PULSED ELECTRIC FIELDS IN WINE PROCESSING – A NARRATIVE REVIEW

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Abstract: Non-thermal technologies have emerged to meet the demand for safe and high-quality foods. Among these, the pulsed electric field stands out for its eco-friendly and cost-effective characteristics, particularly in the microbial inactivation and extraction of intracellular compounds from different vegetable matrices. It involves intermittent high-voltage pulses that increase the permeability of biological cell membranes through reversible or irreversible electroporation. Reversible electroporation is valuable in biotechnology, medicine, and molecular biology, while irreversible electroporation is especially important in maintaining food safety and quality. The pulsed electric field is notably effective in wine processing, facilitating must or wine decontamination and the extracting of bioactive intracellular compounds from grapes. This extraction significantly impacts wine's sensory quality and stability. The technology aids in controlling spoilage microorganisms, thereby preventing wine deterioration and reducing economic losses for manufacturers. The article synthesizes scientific literature to explore the pulsed electric field's antimicrobial and extraction mechanisms, emphasizing its advantages in winemaking.

Keywords: pulsed electric field, electroporation, winemaking optimization

Introduction

During the last few decades, numerous non-thermal technologies have been developed in response to the growing demand for safe and high-quality foods (Nowosad *et al.*, 2021, Ozturk and Anli, 2017; Stoica *et al.*, 2013, 2014). Among these innovative technologies, Pulsed Electric Field (PEF) technology is an eco-friendly and cost-effective approach for microbial inactivation

and enhancement of mass transfer in foods (Chatzimitakos et al., 2023). PEF technology utilizes intermittent high-voltage pulses, ranging from 0.1 to 50 kV/cm or higher (depending on the electric potential difference between the electrodes), which increase the permeability of biological cell membranes by forming hydrophilic pores (Bocker and Silva, 2022; Lytras et al., 2024; Stoica et al., 2022). This phenomenon, known as electroporation, can be either reversible (membrane is able to recover) or irreversible (membrane is unable to recover from pore formation), and it can be controlled based on specific applications (Napotnik et al., 2021; Mihalcea and Stoica, 2023). Reversible electroporation (characterized by pore resealing) has applications in biotechnology, medicine, and molecular biology (Bălănică Dragomir et al., 2020; Ding et al., 2023; Peng et al., 2024). In contrast, irreversible electroporation is applied in vegetable processing, primarily for cell disintegration and tissue permeabilization (Bălănică Dragomir et al., 2020). PEF is highly suitable for wine processing (Akdemir Evrendilek, 2022; Silva et al., 2024). It can be used for must or wine decontamination through the electroporation of spoilage microorganisms (which is critically important for controlling wine deterioration and mitigating economic losses for wine manufacturers), as well as for the extraction of intracellular compounds from grapes (e.g. extraction of phenolic compounds has a profound impact on both the sensory quality and wine stability (Barba et al., 2015; Bocker and Silva, 2022; Carpentieri et al., 2023; Gałązka-Czarnecka et al., 2018; Maza et al., 2020; Ozturk and Anli, 2017; Puértolas et al., 2010; Silva et al., 2024). This article offers an insight into antimicrobial and extraction mechanisms of pulsed electric fields, highlighting their significant advantages in the winemaking process. The discussion is based on the synthesis of scientific literature, creativity, and knowledge in the field of non-thermal food processing.

Principles of PEF technology

One of the key principles of PEF technology is its ability to inactivate microorganisms and enzymes in food products without the need for high temperatures, making it an attractive alternative to traditional thermal processing methods. This non-thermal nature of PEF ensures that the nutritional and sensory attributes of the food are better preserved, as compared to conventional heat treatments. The technology has been successfully applied to various food products (fruit juices, dairy products, meat, wine) (Adhipaththu *et al.*, 2024; Akdemir Evrendilek, 2022; Akdemir Evrendilek and Hitit Özkan, 2024; Silva *et al.*, 2024). PEF

technology also shows promise as a pretreatment for enhancing the extraction of valuable compounds from food materials. For instance, PEF has been effectively used to increase the yield of polyphenols and other bioactive compounds from plant materials, thereby improving the efficiency and sustainability of extraction processes in the food industry. This is particularly beneficial for producing extracts with higher concentrations of bioactive compounds, which are in demand for their health-promoting properties (Athanasiadis *et al.*, 2023; Ranjha *et al.*, 2021). The application of PEF in food processing is not limited to microbial inactivation but also extends to improving the functional properties of food, such as enhancing the accumulation of nutrients like vitamin C in yeast cells used in food products (Nowosad *et al.*, 2022).

