Food Research International 123 (2019) 95–105

Contents lists available at ScienceDirect

Food Research International

journal homepage: www.elsevier.com/locate/foodres

Review

Application of pulsed electric fields in meat and fish processing industries: An overview

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ARTICLE INFO

Keywords:
Electroporation
Non-thermal technology
By-products valorization
Tenderization
Aging
Preservation

ABSTRACT

The market demand for new meat and fish products with enhanced physicochemical and nutritional properties attracted the interest of the food industry and academia to investigate innovative processing approaches such as pulsed electric fields (PEF). PEF is an emerging technology based on the application of electrical currents between two electrodes thus inducing electroporation phenomena and enabling a non-invasive modification of the tissues' structure. This review provides an overview of the current knowledge on the use of PEF processing in meat and fish to enhance the physicochemical and nutritional changes, as a preservation method, as well as for improving the extraction of high added-value compounds. PEF treatment had the ability to improve several processes such as preservation, tenderization, and aging. Besides, PEF treatment could be used as a useful strategy to increase water holding properties of fish products as well as for fish drying. Finally, PEF could be also used in both meat and fish foods for by-products valorization, due to its potential to enhance the extraction of high added-value compounds. However, more studies are warranted to completely define specific treatments that can be consistently applied in the industry. This review provides the directions for this purpose in the near future.

1. Introduction

Pulsed electric field (PEF) is among the electrical-based processing techniques (Barba et al., 2015; Mannozzi et al., 2018). However, the application of short electrical pulses at high voltages enables the control of thermal effects to remain low, making it different from those of thermal electrical-based techniques, such as ohmic heating (Gavahian & Farahnaky, 2018; Gavahian, Farahnaky, Javidnia, & Majzoobi, 2012), and moderate electrical field (Gavahian, Chu, & Sastry, 2018). These characteristics make PEF a promising technique for disrupting biological cells in the food matrix without any detrimental effect on the attributes of food products (Kumar, Kumar Patel, & Kumar, 2015; Puértolas, Koubaa, & Barba, 2016).

Although the concepts of this technique were introduced to the food industry about 50 years ago, PEF can be still considered an emerging technology due to the recent developments in its industrial applications. The applicability of PEF for the treatment of food commodities is well explained in the literature (Barba, Koubaa, do Prado-Silva, & Orlien, 2017; Gabric et al., 2018; Misra et al., 2017; Soliva-Fortuny, Balasa, Knorr, & Martín-Belloso, 2009), indeed it has been reported as a novel preservation method that has the capacity to produce foods with great nutritional and sensory quality and shelf-life (Barba et al., 2017; Horita, Baptista, Caturla, Lorenzo, & Barba, 2018; Kumar et al., 2015). Particularly, research is currently focused on two major applications of this technique including non-thermal microbial inactivation and improvement of mass transfer through cell disruption. These applications

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https://doi.org/10.1016/j.foodres.2019.04.047
Received 7 March 2019; Received in revised form 16 April 2019; Accepted 18 April 2019

are accomplished under different processing conditions as microbial inactivation is acting on small cells while mass transfer phenomena usually involve the disruption of bigger structures such as the glands of aromatic herbs (Barba et al., 2015; Barba et al., 2017; Jäger, 2013; Puértolas et al., 2016). The ability of PEF to perform efficiently the aforementioned goals depends on different factors, such as number and duration of electric pulses and cell properties (Mannozzi et al., 2018; Martínez, Delso, Álvarez, & Raso, 2019). The process parameters of PEF can be adjusted depending on the application desired. In addition, the diversity of raw materials and the availability of PEF equipment may affect the PEF process conditions.

Likewise, there are various ways for the generation of PEF with different pulse features. Kumar et al. (2015) explained that different voltage waveforms, including exponential pulses, square wave, bipolar and oscillatory pulses, can be applied in a PEF treatment. In addition, the design of the treatment chambers is probably the most significant element in terms of the comparability of results. The shape of the chamber has an important effect on the electric field distribution and on the total resistance and also on the discharge circuit. With this in mind, it is correct to affirm that PEF treatments have multi-scale nature and all the aspects must be considered. It is indispensable to analyze the PEF process into complex food processing systems (Jäger, 2013). Besides the treatment chamber, high-intensity PEF involves a number of components such as a power source, a switch, a capacitor tank, temperature sensors and aseptic packaging equipment (Kumar et al., 2015).

There are different tools available for analyzing the impact of the electric field on cellular and tissue levels. The degree of cell permeabilization requires being evaluated and quantified. The membrane permeabilization influences the turgor of the cell, thus turgor and texture measurements, such as stress deformation and relaxation assays of complex tissue, may be used for the valuation of the degree of tissue damage and cell rupture (Lebovka, Praporscic, & Vorobiev, 2004; Tomos, 2000). Similarly, other methodologies used to evaluate the cell rupture or membrane disintegration have been used to promote the release of plant pigments (Barba et al., 2015; Dörnenburg & Knorr, 1993). Methodologies reported in the literature for monitoring the degree of electroporation include the measurement of conductivity-frequency spectra (in the range from 10^3 to 10^7 Hz) (Lebovka, Bazhal, & Vorobiev, 2001), and the application of an acoustically derived determination (Grimi, Lebovka, Vorobiev, & Vaxelaire, 2009).

In addition to the ability of reducing the microbial load in different food products such as fruit or vegetable juices (Barba et al., 2017; Evrendilek, Zhang, & Richter, 2004; Gabric et al., 2018; Hodgins, Mittal, & Griffiths, 2002; Marsellés-Fontanet, Puig, Olmos, Mínguez-Sanz, & Martín-Belloso, 2009; Saldana, Puertolas, Monfort, Raso, & Alvarez, 2011), PEF treatment can be a good strategy to change the textural properties during production and processing of numerous products, since modifications in the tissue structure can largely affect product characteristics (Fincan & Dejmek, 2003; Lebovka et al., 2004; Pereira, Galindo, Vicente, & Dejmek, 2009).

The utility of PEF in the food industry is gaining reputation, being considered as an innovative food processing system appropriate for the pre-treatment of liquid and semi-solid edible products (Ricci et al., 2018). However, it is an application restricted to products with low electrical conductivity and no air bubbles in order to avoid the dielectric breakdown (Chauhan & Unni, 2015).

Despite of the fact that PEF application is a non-thermal food manufacturing technology, there is a considerable temperature increase during high intensity PEF treatment, which must be considered with sensitive compounds like proteins (Barba et al., 2015; Jäger, 2013). Depending on the processing parameters and treatment conditions, PEF side effects such as a certain temperature increase or the occurrence of electrochemical reactions must be taken into account to preserve the food quality. Therefore, little damage to pigments, flavor compounds or vitamins, and consequently the degradation of some sensory characteristics and nutritional value of foods could be perceived. The detection and knowledge of the possible negative side effects can be useful to improve the technique, for instance the treatment chamber design.

