electrical treatment, $t=nt_i$ is the PEF application time. Flaumenbaum & Kazandzhyi (1966) proposed the coefficient $K=tE^2$ for estimation of biological tissue resistance to electric field. The recent investigations prove that treatment time t depends on the electric field E (Lebovka *et al.*, 2002). Energy input can be estimated by the factor of $\tau(E)E^2$, where τ is the characteristic electroplasmolysis time.

Lebovka *et al.* (2002) established a general exponential dependency between τ and E in the form of Equation 5 for different experimental data obtained for various vegetable tissues:

$$\tau = A\exp\frac{B}{C + DE^f} \qquad (5)$$

where A, B, C, D and f are fitted parameters. The authors suggested that the f factor varies from 0.50 to 0.56. The higher the electric field strength, the less time needed for achieving the same degree of plasmolysis. However, increasing the electric field value increases energy consumption in the square-law dependency on electric field strength. The optimal electric field value that can be estimated by minimizing the function τE^2, depends on the type of tissue. Bazhal *et al.* (2003b) indicated that the optimal electric field strength falls in the range between 200 and 1100 V/cm for most vegetable tissues, whereas characteristic treatment time varies from 2.5 to 0.3 ms, respectively.

Optimal electric field strength maybe attained by increasing applied voltage or by decreasing the distance between electrodes. Low processing capacity is an inherent practical problem associated with reducing electrode gap. Also, very small electrode gap may provoke sparking and complicate control of PEF plasmolysis in industrial applications. On the other hand, increase in applied voltage is limited by increase in design complexity, cost of pulse generator and industrial security requirements.

Power consumption, w, per volume unit of the treated material can be expressed as:

$$w = \frac{q_i}{t_i} = s\bar{\sigma}E^2 \qquad (4)$$

The power delivered by a pulse generator to a given material is in the square-low dependency from electric field strength and linearly proportional to volume of the treated material. Maximum power consumption is observed at the end of electroplasmolysis operation because in contrast to electrical treatment of liquid products, electropermeabilization of solid foods can increase tissue electrical conductivity up to more than ten times (see Table 1). Although higher electric field strength should decrease treatment time, it elevates power consumption and results in higher cost of electrical generators. For PEF treatment with energy input density q_i≈3 J/kg per pulse and pulse width $t_i = 10^{-4}$ s, power consumption yields w≈30 kW/kg (Bazhal & Vorobiev, 2000). Despite the low energy density input, high power consumption limits industrial application of high electric field pulses due to the required high processing capacity.

Further development in industrial electroplasmolysis will include search for effective means of reducing electric field strength. One of the emerging and promising approaches is the using of combined treatment, which has shown positive potential. Several combinations of PEF with other technologies such as ultrasound, high pressure, pH, etc. show advantages over individual applications. Combined treatments reveal additive or synergistic effects (Gulyi *et al.*, 1994; Rastogi *et al.*, 2000b; Knorr *et al.*, 2001; Bazhal et al., 2001). Only little studies have been conducted on combine plasmolysis. Also, the mechanisms of the synergistic effects are not well understood. The major problem arising from combined treatment is the determination of critical process factors and the choice of optimal modes of treatment.

Conclusion

Electropermeabilization hold good promise for enhancement of some processes during vegetable processing. The current increased demand for minimal thermally processed products will encourage development of novel non-thermal technologies such as PEF. Effective application of electroplasmolysis will require adequate understanding and control of pertinent parameters related to the PEF system intended for the process. Despite progress on PEF technologies, further work will be needed to improve electroplasmolysis efficiency, decrease of electric fields, treatment time, energy requirements, and to exploit the advantages of synergistic combined treatment with other technologies.

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