

# TiO<sub>2</sub>-Coated Electrodes for Pulsed Electric Field Treatment of Microorganisms

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**Abstract**— Pulsed electric fields (PEF) can cause irreversible damage to bio-membranes and may result in inactivation of microorganisms. The aim of this paper is to investigate the PEF treatment of the yeast *Saccharomyces cerevisiae*, using a novel treatment cell with parallel-plane electrodes coated with a 2 μm thin TiO<sub>2</sub> film. Two different PEF waveforms, square and exponential, with magnitudes of 67 kV/cm and 80 kV/cm, were used in this study. The efficacy of the PEF treatment was assessed by comparison of the surviving treated and untreated yeast populations, and it was shown that a treatment cell with TiO<sub>2</sub>-coated electrodes can be successfully used for the PEF treatment of microorganisms: 3-log<sub>10</sub> reduction in the yeast population was achieved with 100 impulses. The energy efficacy of the PEF process in the proposed treatment cell has been compared with the energy losses in the PEF treatment cell with uncoated, conductive electrodes. It is shown that the electrodes coated with TiO<sub>2</sub> provide better performance as compared with the traditional uncoated electrodes.

**Index Terms** — Pulsed electric field, TiO<sub>2</sub>-coated electrodes, inactivation of microorganisms, yeast.

## I. INTRODUCTION

INTENSIVE electric field impulses with durations from a few nanoseconds to a few tens of microseconds can generate electro-mechanical stresses in biological membranes which allow pores to be formed in the membrane [1]. Such field-induced pores become irreversible if the transmembrane potential exceeds a critical value, ~1 V [2]. Irreversible electroporation results in the dysfunction of the biological membrane and the death of the cell. Therefore, pulsed electric field (PEF) treatment can be used for non-thermal inactivation of microorganisms. In recent decades, the PEF inactivation process has been intensively investigated and this technology is used for treatment of yeast and bacteria [3 - 5], for processing of liquid foodstuffs such as juice and milk [6, 7], and for extraction of lipids from microalgae [8 -10].

Typically, the electrodes in traditional PEF systems, including commercially-available apparatuses, are made of stainless steel. The liquid being treated in such PEF systems is

in direct contact with the metallic electrodes, which creates some significant drawbacks. First, significant conduction current, as high as a few hundreds of Amperes, can flow through the liquid during PEF treatment [11, 13]. As a result of Joule heating [5, 13], the treated liquid should be cooled to maintain a desirable temperature during the PEF treatment. Energy losses due to Joule heating and cooling imply significant energy consumption of the PEF process. Secondly, the direct contact between the liquid and the electrodes can cause undesirable electro-chemical reactions during the treatment. Metallic particles can be introduced into the liquid and other toxic reactive chemical species such as hydrogen peroxide can also be produced [14]. Moreover, air bubbles can be generated at the interface between the liquid and the metallic electrodes [15], which increases the probability of undesired dielectric breakdown in the PEF treatment cell.

A conceptually new treatment cell with dielectric electrodes is discussed in [16] and [17]: it is proposed to cover the electrodes of the cell with a layer of dielectric material with a high permittivity. Thus, the direct contact between the metal surface and the liquid is prevented, and conduction current through the liquid is eliminated. This concept was proven theoretically in [16] and tested experimentally in [17]. The results presented in [17] showed that a treatment cell with dielectric electrodes could be successfully used for inactivation of the bacterium *Escherichia coli*, while maintaining a relatively low sample temperature and low energy consumption. However, the maximum field magnitude which was tested in [17], 200 kV/cm, is significantly higher than a typical field used in the PEF treatment, ~ 30 kV/cm. Also, no comparison between the PEF performance achieved in the novel and traditional PEF treatment cells was made.

In the present study, a treatment cell with metallic electrodes coated with a 2-μm thick TiO<sub>2</sub> film has been developed. Titanium dioxide can form a stable dielectric ceramic in low temperature condition [18, 19]. It was reported in [18] that the dielectric constant of TiO<sub>2</sub> is ~ 95 at room temperature. It was also shown in [20] that the conductivity of a dry, thin TiO<sub>2</sub> film is ~ 5×10<sup>-7</sup> μS/cm. However, as found in the present study, when TiO<sub>2</sub> film is in contact with water, its conductivity increases significantly, and an ionic current can also flow through the liquid when treated with PEF impulses. Nevertheless, the magnitude of this conduction current is lower than the magnitude of the current in the case of metallic

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electrodes. Therefore, the  $\text{TiO}_2$ -coated electrodes potentially can be considered for design and development of a low-conductivity PEF treatment cell.