Despite numerous research on the principle (Wouters, Alvarez, & Raso, 2001) and the applications of the PEF to various products, including alcoholic beverage (Yang, Huang, Lyu, & Wang, 2016), avian eggs (Yogesh, 2016), and dairy products (Buckow, Chandry, Ng, McAuley, & Swanson, 2014), there is still a need to better understand the applicability of this emerging technology in meat and fish industries. Therefore, this review aims to provide detailed information about the potential benefits and applications of this innovative technology for both researchers and the industry. The mechanisms involved in a PEF process and considerations for successful industrial implementation of PEF in these industries are also taken into account. The research needed in this area of the science is also discussed.

2. General description, advantages, and disadvantages of PEF

PEF processing is the application of high voltage pulses for short duration times to foods placed between two electrodes. In general, PEF affects biological cells resulting in specific structural modifications and cell membrane disruption (Barba et al., 2015). There is a field threshold value of about 1–10 (kV/cm) depending on the cell type (e.g. microbial, plant or animal), that when it is exceeded, the electro-compressive force induces a local dielectric breakdown of the membrane resulting in the creation of a pore, which can then work as a conductive channel (Kumar et al., 2015; Oziembłowski & Kopeć, 2005; Puértolas & Barba, 2016). More specifically, due to the high electric field pulses, cell membranes develop pores, either by enlargement of existing pores or by the generation of new ones, which may be permanent or temporary, depending on the operating conditions, and causing the increase of membrane permeability (Chauhan & Unni, 2015). Another proposed electroporation model consists of an external applied electric field of sufficient strength that promotes an increase in the permeability of cell membrane. This increase is associated with the formation of aqueous pathways, i.e., pores, in the lipid bilayer of the membrane (Mahnič-Kalamiza, Vorobiev, & Miklavčič, 2014).

If the cell membrane is disrupted, intracellular contents leak out with the consequent loss of cell metabolic activities (Chauhan & Unni, 2015). PEF lethal impact will depend on different factors as the electric field strength (EFS) and the treatment time, as well as on the microorganism itself, treatment temperature and the medium characteristics (Barba et al., 2015; Morales-de la Peña, Welti-Chanes, & Martín-Belloso, 2019; Ohshima, Tamura, & Sato, 2007). In general, an increment in the number of pulses improves microbial inactivation, but it can also induce a significant heating of product (Oziembłowski & Kopeć, 2005). It is of interest to notice that the inactivation of larger microbial cells needs less intense field strengths to suffer a similar inactivation than smaller cells. In fact, the field induces a modulation of the transmembrane potential in a cell size-dependent way. Moreover, cells in their exponential growth phase are more susceptible to PEF treatments than the same cells in lag or stationary phase (Álvarez, Raso, Palop, & Sala, 2000).

Fig. 1 illustrates the mechanism of electrical reversible (leaving the cell either damaged or able to fully recover) and irreversible breakdown (the cell losses membrane integrity and cell material leading to cell death) of a microbial cell. When the applied field exceeds a critical threshold value, this causes cell rupture, and large pores are formed due to an increment in the intensity of the electric field and/or in the pulse duration (Chauhan & Unni, 2015; Rodrigo, Sampedro, Silva, Palop, & Martínez, 2010).

The treatment can be carried out at ambient, sub-ambient, or slightly above ambient temperature, and it is possible to minimize energy loss due to heating of foods (Pourzaki & Mirzaee, 2008). The number of pulses may vary from hundreds to thousands, as well as their time duration ranging from micro to milliseconds, and the particular

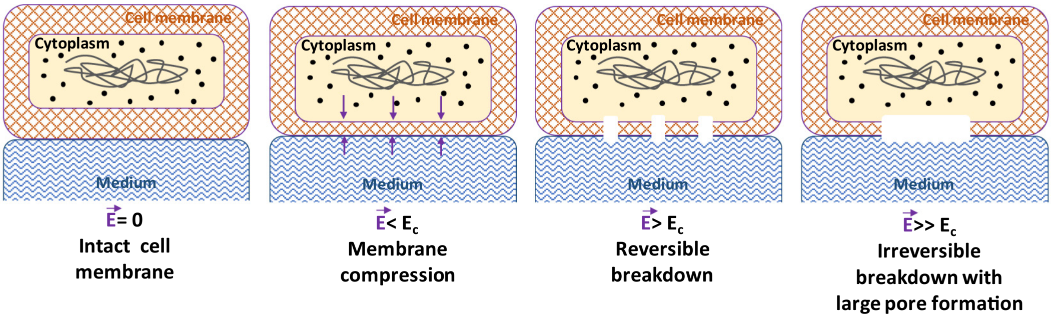

Fig. 1. Schematic representation of the compression and possible breakdown by pore formation of a biological cell subjected to a pulsed electric field treatment (E). E_c is the critical electric field.

intensity of the process can be adjusted on the basis of the geometry and distance of the working electrodes, the voltage delivered, and the conductivity of the material treated (Ricci et al., 2018). Particularly important is the EFS that goes through the food sample, which is inversely proportional to the gap distance between the electrodes, but directly proportional to the voltage supplied across them (Chauhan & Unni, 2015).

PEF, as a non-thermal cell membrane permeabilization method, is characterized by its low energy consumption, its continuous operability with short processing times, and is a waste-free process, allowing the development of innovative, cost-effective and sustainable processing concepts in the food and drink industry, as well as in the biotechnology and pharmaceutical industry (Barba et al., 2015; Puértolas et al., 2016; Puértolas & Barba, 2016). In fact, treatment with high voltage electric field pulses with a very short duration (1–100 μs) has been of interest to many researchers as enables the inactivation of microorganisms without losing quality neither in the flavor nor in the taste of food (Ohshima et al., 2007).

PEF equipment consists of a pulse generator, the treatment chamber, electrodes correctly designed to avoid the effect of electrolysis, and a system for control and data acquisition (Puértolas et al., 2016). These basic components are schematically represented in Fig. 2. There are different manners to implement the pulse generator (Pourzaki & Mirzaee, 2008). The treatment chamber contains the fundamental electrodes between which there is a space to place the food to be treated. Energy from the power source is retained in the capacitor and is discharged through the treatment chamber to induce an electric field in the food material.

In addition to the set-up of the entire device and the EFS, several process parameters must be defined while designing a PEF process. These include the number of pulses, pulse duration, pulse width, pulse shape, pulse specific and pulse frequency (Puértolas & Barba, 2016; Puértolas, Luengo, Álvarez, & Raso, 2012). The above-mentioned variables and the duration of the PEF treatment should be taken into account for evaluating the efficiency and economic impacts of PEF treatment, especially for up-scaled application. It was previously explained that the specific energy input of the process (which expresses as kJ/kg and depends on the input voltage, the ohmic resistance of the treated products, and the processing time) is closely associated to the economic cost and environmental footprint of a PEF process. Hence, researchers use this parameter to compare the cost and sustainability of PEF processes with those of conventional methods (Puértolas & Barba, 2016).

According to Ricci et al. (2018), the habitually used pulse shapes are exponential decays and square waveform. The latter one can be cataloged as the most suitable to optimize the effect of a PEF application: in square waveform any power decay happens and the intensity remains without changes for the whole duration pulse width, boosting efficiency compared to the pulses sent per unit of time.