PEF – Microbial Inactivation Mechanism

The inactivation of biological cells (both eukaryotic and prokaryotic) by an external electric field (EF) is a dynamic multi-phase process. This process includes increasing the cell transmembrane potential, initiation phase of pore formation, population pore evolution, and post-treatment phase where either pore resealing occurs or the cell plasma membrane fails to recover the pores (Bălănică Dragomir et al., 2020; Ding et al., 2023; Peng et al., 2024). When the biological cells are exposed to EF, membrane stability changes, causing ions to migrate toward the cell plasma membrane (Bocker and Silva, 2022; Lytras et al., 2024). This migration results in the accumulation of free charges with opposite signs along the outer and inner sides of the membrane, which attract each other, inducing electrocompression that replaces the membrane's elastic resistance. The accumulation of free charges on the plasma membrane generates an initial transmembrane potential (TP). The application of EF generates an induced potential (IP), which combines with TP. This interaction causes the TP to reach a critical threshold, referred to as the breakdown potential (BP) (Bălănică Dragomir et al., 2020). When this threshold is reached, it leads to the formation of pores/nanopores, particularly in the context of nanosecond PEF (electroporation) (Bocker and Silva, 2022; Lytras et al., 2024; Ruiz-Fernández et al., 2022). Electroporation can range from the enlargement of pre-existing pores to the initiation of new pores, which may be temporary or permanent, and can also affect intracellular structures. Specifically, membrane rupture occurs at the locations where the TP and IP reach the BP. The cell plasma membrane can be disrupted due to Joule heating of the membrane surface or chemical imbalances caused by enhanced transmembrane transport through the membrane pores.

The significant transmembrane potential exerts pressure on the plasma membrane, resulting in reduced membrane thickness, compromised membrane integrity, and eventual cell rupture (Bălănică Dragomir *et al.*, 2020). The electrical breakdown resulting from PEF can be either reversible (allowing the cells to survive) or irreversible, resulting in the efflux of small compounds from the cells (the cells cannot repair the damage or restore metabolism, leading to cells death) (Ding *et al.*, 2023; Peng *et al.*, 2024) (Figure 1).

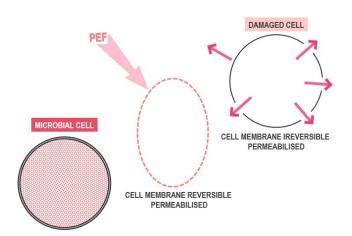


Figure 1. Mechanism of cellular inactivation by PEF. Schematic presentation (adapted from FAQ about Pulsed Electric Field Processing, 2024; Stoica *et al.*, 2011).

The mechanisms of cell death induced by electroporation are still not well understood (Peng et al., 2024).

PEF – Extraction Mechanism

This technique is proposed to reduce the mass transfer resistances of target solutes and solvents through the cell envelope, including the membrane and wall. Specifically, pulpy plant tissues (pericarp/mesocarp) are subjected to repetitive short pulses (ranging from 1µs to 1ms) of moderate electric fields (0.1–10 kV/cm). In contrast, hard vegetable materials (such as seeds) are exposed to electric fields up to 20 kV/cm (Mihalcea and Stoica, 2023). These conditions increase membrane permeability by forming hydrophilic pores in the lipid bilayer due to the induced transmembrane voltage (electroporation) and chemical changes of lipids and alterations of the membrane's protein functions (Bocker and Silva, 2022; Lytras *et al.*, 2024). This process facilitates tissue softening and enhances the extraction of intracellular compounds (e.g. anthocyanins, polyphenols and resveratrol from grapes) (Barba *et al.*, 2015; Bălănică Dragomir

et al., 2020; Bocker and Silva, 2022; Carpentieri *et al.*, 2023; Comuzzo *et al.*, 2020; Gałązka-Czarnecka *et al.*, 2018; Maza *et al.*, 2020; Mihalcea and Stoica, 2023; Puértolas *et al.*, 2010) (as can be seen in the Table 1).