PEF treatment of the yeast *Saccharomyces cerevisiae* has been conducted using this  $\text{TiO}_2$ -coated treatment cell. In order to provide a direct comparison between this  $\text{TiO}_2$ -coated and traditional metallic PEF treatment cells, the PEF treatment procedure was identical to the procedure used in [12], where the same microorganisms were subjected to pulsed electric fields in a treatment cell with uncoated, conductive electrodes. In [12], voltage impulses with two different waveshapes, square and exponential, and two peak electrical-field magnitudes, 67 kV/cm and 80 kV/cm, were used. In order to make comparison with this test cell, identical field strengths and waveshapes were used in the present study. The inactivation kinetics and energy consumption for the PEF process performed in the treatment cell with  $\text{TiO}_2$ -coated electrodes have been obtained. The results of PEF inactivation obtained using the treatment cell with  $\text{TiO}_2$ -coated electrodes are compared with recently published results [12]: the results of the PEF treatment obtained using the treatment cell with uncoated conductive electrodes, and the same high-voltage (HV) waveforms.

## II. EXPERIMENTAL PROCEDURES

### A. PEF Treatment Cell with Coated Electrodes

The topology of the PEF treatment cell used in the present study is shown in Fig. 1. In this treatment cell, 50 mm  $\times$  50 mm square metallic plates, coated with a 2- $\mu\text{m}$   $\text{TiO}_2$  thin-film, were used as parallel-plane electrodes. The inter-electrode gap was maintained by a 1-mm-thick Polytetrafluoroethylene (PTFE) spacer. The electrodes and the spacer were clamped tightly together by nylon screws in order to seal the treatment cell. The treatment cell was filled with the yeast suspensions through its open top.

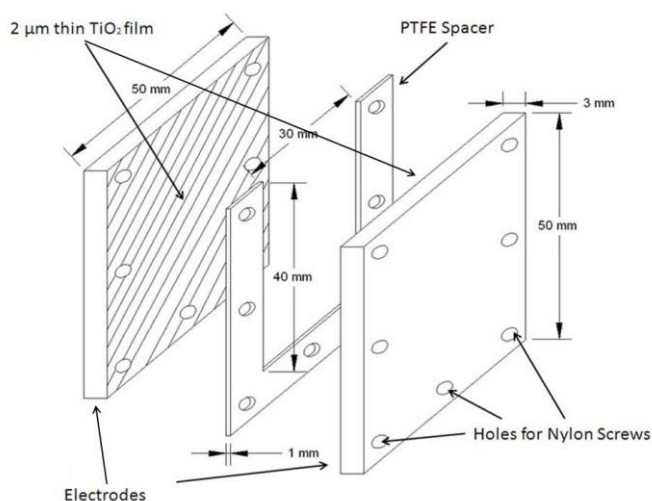


Fig. 1 Schematic diagram of the PEF treatment cell with  $\text{TiO}_2$ -coated electrodes.

### B. PEF System and Voltage Waveforms

Under atmospheric pressure, an electrical breakdown event is likely to occur across the top liquid-air interface, due to the short (1 mm) gap between the electrodes. To avoid this undesirable breakdown, the PEF treatment cell was placed inside a sealed metallic chamber, described in [12], which was pressurised up to 7.5 bar (absolute pressure) with compressed air. The elevated air pressure suppresses the development of interfacial spark breakdowns.

Two different voltage waveforms, square and exponential, were used in this PEF study. An impulse generator based on a transmission-line topology (SAMTECH Ltd., UK [8]), was used to generate square HV impulses. This impulse generator was used to produce negative impulses with magnitude of 30 kV and duration of 1  $\mu\text{s}$ .