After the PEF treatment, the material is cooled if necessary, packed aseptically, and then stored at refrigerated or ambient temperatures depending on the type of food and their future use/s. The major

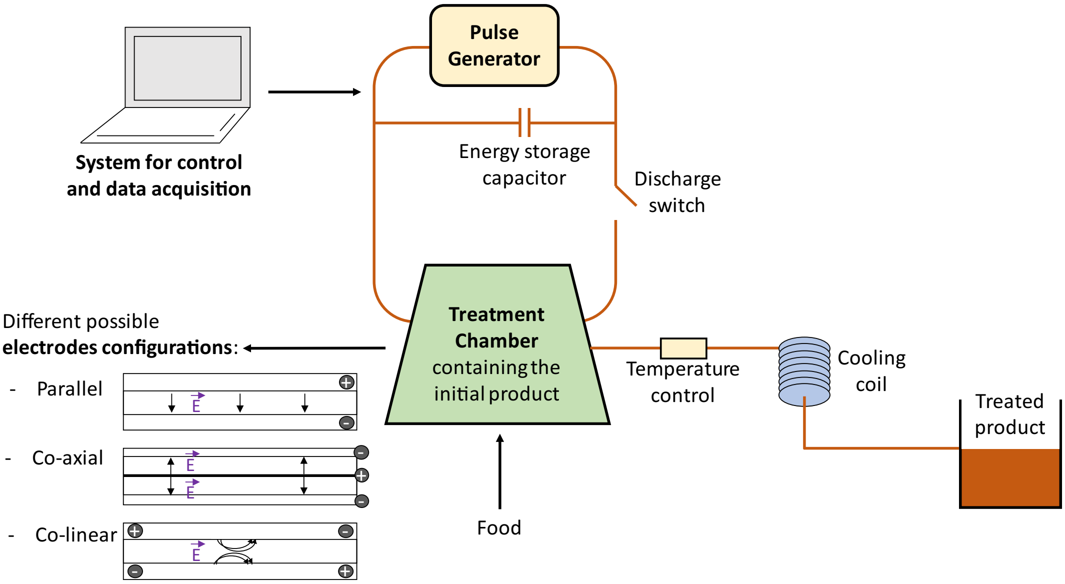

Fig. 2. Scheme of a pulsed electric field (PEF) processing system which generates exponential decay pulses, including different configurations of electrodes (parallel, co-axial and co-linear).

Table 1
Enumeration of advantages and disadvantages of the application of pulsed electric fields (PEF) in meat and fish industries.

Pulsed electric field for food processing	
Advantages	Drawbacks
Efficient inactivation of cells	High capital cost
Suitable for procesing heat-sensitive foods	Inefficient in spore inactivation
Applicable to many solid and liquid foods	Unavailability of commercial units in many regions of the world
Can perform the pasteurization of a wide range of meat products	
Better retention of color, flavors, and nutrients	The presence of bubbles may lead to non-uniform treatment as well as operational problems
Relatively short treatment time	
Free from health-threatening solvent and Environmental friendly	Limited economic and engineering studies for up-scaled continuous process
Possibility of combination with other processing techniques	

concern for industrialization of the application of PEF in food processing is the initial capital investment. However, in spite of some limitations, there are many advantages which have been collected in Table 1.

PEF technology provides a variety of opportunities including continuity of the process, high retention of nutrients and vitamins, and high organoleptic quality of the product. PEF technology can be more effective when applied with other methods, as high hydrostatic pressure, ultrasound and effective temperature control technologies upstream and downstream (Huang, Mittal, & Griffiths, 2006; Jaeger, Meneses, Moritz, & Knorr, 2010; Riener, Noci, Cronin, Morgan, & Lyng, 2008). Nevertheless, the combination with different methods and the optimization of the corresponding parameters will depend on the material and of the objective of the treatment. Therefore, it is essential to continue investigating. Moreover, PEF technology offers the potential to efficiently and economically improve energy consumption, to control the presence of microorganisms in foods in a fast and homogeneous way and to preserve the nutritional properties. For instance, the life cycle assessment (LCA) of PEF in potato processing was recently assayed, allowing to characterize its general environmental impact. The study revealed that replacing conventional pre-heaters with PEF equipment can reduced the energy and water consumptions by 85% and 90%, respectively. In addition, the authors observed an improvement in both product quality and product yield when PEF was used in this process (Fauster et al., 2018). In the same line, the LCA was used to evaluate the potential of PEF technology for facilitating the steam peeling of tomato fruits at an industrial scale (Arnal et al., 2018). The article concluded that PEF is an environmentally friendly alternative to traditional processing techniques as it improved all the studied environmental indicators by 17–20%. Furthermore, Davis, Moates, and Waldron (2010) investigated the environmental impact of PEF treatment of carrot juice and concluded that the packaging must be carefully considered for a sustainable PEF process. According to the authors, selecting appropriate packaging system enables the food processors to benefit from the possible environmental advantage of this emerging technology (Davis et al., 2010).

On the other hand, products of electrolysis may adversely affect foods and the presence of bubbles can derive in non-uniform treatments, as well as in safety and operational problems. In addition, this technique for pasteurization purpose is rather limited to liquid food products as the high EFS required for microbial inactivation normally implies very small distance gaps between the electrodes (in the magnitude of millimeters), what is not the case for standard meat products. Other negative points include the resistance of microbial spores to inactivation, and its upscaling is still under development (Chauhan & Unni, 2015; Kumar et al., 2015; Oziembłowski & Kopeć, 2005).

3. Use of PEF technology for meat and meat products

The elaboration of meat and meat products includes curing periods with the aim of increasing the shelf-life and developing specific flavors and aroma profiles. The uptake of the common substances used in this process, such as salt, nitrites, and spices, could be improved after PEF treatment due to the partial disruption of cellular tissues and its effect in the mass transfer processes. In summary, PEF would be able to reduce the concentration of additives and the process duration required, because of an improved absorption rates. In relation with this, McDonnell, Allen, Chardonnereau, Arimi, and Lyng (2014) described the potential use of PEF for accelerating the salting of pork without causing significant effect on weight change post-curing, water-binding capacity or texture, and highlighted the necessity of optimizing the different treatment parameters.

Most of the traditional processes for meat preservation, such as drying, frozen storage, and salting, exert negative effects on the microstructure of the product compared to the fresh product (Gudmundsson & Hafsteinsson, 2001). This fact makes evident the emerging possibilities that PEF treatments can have in this field. However, it is important to consider that some of the electrical energy input is converted to heat during processing which may affect the meat quality attributes (McDonnell et al., 2014). Although the heat originated by thermo-electric effect is moderate, the synergistic effect of the two could influence the physical characteristics of beef. O'Dowd, Arimi, Noci, Cronin, and Lyng (2013) employed low PEF electric fields on beef muscle and observed significant weight loss while the texture was unaffected.