Processing parameters	Findings	References
Pulse frequency: 0,5 Hz Electric field strength: 13.3 kV/cm Specific energy: 0–564 kJ/kg	Increased anthocyanin extraction	(Barba <i>et al.</i> , 2015)
Pulses number: 20	Higher productivity of	(Gałązka-
Electric field strength: 3.3–5	extraction of color	Czarnecka et al.,
kV/cm	compounds from grapes	2018)
Specific energy: 10–20 kJ/kg	Higher anthocyanin extraction	(Comuzzo <i>et al.,</i> 2020)
Pulse duration: 100 μs Electric field strength: 4 kV/cm Specific energy: 6.2 kJ/kg	Polyphenolic compounds extraction	(Maza <i>et al.</i> , 2020)
Pulse duration: 20 μs Pulse frequency: 5 Hz Electric field strength: 0.5–5 kV/cm Specific energy: 1–20 kJ/kg	Considerably increasing of health-related intracellular compounds from red grape pomace	(Carpentieri <i>et al.,</i> 2023)
Pulse duration: 100 ms Pulse frequency: 2 Hz Electric field strength: 3 kV/cm Specific energy: 100 kJ/kg	Recover bioactive glycosylated and lipidic compounds	(Salgado-Ramos <i>et al.</i> , 2023)

Table 1. Examples of bioactive compounds extraction from grape matrices by PEF technology

Extraction mechanism by electroporation is affected by various electrical parameters, including amplitude, waveform type (exponential, oscillatory, or square), pulse duration, pulses number, and frequency (Bocker and Silva, 2022). Typically, the extraction of valuable biomolecules is effectively achieved using low specific energy (1–10 kJ/kg), low electric field strength, and

short-duration pulses. The electroporation of plant biological cells using PEF is a complex, multiphase dynamic process which includes the following stages: an increase in cell transmembrane potential, initiation of hydrophilic pores on the cell membrane, evolution of the pore population, and a post-treatment phase during which either pore resealing occurs or the cell plasma membrane fails to recover from the pores. Once the pores are formed, the membrane loses its functionality, resulting in the extraction of different intracellular biocompounds (Mihalcea and Stoica, 2023) (Figure 2).

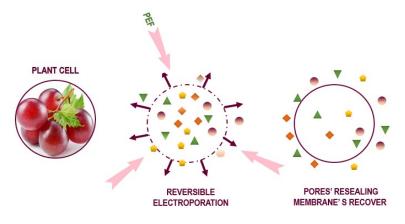


Figure 2. Extraction of health-related intracellular compounds by PEF (pink-purple circles – anthocyanins, yellow pentagons – carotenoids, green triangles – quercetin, brown diamonds – resveratrol; purple circle on the left – intact cell membrane, dotted purple circle – cell membrane reversible electroporated, purple circle on the right – recovered membrane). Schematic presentation (adapted from Mihalcea and Stoica, 2023).

During the extraction process, both reversible and irreversible electroporation may occur, irreversible electroporation enhancing the efficacy of the extraction process (Barba *et al.*, 2015; Mihalcea and Stoica, 2023). However, the efficacy of PEF extraction is influenced not only by electrical parameters (specific energy, electric field strength, pulse duration), but also by the characteristics of the treated raw materials (such as size, shape, conductivity, and ionic strength) and the properties of extracted components (including size and location within the cytoplasm or vacuoles) (Arcena *et al.*, 2021; Bălănică Dragomir *et al.*, 2020; Bocker and Silva, 2022; Mihalcea and Stoica, 2023; Stoica *et al.*, 2011).

PEF Technology in Winemaking Process

PEF can be applied at the grape level to enhance juice extraction contained inside the grape cells (at the beginning of vinification) and at various stages of vinification for must or wine decontamination (through the electroporation of wine spoilage microorganisms), helping in reducing the need for SO₂ (sulphur dioxide) additions (Feng *et al.*, 2022; Raso, 2023; Silva *et al.*, 2024) (Figure 3).

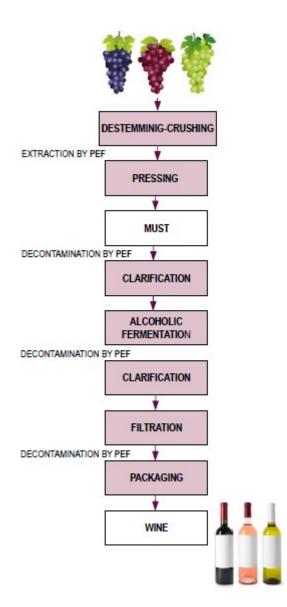


Figure 3. PEF technology in wine production (adapted from Silva et al., 2024).