A capacitive pulsed-power system was built to generate exponential HV impulses. This system consisted of a HVDC charging unit (Glassman, EH50P2), a protective resistor of 28 M $\Omega$ , a 40-nF HV capacitor (nominal capacitance), a self-breaking plasma closing switch, and a 2-m length of URM67 coaxial cable, which connects the output terminal of the capacitor with the PEF treatment cell. The magnitude of the voltage across the PEF treatment cell was regulated by the inter-electrode gap distance of the switch, and the pulse repetition rate was controlled by the current regulator of the HVDC source. A 50- $\Omega$  resistor was connected in parallel with the PEF treatment cell to match the impedance of the pulse generator. A Tektronix P6015A HV probe with a bandwidth of 75 MHz was used to measure the voltage across the PEF treatment cell, and a Pearson 6585 current monitor was used to measure the current through the PEF treatment cell. Both voltage and current waveforms were recorded using a Tektronix TDS2024 digital storage oscilloscope with a bandwidth of 200 MHz and a sampling rate of 2 GS/s. Fig. 2 shows the typical voltage waveshapes across the treatment cell during PEF treatment. The current waveforms for both types of applied HV impulses will be discussed in Section III-A.

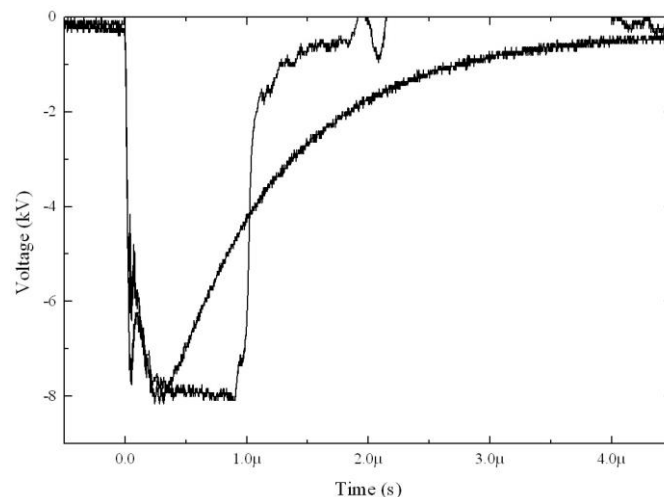


Fig. 2. Exponential and square voltage waveforms used to stress the PEF cell.

### C. Preparation of the Yeast Sample

The yeast used in the paper was *S. cerevisiae* (MUCL 28749), obtained from The Belgian Co-ordinated Collections of Microorganisms. The yeast was cultured in 100 ml 2% (w/v) malt extract broth (Oxoid Ltd, UK) at 30°C in a shaking incubator (120 rpm). After 20 hours incubation, the *S. cerevisiae* culture was centrifuged at 4300 rpm for 10 minutes and re-suspended in 100 ml 0.05% (w/v) mycological peptone (Oxoid Ltd, UK). The population density of the resulting yeast suspension was  $\sim 10^7$  CFU/ml. The yeast suspension was then serially diluted 0.05% (w/v) with mycological peptone to prepare samples with population densities of  $10^4$  to  $10^7$  CFU/ml.

### D. Assessment of PEF Performance

Yeast suspensions were treated in the PEF treatment cell with TiO<sub>2</sub>-coated electrodes. *S. cerevisiae* samples were subjected to 25, 50 and 100 HV impulses which had square and exponential waveforms, with peak magnitudes of 67 kV/cm and 80 kV/cm. The pulse repetition rate was 1 pulse per second (pps) for all tests. In order to exclude any potential antimicrobial effect from the electrode itself, a control group of sample was also used. In this group, the samples were placed inside the treatment cell for the same period of time without PEF treatment, to detect any antimicrobial effect. The treated and untreated (control) yeast samples were plated onto malt extract agar (Oxoid Ltd, UK) using spiral- and spread-plating methods, as detailed in [12]. Agar plates were then incubated at 30 °C for 20 hours. Post-incubation, the surviving populations of the treated and untreated (control) *S. cerevisiae* samples were enumerated and compared to assess the effect of the PEF treatment. The mean of the yeast populations (log<sub>10</sub> CFU/ml) and its standard deviation have been obtained from triplicate independent PEF tests ( $n=3$ ), with each test using a fresh yeast culture.

## III. RESULTS AND DISCUSSION

### A. Current Waveform

Fig. 3 and Fig. 4 showed the current waveforms obtained during PEF treatments using the square impulse and exponential impulse respectively. As can be seen from both figures, there is a noticeable ionic conduction current in the case of both PEF treatment cells, conductive (uncoated) and TiO<sub>2</sub>-coated cells. It was found that the conduction current through the treated liquid was reduced by  $\sim 23\%$  with TiO<sub>2</sub>-coated electrodes as compared with the conduction current with the stainless steel treatment cell.