3.1. Meat tenderization

Increased tenderness can improve the value of meat, as tenderness is one of the main contributors to sensory quality and consumer acceptability. For instance, some studies performed in beef have reported that PEF application to muscles before aging improved the tenderization process, as well as increased the proteolysis of myofibrillar proteins (Table 2). Particularly, the application of repeated (1×, 2×, 3×) PEF had a positive impact on the tenderization of beef *M. longissimus lumborum* muscles (LL), while *M. semimembranosus* muscle (SM) was not affected by PEF treatments (10 kV, 90 Hz and 20 μs) and using a PEF system in batch mode with the meat fiber direction in parallel to the electrodes (Suwandy, Carne, van de Ven, Bekhit, & Hopkins, 2015c). Authors indicated that accelerated proteolysis was probably the responsible mechanism for the improved tenderization of cold-boned beef LL muscle subjected to 1 × PEF, whereas the cause for the improved tenderization of cold-boned beef LL muscle subjected to 2× and 3× PEF treatments could be an increase in proteolysis together with a physical alteration to the muscle structure. In addition, this study did not found changes in lipid oxidation, which was suggested as an advantage due to the non-occurrence of undesirable flavors and odors. This can be considered as an advantage of PEF over some other non-thermal processing techniques such as cold plasma (Gavahian, Chu, Mousavi Khaneghah, Barba, & Misra, 2018). The same PEF repetitions and conditions were evaluated on the quality of hot-boned beef loins and topsides, and the analysis of tenderness was again the main focus. The PEF-treated samples of both muscle types were aged during 3, 7, 14 and 21 days (Bekhit, Suwandy, Carne, van de Ven, & Hopkins, 2016).

Table 2
Recent studies of pulsed electric fields (PEF) processing on meat tenderization.

Meat material	PEF conditions	Findings/effects	Reference
Beef loins (LL) and topsides (SM)	Batch mode, the meat fiber direction was parallel to the electrodes. Different combinations were used: EFS 0.27 to 0.56 kV/cm; input voltages 5 and 10 kV; frequencies 20, 50 and 90 Hz; pulse width 20 μs; a variety of pulse numbers	Promising potential of PEF to tenderize meat and to improve the cooking loss	Bekhit et al. (2014)
Beef LL and SM muscles	Batch mode, the meat fiber direction was parallel to the electrodes. EFS 0.28 to 0.51 kV/cm; voltages 5 and 10 kV; frequencies 20, 50 and 90 Hz; pulse width 20 μs; a variety of pulse numbers	Potential of PEF to tenderize beef SM muscles, which can achieve a 21.6% reduction in the shear force. LL muscles tended to get tougher with increasing treatment frequency. Each muscle type must be treated with the optimum PEF treatment intensity	Suwandy et al. (2015a)
Cold boned beef loins (LL)	PEF system in batch mode. EFS 0.58 to 0.73 kV/cm; voltage 10 kV; frequency 10 Hz; pulse width 20 μs	PEF treatment had potential to improve the tenderization of cold-boned beef and was effective in improving the proteolysis of low pH meat samples	Suwandy et al. (2015b)
Cold-boned beef LL and SM muscles	PEF system in batch mode, and the meat fiber direction was parallel to the electrodes. Four levels of PEF treatments (0×, 1×, 2× and 3× repeats of electrical stimulation); EFS 0.50 to 0.58 kV/cm; voltage 10 kV; frequency 90 Hz; 20 μs	The use of repeated PEF had positive impact on the tenderization of beef LL muscles without increasing off-flavor or off-odor, but not in the SM muscle	Suwandy et al. (2015c)
Beef semitendinosus muscles	Batch mode configuration. EFS 1.4 kV/cm; frequency 50 Hz; pulse width 20 μs; pulse number 1032; total specific energy input of 250 kJ/kg	PEF affected microstructure of beef tissue, influencing its water holding capacity and textural properties. Both applied PEF conditions and sample pre-treatment (fresh or frozen-thawed) should be considered for determining the effect of PEF on meat tenderization	Faridnia et al. (2015)
Turkey breast meat	Batch PEF chamber (distance between electrodes 4 cm). EFS up to 3 kV/cm; frequency 5 Hz; pulse width 20 μs; pulse number 300	PEF treatments did not induce any major adverse side effects on the lipid oxidation of the turkey meat, neither in weight loss, cook loss, lipid oxidation, texture, and color either on fresh or frozen samples	Arroyo, Eslami, et al. (2015)
Beef longissimus thoracis et lumborum	Batch mode using two parallel electrodes. EFS 1.4 kV/cm; frequency 10 Hz; pulse width 20 μs; pulse number 300 and 600	PEF treatments resulted in no detrimental effect on cook loss, storage loss and color regardless of the length of aging before PEF application and of the length of aging after PEF application	Arroyo et al. (2015)
Hot-boned beef loins and topsides	Batch mode and the meat fiber direction was parallel to the electrodes. Repeats of 1×, 2× and 3×; EFS 0.44 to 0.48 kV/cm on LL muscle and 0.32 to 0.35 kV/cm on SM muscle; voltage 10 kV; frequency 90 Hz; pulse width 20 μs	PEF treatment had differential effect on water holding capacity and tenderness of hot-boned LL and SM muscles	Bekhit et al. (2016)
Cold boned beef *M. longissimus et lumborum*	Batch mode and the meat fiber direction was parallel to the electrodes. EFS 0.23 to 0.68 kV/cm; voltage 2.5 and 10 kV; frequency 200 Hz; pulse width 20 μs	There is an optimal PEF treatment for beef cuts within a range of processing parameters. Low and high PEF treatments can lead to ultrastructural changes in beef LL.	Khan et al. (2017)
Briskets (deep pectoralis muscle) from beef animals	Triangular PEF chamber with an electrode distance of 4 cm. Batch mode configuration EFS 1 and 1.5 kV/cm; range of voltages 15–35 kV; specific energy levels of 40–50 and 90–100 kJ/kg; frequency 50 Hz; pulse width 20 μs; a variety of pulse numbers	PEF can be used to reduce the cooking time of tough meat cuts since it physically weakens the connective tissue and increase the collagen solubility	Alahakoon, Oey, et al. (2017)

EFS: Electric field strength. *LL: longissimus lumborum. SM: semimembranosus* muscle.

The outcome of this research suggested that the same PEF treatment can lead to different effects on water holding capacity and tenderness of hot-boned LL and SM muscles. In fact, the shear-force of hot-boned beef LL muscles was significantly increased by 3× PEF treatment over 21 days of post-treatment period, whereas the shear-force of hot-boned beef SM muscles was significantly decreased by 3× PEF treatment at 3 days post treatment. This indicated the dependence of PEF-induced effects to the specific primal cuts being treated.

In the same direction, the impact of PEF on the quality of beef loins (LL) at 1 day postmortem and topsides (SM) at 1 and 3 days postmortem was evaluated by Bekhit, van de Ven, Suwandy, Fahri, and Hopkins (2014) varying the voltages (5 and 10 kV) and frequencies (20, 50 and 90 Hz). Authors used batch mode, the meat fiber direction in parallel with the electrodes and pulse width = 4–32 μs. The main objective was to select the optimal treatment for each muscle and determine the economic and texture benefits. As a result, these authors observed a promising capacity to tenderize meat using PEF, with almost 20% reduction in the shear force. Though in the case of the SM muscle treated with PEF, this reduction was dependent on the treatment. Additionally, the improvements observed in the SM tenderness were not influenced by the muscle postmortem time, which translates into greater flexibility in the use of this technology in the meat processing plant.