 SO_2 is typically added at various stages of white wine production for its antimicrobial and antioxidant properties, but excessive consumption of this additive can have negative health

influences on consumers (Silva *et al.*, 2024). Wineries can leverage the capability of PEF to electroporate the cell membranes of red grape skins and microbial cells, thereby enhancing various winemaking processes. These enhancements include improving the extraction of primordial compounds, increasing juice yield, decreasing maceration time, increasing color intensity, enhancing polyphenol content (which increases the wine's antioxidant potential), inactivating spoilage microorganisms (helping to stabilize the wine), and forming aroma substances during aging (Feng *et al.*, 2022; Raso, 2023; Silva *et al.*, 2024) (Figure 4).

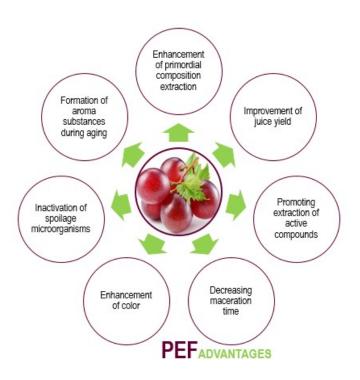


Figure 4. PEF advantages in winemaking (adapted from Feng et al., 2022)

The effectiveness of PEF in achieving these goals on grapes depends on the intensity of EF strength and total energy applied, as well as the characteristics of the biological cell, such as its morphology, electrical conductivity, and chemical composition (Arcena *et al.*, 2021). In traditional winemaking, only 40% of anthocyanins and 20% of tannins from red grape skins are transferred to the wine due to insufficient permeabilization of cell walls and cytoplasmic membranes (Feng *et al.*, 2022; Piccardo *et al.*, 2019). PEF can disrupt the cell membranes of grapes without affecting aroma, alcohol content, total acidity, pH, volatile acidity and

concentration in reducing sugar (Feng et al., 2022; Scharf and Sandmann, 2023; Vaquero et al., 2021). Although high voltage is required for electroporation, high-intensity PEF treatment exhibits a significant electrolytic effect, and its high EF strength combined with prolonged treatment duration may result in the degradation of certain macromolecular substances. PEF treatments increase the yield of grape juice, which may be related to the improvement of mass transfer in pulp tissues with a positive impact on wine quality (Fauster et al., 2022; Feng et al., 2022; Li et al., 2024; Scharf and Sandmann, 2023). PEF application during the maceration stage can significantly increase the phenolic compounds and tannin contents as well as color intensity, the color being the most important quality characteristic of red wines (Maza et al., 2019; Puértolas et al., 2010). However, an increase in EF strength can result in a reduced extraction rate of polyphenols, indicating that there is an optimal range for PEF treatment conditions. PEF treatment can substantially reduce the maceration time during winemaking, ensuring sufficient pigment extraction (Feng et al., 2022). PEF treatment enhances the removal of suspended solids such as microorganisms (especially yeast) and other particulates, resulting in clearer red and white wine (an essential quality characteristic valued by consumers) (Mierczynska-Vasilev and Smith, 2015), and ensures wine stability before bottling or other packaging methods (Silva et al., 2024).

Conclusions

Pulsed electric field (PEF) is a non-thermal, eco-friendly, and cost-effective technology that significantly enhances microbial inactivation and mass transfer in food processing, particularly in the winemaking process. The effectiveness of PEF in microbial inactivation involves a dynamic, multiphase process where the electric field induces electroporation, leading to either reversible or irreversible disruption of biological cell membranes. This technology facilitates the extraction of valuable intracellular compounds, such as anthocyanins, polyphenols, and resveratrol, from plant tissues by increasing membrane permeability through the formation of hydrophilic pores. The application of PEF in winemaking improves juice yield, decreases maceration time, enhances color intensity, increases polyphenol content, and inactivates spoilage microorganisms, thereby stabilizing the wine and reducing the need for sulphur dioxide additives. The efficacy of PEF is influenced by various electrical parameters (amplitude, waveform, pulse duration, number of pulses, and frequency) and the characteristics of the raw materials (size, shape, conductivity, and

ionic strength), necessitating optimization for each specific application. PEF-treated wines exhibit improved clarity and stability, without negatively affecting key quality parameters such as aroma, alcohol content, total acidity, pH, and volatile acidity.

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