### B. Temperature Effects

The temperature of the yeast suspension was measured using a K-type thermocouple before and after PEF treatment, in order to identify any notable thermal effects. The maximum registered change (increase) in the temperature of the yeast samples treated in the test cell with TiO<sub>2</sub>-coated electrodes was only  $\sim 0.5$  °C. As a comparison, an increase of  $\sim 1.5$  °C in the sample temperature was observed after PEF treatment of a yeast suspension in a test cell with uncoated, stainless steel

electrodes using the same treatment parameters, i.e. the same field magnitude, pulse waveshape and pulse repetition rate [12]. As the Joule heating of the liquid sample is directly related to the conduction current, the lower temperature increase of the sample treated in the TiO<sub>2</sub>-coated treatment cell can be explained by the lower conduction current through the sample. Low electrical conductivity of the yeast solutions used in the present study is also an important factor which helps to minimise the thermal effect of PEF treatment [8, 12]. In contrast, in the case of highly conductive solution, an increase of a few tens of °C in temperature was reported [5, 13]. Therefore, the lower temperature change observed in the present paper ( $\sim 0.5$  °C) is likely to be explained by the use of less conductive TiO<sub>2</sub>-coated electrodes and the low conductivity sample solution.

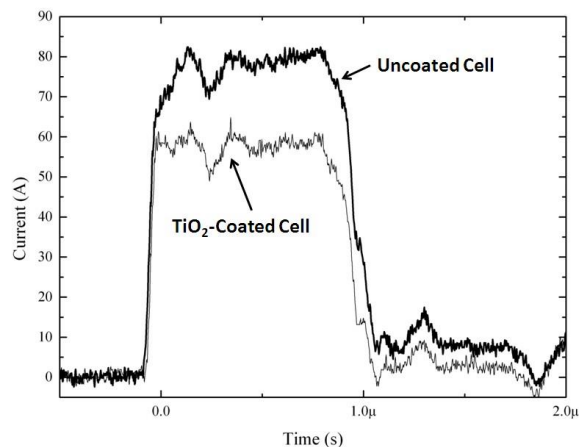


Fig. 3. Current waveform obtained during PEF treatment using exponential impulse. Dark line: Current waveform in the treatment cell with stainless steel electrodes; Grey line: Current waveform in the treatment cell with TiO<sub>2</sub>-coated electrodes.

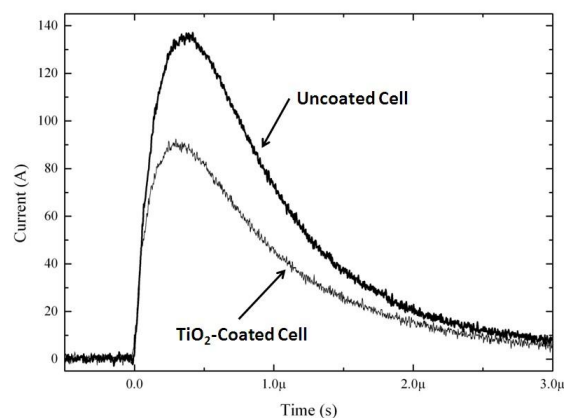


Fig. 4. Current waveform obtained during PEF treatment using exponential impulse. Dark line: Current waveform in the treatment cell with stainless steel electrodes; Grey line: Current waveform in the treatment cell with TiO<sub>2</sub>-coated electrodes.

### C. PEF Inactivation of Yeast in the Treatment Cell with TiO<sub>2</sub>-Coated Electrodes

The results of the PEF treatment of yeast suspensions ( $\sim 10^7$  CFU/ml and  $\sim 10^4$  CFU/ml) with TiO<sub>2</sub>-coated electrodes are shown in Fig. 5 and Fig. 6. These figures demonstrate that both types of impulses (square and exponential) lead to notable reduction in the yeast population. Control population, which

were held in the TiO<sub>2</sub>-coated test cell but not subjected to electrical fields, were unchanged compared to the untreated starting populations, thus, confirming that direct contact with the TiO<sub>2</sub> coating alone had no antimicrobial effect.

It was found that the PEF treatment with the higher field strength, 80 kV/cm, provides ~ 1-log<sub>10</sub> greater reduction of the yeast population as compared with 67 kV/cm treatment, for all initial populations. The energy efficacy of the PEF treatment in the treatment cell equipped with TiO<sub>2</sub>-coated electrodes is discussed in Section III-F.