Likewise, Suwandy, Carne, van de Ven, Bekhit, and Hopkins (2015a) concluded that PEF represents a promising method to tenderize beef SM muscles after achieving a 21.6% reduction in the shear force. Nevertheless, a different situation was detected for beef LL muscles in which the increase in the treatment frequency derived into tougher meat. Not only different behaviors were observed in the shear force measurements of both muscles, but also in the water holding capacity. Meanwhile, another study highlighted the potential of PEF for improving the proteolysis of low pH meat samples (Suwandy, Carne, van de Ven, Bekhit, & Hopkins, 2015b) after detecting a significant increased proteolysis (based on degradation of troponin-T and desmin) in the low pH (5.5–5.8) samples compared to the high pH (> 6.1) ones. This fact was reflected in the different shear force values found for each of the samples.

In another study, Alahakoon, Oey, Silcock, and Bremer (2017), reported the improvement of the tenderness, with the corresponding reduction of the cooking time in collagen-rich meat cuts using a triangular PEF chamber with an electrode distance of 4 cm (length of 6 cm and depth of 6 cm). This highlighted the possibility of using PEF processing for lower value cuts rich in connective tissue. Particularly, they designed a novel model system in which was possible to decrease the thermal stability of connective tissue obtained from beef *deep pectoralis* muscle (brisket).

Recently, Bhat, Morton, Mason, and Bekhit (2018) summarized emerging methods useful for meat tenderization and included PEF as a novel technology with numerous advantages in front of traditional processing methods including limited side effects, short processing time and low treatment temperature. Moreover, many studies did not find any significant impact of PEF on water holding capacity, color and lipid oxidation (Arroyo et al., 2015; McDonnell et al., 2014; Suwandy et al., 2015b), which is very relevant to guarantee an industrial application.

The potential of PEF to improve meat tenderness in beef muscles has been confirmed by several authors and appear to depend on a variety of factors such as the sample under study, the EFS or the presence of an aging stage. However, PEF has been also characterized for inducing negative effects on tenderness. According to the previously mentioned work of Suwandy et al. (2015a), beef LL was found to get tougher with high-intensity PEF treatment (10 kV, 90 Hz, pulse width 20 μs) using batch mode and the meat fiber direction in parallel to the electrodes. The outcome could be explained by the ohmic heating during the application of this treatment, promoting denaturation of the proteins and enzymes involved in the tenderization process.

The previously conducted studies revealed that, in some cases, combination of PEF with other processing techniques can improve the non-thermal effects of PEF on the treated sample. For example, Faridnia et al. (2015) reported that combined freezing-thawing and PEF led to improved tenderness based on the reduced shear force observed. The results of this study suggested that both selected PEF conditions and sample pre-treatment (fresh or frozen-thawed) should be considered when evaluating the impact of PEF on meat tenderization. In addition, this research group explained the mechanisms involved in meat tenderization by PEF according to the results from transmission electron microscopy. It is believed that changes in the microstructural structures can alter the tenderness of meat samples.

3.2. Meat aging

Like other food products such as cheeses or wines, beef meats can improve their properties as texture after an aging stage of the pieces. Currently, aging is one of the main options in the industry to carry out the meat tenderization. This process involves the breakdown of muscle fibers by endogenous proteolytic enzymes (calpains, lysosomal cathepsins, multicatalytic proteinase complex) but has the drawback of being a slow, time-consuming and expensive process due to long refrigeration storage (Alahakoon, Faridnia, Bremer, Silcock, & Oey, 2017; Faridnia, Bekhit, Niven, & Oey, 2014). Aging in combination with PEF treatment appears to be an appropriate alternative to improve meat tenderness due to an increased rate of proteolysis (Faridnia et al., 2015).

Bekhit et al. (2016) investigated the effect of different PEF treatments followed by aging for a variety of time periods (3, 7, 14 or 21 days) on the tenderness of beef LL and SM muscles, observing an interaction between PEF treatment and aging. The PEF system was used in batch mode, the meat fiber direction was parallel to the electrodes, and the treatment time was 30 s. The detailed technical specifications of this PEF treatment are presented in Table 2. According to the authors, both aging period and PEF repeats affected purge loss (%) and textural (rheological) properties of hot-boned beef muscles. Previously, shear force values were significantly reduced in response to aging times regardless of the PEF treatment (Faridnia et al., 2014). Meanwhile, Faridnia, Bremer, Burritt, and Oey (2016) assayed the impact of PEF application in batch mode and parallel electrodes (EFS of 1.7–2.0 kV/cm, pulsed electrical energy of 185 kJ/kg, frequency of 50 Hz and pulse width of 20 μs) on beef muscles and its potential use to reduce meat aging duration and cost. Particularly, this last work did not find any detrimental effect on meat cook loss and color stability in the PEF-treated samples regardless of the duration of aging period. In addition, authors observed that PEF causes alterations in the microstructure and texture of the meat. Therefore, it can be a suitable strategy not only to improve the tenderness but also to reduce the aging time and to modify functional properties. However, the use of this technique cannot be generalized since existing significant differences depending on the animal species and the meat cuts have been observed. Further research and experiments are warranted to optimize the aging conditions and to determine its effects on PEF-treated meat.

3.3. Changes in physicochemical properties

As it is already known, the PEF-induced effects are more complex in multicellular systems that are made up of different cellular types with numerous properties within a variety of tissues. Namely, a PEF-treatment which is applied to enhance mass transfer, will modify the characteristics of a product in a different way depending on the nature of the product itself. In this context, it is important to take into account that meat is a complex set of connective tissue, adipose, vascular and nervous tissues, and longitudinal, multinucleated muscle cells. In addition, in accordance with Alahakoon, Faridnia, et al. (2017), muscle fiber composition is defined among the most important by the species muscle type, breed, age, environmental conditions, history of physical activity and nutrition.

In general, PEF processing affects the muscle cell membranes influencing the interaction between fatty acids and cell membrane phospholipids with prooxidant effect in meat (Faridnia et al., 2015). Such interactions can originate undesirable compounds able to decompose into secondary products which can cause off-flavors and odors in meat and reduce its sensorial and nutritional quality. Regarding meat color, high PEF intensities and numbers of PEF repeats may lead to negative effects on the meat appearance as increases the temperature of the product and promotes myoglobin oxidation, in contrast to mild PEF conditions (Alahakoon, Faridnia, et al., 2017). Moreover, water holding capacity is another factor that can be altered in the PEF-treated meat samples due to the generation of pores and the facilitation of water movement.

It has been suggested that frozen meat may be more susceptible to PEF applications than fresh samples since the synergistic effect of freezing and PEF can result in an accelerated proteolysis with the subsequent impact in meat tenderization (Alahakoon, Faridnia, et al., 2017; Faridnia et al., 2015). Thus, when cell's susceptibility for pore formation is reduced, it is so the efficiency of PEF treatment (Faridnia et al., 2015). In fact, Faridnia et al. (2014) kept the beef muscle samples at refrigeration temperatures (4 °C) before mild PEF treatments using a batch mode configuration and different combinations of EFS and frequency (0.2–0.6 kV/cm, 1–50 Hz, 20 μs), and they did not find changes in the appearance and physical properties of meat based on no significant differences in color stability, pH, cooking losses and protein profile. Another study observed that samples of cooked lamb meat cuts subjected to prolonged storage time and frozen-thawed pretreatment prior to PEF led to significant increases in volatile compounds due to lipid and protein oxidation. In this case, PEF-treated for all cuts of samples were associated with brown color, juicy, livery, and meat flavor attributes (Ma et al., 2016). Therefore, it is important to establish a proper thawing step and meat temperature control prior to application of PEF treatment in order to obtain a good meat quality product.

In a recent work, Khan et al. (2017) demonstrated that high PEF (10 kV, 200 Hz and 20 μs) treatment can negatively influence the quality of beef in comparison to the effects observed for low PEF treatments (2.5 kV, 200 Hz and 20 μs). They worked with a treatment chamber containing two electrodes consisting of two parallel stainless steel plates that were positioned apart at a distance of 8 cm by a Teflon insulating material. Specifically, the parameters that were negatively affected by the intense PEF treatment were temperature, shear force, color, lipid oxidation and mineral levels (P, K, and Fe).

Moreover, it has also been reported that PEF treatment prior to *sous vide* processing had no significant effect on color stability, lipid oxidation or cooking loss of brisket. But increasing the EFS and/or prolonging the *sous vide* time significantly increased collagen solubilization in the sous vide brisket.

More recently, Bhat, Morton, Mason, Mungure, et al. (2019), Bhat et al. (2019) evaluated the effect of PEF on the proteolytic, enzymatic and physicochemical changes of venison and did not find a significant impact of the PEF treatment on shear force, neither on myofibrillar fragmentation index after aging for 21 days. Nevertheless, PEF processing resulted in a slight tendency towards increasing the calpain activity and the proteolysis of venison samples. Previously, similar PEF conditions led to a significant tenderization effect on beef, hence different processing conditions must be tested to promote significant responses in muscles from different species.

3.4. Protein digestibility

The presence of significant amounts of non-hydrolyzed proteins in the colon can induce the production of harmful metabolites through their fermentation by colonic microbiota. Therefore, the idea of promoting the intake of foods products with improved digestibility and nutritive value is gaining interest in a society concerned about their health and quality food (Bhat, Morton, Mason, & Bekhit, 2019). In this sense, recently conducted research have showed that PEF treated beef samples possess faster and greater in vitro digestion properties compared to the untreated beef samples.

In a recent study, PEF treatments in batch mode and parallel electrodes (T_1: 5 kV, 90 Hz, 20 μs and T_2: 10 kV, 20 Hz, 20 μs) enhanced the values of protein digestibility and solubility (Bhat, Morton, Mason, & Bekhit, 2019). In a second recent study, PEF treatment in batch mode (EFS 1–1.5 kV/cm, 48–178 kJ/kg, pulse width of 20 μs and frequency of 50 Hz) improved in vitro protein digestibility by at least 18% (up to 31%) and the digestive profiles (SDS-PAGE) of both control and PEF-treated samples were different. This corresponded to more severe *Z*-disks and I-bands disruption in PEF-treated samples after 180 min of simulated digestion, showing that the PEF-treated muscle is more susceptible to enzymatic degradation (Chian et al., 2019). Electroporation of muscle fibers by PEF and the rate of movement of intracellular constituents have been reported as responsible for proteolysis processes (Alahakoon, Faridnia, et al., 2017; Alahakoon, Oey, et al., 2017). Meanwhile, PEF pre-treatment of beef briskets, under certain processing conditions, can significantly alter their textural properties (shear force, hardness and chewiness) which results in increased tenderness and collagen solubilization after sous vide processing without having any adverse effects on protein digestibility (Alahakoon, Oey, Bremer, & Silcock, 2018).

On the other hand, a greater effect in proteolysis of beef samples was observed after treatment with 1 × PEF than increasing the number of PEF treatments (2 × and 3 ×), thus indicating a different mechanism for the tenderization including physical disruption. This may be explained by the generation of high temperature during the repeated PEF treatments in batch mode (10 kV, 90 Hz, 20 μs), which could have inactivated the endogenous proteolytic enzymes responsible for meat tenderization (Suwandy et al., 2015c). Similarly, Bekhit et al. (2016) evaluated the effects of repeated PEF treatments (1 ×, 2 ×, 3 ×) on the quality of hot-boned beef loins and topsides and found a reduction in proteolysis (based on troponin T) with every extra application. The design and optimization of pre-treatments to enhance protein digestibility is important to promote a better utilization of proteins in the gut and limit undesirable effects of protein rich products consumption.

3.5. Effects on minerals

PEF processing has been reported to significantly modify the levels of nutritionally important minerals in beef and chicken (Khan et al., 2018, 2017). This is of relevance due to meat is worldwide cataloged as a significant source of minerals, such as Fe and Zn, therefore any change in their concentrations could have negative commercial consequences. That is why more studies about the impact of PEF processing on the concentrations of different minerals in meat should be carried out in the next years.

In relation to this, Bhat, Morton, Mason, and Bekhit (2019) did not observe negative changes in the release of minerals such as Fe, K, P, Ca, Na and Mg from the meat material during in vitro gastrointestinal digestion. On the contrary, higher mathematically ($P > .05$) amounts of these minerals were detected for PEF treated samples in comparison to control, which may be due to a greater release of minerals by increasing the permeability of the membrane after PEF treatment. Previously, Khan et al. (2017) concluded that high PEF application (10 kV, 200 Hz, 20 μs) induced lower concentrations in P, K, and Fe and negatively affected the quality parameters of beef. On the other hand, this was not the case when PEF was applied at low intensities (2.5 kV, 200 Hz, 20 μs). The PEF system was used in batch mode and the meat fiber direction was parallel to the electrodes. More recently, Khan et al. (2018) focused their investigation on the consequences of low and high PEF (2.5 kV and 10 kV, respectively) on 40 macro- and micro-minerals in raw and cooked beef loins and in chicken breasts. PEF treatments caused changes in element concentrations and, interestingly, there were differential effects depending on the treated meat material (beef or chicken). Furthermore, some of the increased mineral contents suggested potential migrations from the electrodes, especially at high PEF.

3.6. Valorization of meat by-products

Gudmundsson and Hafsteinsson (2001) suggested another interesting potential use of PEF for meat products by investigating the applicability of PEF for facilitating the extraction of substances from waste material. In this context, a recent study has been carried out with the objective of recovering functional proteins from waste chicken meat by a PEF-based process (two-step protocol consisting of high voltage, short pulses followed by low voltage, long pulses, with the total invested energy of 38.4 J/g). As a result of this non-thermal, chemical-free process, a fraction enriched in proteins with possible antioxidant properties was obtained (Ghosh, Gillis, Sheviryov, Levkov, & Golberg, 2019). Likewise, animal blood is a slaughter by-product that can be collected at abattoirs in large quantities. According to this, Boulaaba, Egen, and Klein (2014) investigated the effectiveness of PEF treatment of blood for the inactivation of microorganisms and its effect on the physicochemical and sensory properties. They used a coaxial continuous treatment chamber composed by TITAN electrodes separated by a gap of 7 mm and the following conditions: EFS 11 kV/cm, 163 and 209 ms, 134 and 175 Hz and a total specific energy input of 91 and 114 kJ/kg, respectively. As a result, authors concluded that PEF can increase the shelf-life of the product and promote the color parameters by decreasing lightness (L^*) and redness (a^*) values. In the same way, porcine blood plasma was treated with PEF with the idea of improving the microbiological quality and to expand its utilization possibilities and increasing storage period (Boulaaba, Kiessling, Töpfl, Heinz, & Klein, 2014).