The results shown in Fig. 5 and Fig. 6 demonstrate that PEF treatment using the exponential waveform provides a greater inactivation effect as compared with the PEF treatment using the square waveform. A similar tendency was reported in [12], where it was shown that exponential HV impulses resulted in a higher PEF efficacy as compared with square impulses. It was suggested that the superior performance of the exponential waveform could be a result of its longer effective duration, during which the field in the treatment cell exceeds the critical inactivation field value, ~ 10 kV/cm, [12].

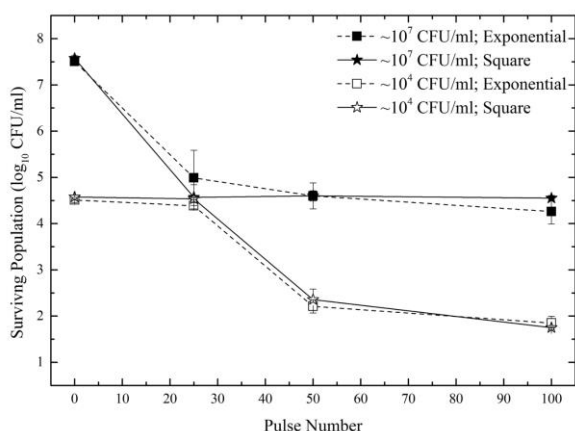


Fig. 5. Surviving populations of yeast (log<sub>10</sub> CFU/ml) as a function of pulse number for 67 kV/cm waveforms: square waveform (stars and solid line) and exponential waveform (squares and dashed line). Error bars show standard deviation ( $n=3$ ).

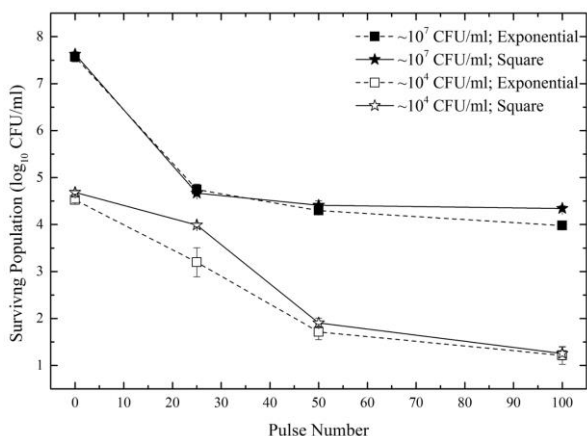


Fig. 6. Surviving populations of yeast (log<sub>10</sub> CFU/ml) as a function of pulse number for 80 kV/cm waveforms: square waveform (stars and solid line) and exponential waveform (squares and dashed line). Error bars show standard deviation ( $n=3$ ).

#### D. Tailing Effect

The PEF inactivation results presented in Fig. 5 and Fig. 6 demonstrate that after a 3-log<sub>10</sub> population reduction (for all initial yeast populations and for all applied HV waveforms), further increase in the number of applied impulses leads to a ‘tailing’ effect in the inactivation curves. This undesirable phenomenon was also reported in [12, 13] and in [21]. In [12], the tailing effect was examined using different impulse waveforms and different initial populations of the yeast, in order to identify potential causes of this effect. It was found that there was a sedimentation of the yeast cells during a ~ 20-min period associated with pressurising/de-pressurising the test chamber. Conducting PEF treatment and re-mixing of the samples after each 100 impulses allowed further reduction in the yeast population to be achieved. The same approach was used in the present study, and it was shown that a further 3-log<sub>10</sub> reduction in the *S. cerevisiae* population was achieved using this re-mixing process (Fig. 7). Therefore, continuous flow of the liquid through the PEF treatment cell may help to eliminate the tailing effect, as was shown in [22]. However, no direct comparison between batch and continuous PEF processes has been made. As suggested in [12], other factors such as potential penetration of liquid in the gaps between the electrodes and the dielectric spacer (areas with a reduced electric field), and clumping of microorganisms, might also contribute to the tailing effect in the inactivation process.

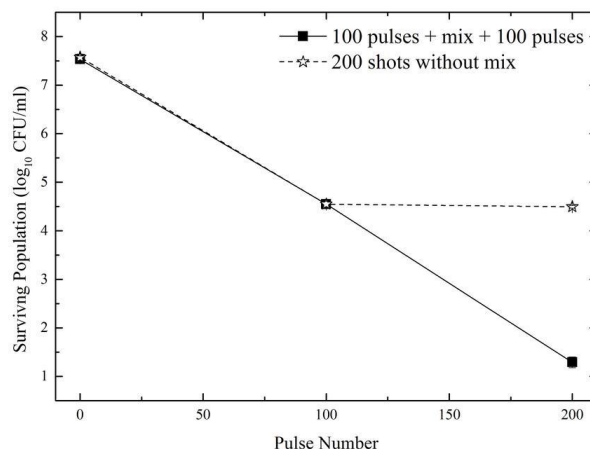


Fig. 7. Surviving populations of yeast in the case of the batch PEF treatment (dashed line, star symbols) and PEF treatment with re-mixing procedure (solid line, square symbols). Field strength: 67 kV/cm; square waveform. Error bars show standard deviation ( $n=3$ ).

#### E. Comparison of PEF Inactivation of Yeast in the Treatment Cell with TiO<sub>2</sub>-Coated Electrodes and Uncoated Electrodes

As discussed in Section III-B, the electrodes coated with TiO<sub>2</sub> film resulted in lower Joule heating of the liquid as compared with a treatment cell with uncoated conductive electrodes [12]. In this section, the results of the PEF treatment of yeast in the treatment cell with TiO<sub>2</sub>-coated electrodes (Fig. 1) are compared with those following PEF treatment of the same strain of yeast in a treatment cell with uncoated conductive electrodes, published in [12]. The same initial populations of yeast were used in both the present study and in [12]. The PEF inactivation curves for two initial populations

$\sim 10^7$  CFU/ml and  $\sim 10^4$  CFU/ml were used for comparison of the PEF performance.

The inactivation curves obtained for PEF treatment of yeast suspensions with TiO<sub>2</sub>-coated electrodes, and with both square and exponential waveforms, are shown in Fig. 8 (67 kV/cm) and Fig. 9 (80 kV/cm). For ease of comparison, the results from [12], using uncoated electrodes, are also displayed. Fig. 8 and Fig. 9 show that the treatment cell with TiO<sub>2</sub>-coated electrodes resulted in a better PEF performance as compared with the treatment cell with uncoated, conductive electrodes: half of the number of impulses were required to achieve an  $\sim 3\text{-log}_{10}$  population reduction in the new treatment cell with TiO<sub>2</sub>-coated electrodes as compared with the traditional treatment cell with uncoated electrodes.

An analysis of the energy dissipation processes in both treatment cells is presented in Section III-F; this analysis confirms that the treatment cell with TiO<sub>2</sub>-coated electrodes resulted in lower Joule energy losses as compared with the treatment cell with uncoated electrodes providing the same level of inactivation.

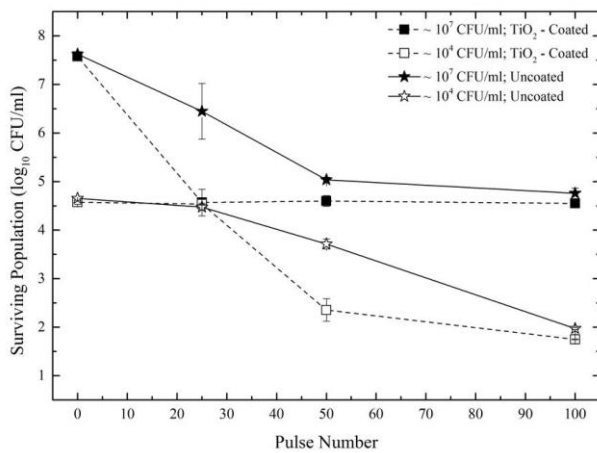


Fig. 8. Surviving populations of yeast ( $\log_{10}$  CFU/ml) as a function of pulse number for 67 kV/cm square waveforms: uncoated electrodes (stars and solid lines, results are from [12]); TiO<sub>2</sub>-coated electrodes (squares and dashed lines). Error bars show standard deviation ( $n=3$ ).