4. Applications of PEF in the fish industry

4.1. Impact on product quality

The utility of PEF in the food industry is gaining attention since it is a non-thermal alternative that could have more impact on the microstructure of muscle food compared to the emerging heat-based processes such as ohmic (Gavahian, Chu, & Farahnaky, 2019) and microwave heating (Farahnaky, Azizi, & Gavahian, 2012). Additionally, several authors have indicated the advantageous utility of this technique to maintain the physical, organoleptic and functional characteristics of the final product, i.e., introducing minimal changes in the flavors, vitamins, and other nutrients (Oziembłowski & Kopeć, 2005; Pourzaki & Mirzaee, 2008). In a previously conducted research, Gudmundsson and Hafsteinsson (2001) evaluated the effects of PEF on microstructure of salmon, chicken, and lumpfish roes. Their results indicated that salmon is more sensitive to mild PEF treatment (< 2 kV/cm, 20–40 pulses) than chicken, but both samples showed significant effects on their texture and microstructure, while roes withstood harder conditions.

A few years later, a different work used PEF technique with the aim of improving water holding properties of fish and the tenderization of shellfish products (Klonowski, Heinz, Toepfl, Gunnarsson, & Þorkelsson, 2006). Unfortunately, no positive effects were observed on the tenderness of shellfish products such as common whelk and Iceland cyprine, while submitting the fish muscle to PEF made its structure more porous. Authors suggested PEF treatment as a promising strategy to increase water holding properties in fish, as well as it could be potentially used as a pretreatment for fish drying.

In addition, Zhou, He, and Zhou (2017) studied the extractive effectiveness of PEF technology to obtain protein from mussel. In case of PEF application (the triangular pulse power waveform and the pulse duration of 2 μs) at the estimated optimum conditions (EFS of 20 kV/cm, pulse number of 8, and the enzymolysis time of 2 h), the extraction yield of protein was maximized to 77.08%.

4.2. Valorization of fish by-products

As in the case of meat, PEF technique also can be very interesting to be used to valorize by-products from fish processing industries (Table 3). In this line, Zhou et al. (2012) designed an improved a method to extract calcium from fishbone using high-intensity PEF. Comparing the results with ultrasonic technique, it is correct to conclude that PEF allowed reaching higher extraction efficiency in short period of times. This finding was in agreement with the negative results concerning the applicability of power ultrasound for accelerating an extraction process (Gavahian et al., 2017). Likewise, another research used the same raw material, fishbone, to extract chondroitin sulphate by high intensity PEF (He, Yin, Yan, & Yu, 2014). This study highlighted the benefits of this technique including reduced process time, enhanced efficient and eco-friendly aspects. Furthermore, He, Yin, Yan, and Wang (2017) combined PEF (exponentially decaying bipolar triangle pulse waveforms was generated with pulse duration of 2 μs) with semi-bionic extraction to optimize not only the extraction of the mentioned calcium and chondroitin sulphate from fishbone, but also of collagen. The maximum contents of the effective ingredients in the extract were obtained with EFS of 22.79 kV/cm and 9 pulses.

Another research on the valorization of fish waste was conducted by Li, Lin, Chen, and Fang (2016). They developed a PEF enzymatic-assisted extraction for isolation of the abalone viscera protein. They studied the effects of various PEF conditions, including treatment time, intensity strength, and material to solvent ratio. They observed the highest extraction yield occurred when the solvent to material ratio was 1 to 4 and the PEF was applied for 600 μs at the intensity strength of 20 kV/cm. According to the authors, the proposed extraction technique resulted in a high yield of abalone viscera protein, which possessed promising emulsifying properties, when compared to conventional enzymatic extraction methods. In contrast, the viscosity and foaming properties of the extracted product were decreased when PEF was applied (Li et al., 2016). This study highlighted the possibility of combining other technique with PEF to improve the efficiency of the process.

A schematic representation of the extraction process of hydrophilic compounds from a fish by-product (fish tail), with and without PEF pretreatment, is shown in Fig. 3. PEF can be easily applied as a pretreatment of fish waste for enhancing the yield of the extraction process.

The high initial capital investment is the main barrier that limits the application of PEF in the fish processing industry at this moment. In addition, inefficiency of this technique against the reduction of natural occurring enzymes in the fish is among the shortcomings of this emerging technology. Similar to the ohmic heating method, the electrical conductivity of the product is a crucial parameter that limits the application of PEF to materials with moderate conductivity (Gavahian, Farahnaky, Farhoosh, Javidnia, & Shahidi, 2015; van Wyk, Silva, & Farid, 2019).

5. Conclusions and future perspectives

PEF is an innovative processing technology that has not only a wide potential for preserving food products, but also for modifying their structure. In addition, it is an energy efficient and environmentally friendly alternative for food processing. With these advantages, it is expected that will be successfully used in the industry shortly, as a single treatment or in combination with other processes that synergistically improve product quality and process yields.

A task still challenging is the development of equipment with a reliable, industrial-scale generation of high-strength electric field

Table 3
Uses of pulsed electric fields (PEF) technology on fish processing industry.

Fish product	Operating conditions	Findings/Applications	Reference
Salmon and lumpfish roes	Different combinations of electric field, number of pulses and high-pressure treatment.	PEF application had greater impact on salmon than chicken samples. Appropriate technique as pretreatment for lumpfish roes.	Gudmundsson and Hafsteinsson (2001)
Pollock fillets, cod loins frozen, cod fresh fillets, haddock loins frozen, Iceland cyprine and common whelk	Electric field strength 1.2–2.0 kV/cm; frequency 1–4 Hz; pulse width 400 μs; pulse number 20, 40, 80 or 120	PEF treatment was not effective with < 90 pulses and field < 2.0 kV/cm. Improvement of the water holding properties and the fish drying.	Klonowski et al. (2006)
Fishbone	Optimal combination of parameters: EFS 25 kV/cm and pulse number 8	Effective calcium extraction	Zhou et al. (2012)
Fishbone	Processing conditions were optimized and the best yield was achieved using a EFS of 16.88 kV/cm and pulse number of 9	Extraction of chondroitin sulfate	He et al. (2014)
Fishbone	Combinations of semi-bionic extraction method with PEF (optimum conditions: EFS 22.79 kV/cm and pulse number 9)	Extraction of effective ingredients	He et al. (2017)
Mussel	Best conditions: EFS of 20 kV/cm, pulse number of 8 and enzymolysis time of 2 h	Extraction of protein	Zhou et al. (2017)
*Haliotis discus hannai Ino*viscera	Treatment time (100–800 μs), Intensity strength 5–20 kV/cm, and the ratio of material to solvent (3:1–10:1)	Extraction of Protein hydrolysate	Li et al. (2016)

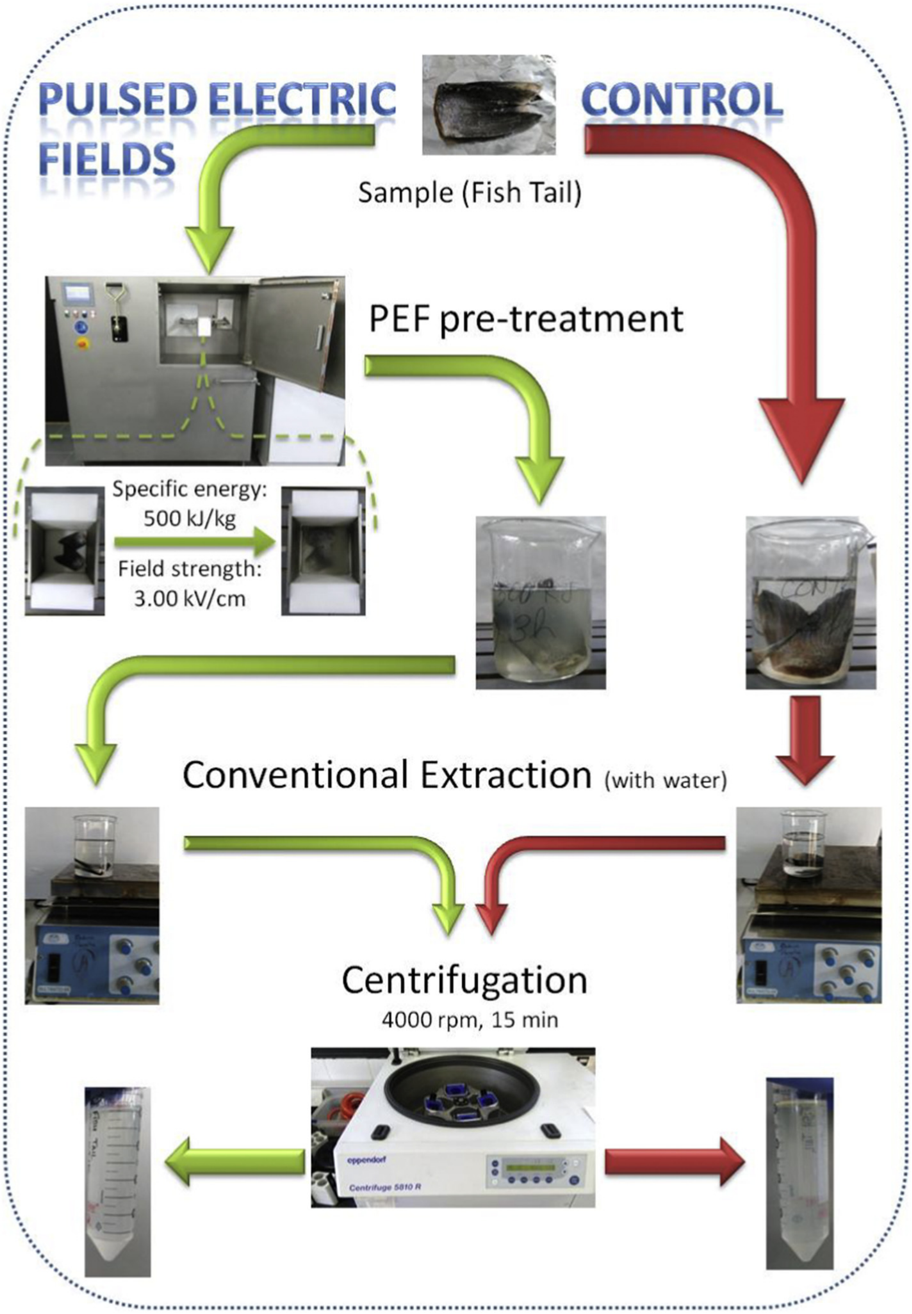

Fig. 3. Extraction process of hydrophilic compounds from a fish by-product (fish tail) with and without pulsed electric field (PEF) pre-treatment.

pulses. Raso and Heinz (2006) reported that important advances have been achieved in the commercialization of PEFs applications. However, translating the technical parameters into an affordable and effective PEF system within legal regulation is not easy. The development of user-friendly and low-cost PEF systems is an important demand for allowing its full deployment in the meat and fish industries. Another under-investigated problem of the last few years is the electrochemical reactions at the electrode/medium interfaces, indicating that there is a challenge to avoid electrode corrosion and migrations of electrode materials into food systems, maybe replacing commonly used stainless steel electrodes by other materials (Khan et al., 2018; Loeffler, 2006; Morren, Roodenburg, & de Haan, 2003; Pataro, Falcone, Donsì, & Ferrari, 2014).

The impact of processing conditions such as temperature, pH, moisture, and lipid content on the safety and quality aspects of new products leaves an area that is still open to food chemists. In addition, different PEF processing parameters, as well as meat properties and pre- and post-treatment meat conditions (freezing, aging, etc.) significantly affect the final product quality.

Finally, despite the existence of several reports on the positive aspects of PEF technology for meat and fish processing at the laboratory scales, its application in the industry is actually rather limited. This is because of several reasons such as the high capital investment (that prevents the industry from investing in PEF equipment), the introduction of changes in the conventional layouts of meat processing plant, and the need for the optimization of PEF conditions for specific product application. To date, there are limited studies regarding the customization of PEF treatments and their effects on the different quality attributes of meat and fish. The transfer of PEF technology to the fish and meat industry would be highly favorable due to the low energy consumption and short processing times required in PEF processing. However, this technology transfer requires further investigations on the impact of PEF treatment on quality parameters of meat and fish products (e.g. tenderness, color, oxidation, weight loss, and water holding capacity).

Acknowledgements

This research was supported by INIA (Spain) under Grant RTA 2017-00024-CO4-04. José M. Lorenzo and Paulo E.S. Munekata are members of the HealthyMeat network, funded by CYTED (ref. 119RT0568). Paulo E.S. Munekata acknowledges postdoctoral fellowship support from Ministry of Economy and Competitiveness (MINECO, Spain) "Juan de la Cierva" program (FJCI-2016-29486). Francisco J. Barba, Jose M. Lorenzo and Francisco J. Martí-Quijal would like to acknowledge the EU Commission for the funds provided by the BBI-JU through the H2020 Project AQUABIOPROFIT "Aquaculture and agriculture biomass side stream proteins and bioactives for feed, fitness and health promoting nutritional supplements" (Grant Agreement no. 790956). Moreover, Francisco J. Barba thanks Generalitat Valenciana for the financial support (IDIFEDER/2018/046 - Procesos innovadores de extracción y conservación: pulsos eléctricos y fluidos supercríticos) through European Union ERDF funds (European Regional Development Fund). Mohsen Gavahian would like to appreciate the support of the Ministry of Economic Affairs, project no. 108-EC-17-A-22-0171, Taiwan, Republic of China.

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