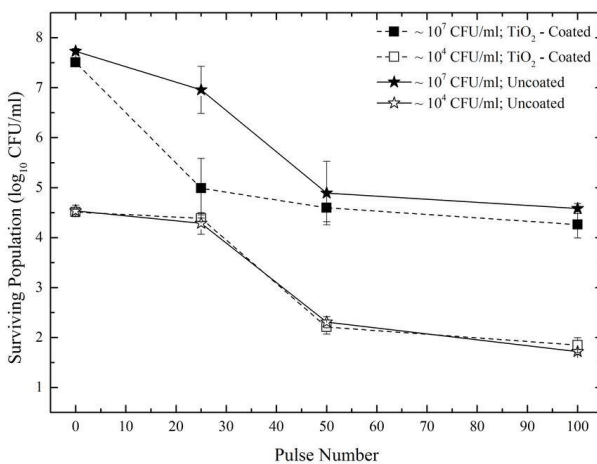


Fig. 9. Surviving populations of yeast ( $\log_{10}$  CFU/ml) as a function of pulse number for 80 kV/cm exponential waveforms: uncoated electrodes (stars and solid lines, results are from [12]); TiO<sub>2</sub>-coated electrodes (squares and dashed lines). Error bars show standard deviation ( $n=3$ ).

### F. Specific Energy Consumption

The thin TiO<sub>2</sub> film acts as a barrier, which limits the ionic conduction current through the bulk of the liquid sample as compared to the treatment cell with uncoated stainless steel electrodes. As shown in Fig. 3 and Fig. 4, a reduced conduction current passes through the bulk of the sample treated with PEF impulses. Therefore, the energy dissipated during the PEF treatment in the cell with TiO<sub>2</sub>-coated electrodes is associated with the losses due to two transient processes: the polarisation process which is manifested by the appearance of the displacement current, and the ionic conduction process which results the Joule energy loss in the treated samples.

The specific energy which is dissipated in the treatment cell during each impulse due to the fast-changing voltage,  $E_{pol}$ , can be calculated using (1) - (3):

$$E_{pol} = R_{ESR} \cdot \int \left( C \frac{dV(t)}{dt} \right)^2 dt \quad (1)$$

$$R_{ESR} = \tan \delta / (2\pi f C) \quad (2)$$

$$C = \frac{\epsilon_r \epsilon_0 A}{d} \quad (3)$$

where  $R_{ESR}$  is the equivalent series resistance of the capacitor in the lumped-circuit model;  $C$  is the equivalent capacitance of the sample;  $V(t)$  is the voltage across the treatment cell;  $\tan \delta$  is the frequency-dependent dissipation factor (the ratio of the imaginary and real parts of the permittivity of the suspension);  $f$  is the frequency that corresponds to the rise- and fall-times of the impulse;  $\epsilon_r$  is the relative permittivity of the sample;  $\epsilon_0$  is the permittivity of free space;  $A$  is the surface area of the electrodes; and  $d$  is the inter-electrode distance.

Analysis of the waveforms shown in Fig. 2 provides an estimation for the frequencies,  $f$ , associated with the rise- and fall-time of the voltage for both waveforms. It was found that, for the square waveform,  $f$  is  $\sim 4$  MHz, and for the exponential waveform,  $f$  is  $\sim 0.75$  MHz. The dissipation factor,  $\tan \delta$ , for the yeast suspension can be modelled using the dissipation factor of water at 25 °C [23]. For the obtained frequencies, this dissipation factor is  $1.94 \times 10^{-4}$  for the square waveform and  $3.64 \times 10^{-5}$  for the exponential waveform.

Using (1) - (3) and the practical voltage waveforms obtained during the PEF treatment,  $V(t)$ , the energy losses associated with the polarisation process and fast-changing voltage have been calculated, and are given in Table I.

TABLE I  
ENERGY LOSSES ASSOCIATED WITH POLARISATION PROCESS

Waveform	Field Strength	Initial Population (CFU/ml)	
		$10^7$	$10^4$
Square	67 kV/cm	0.058mJ	0.055mJ
	80 kV/cm	0.072mJ	0.069mJ
Exponential	67 kV/cm	0.094mJ	0.096mJ
	80 kV/cm	0.152mJ	0.152mJ

Another mechanism of energy loss in the treatment cell with TiO<sub>2</sub>-coated electrodes is the Joule energy dissipation due to ionic conduction. The total specific energy consumption is the combination of polarisation loss and the ionic conduction loss. This total energy losses per pulse,  $E_{total}$  (specific energy delivered to the yeast suspension by each impulse), can be calculated as:

$$E_{total} = \int V(t) \cdot I(t) \cdot dt \quad (4)$$

where  $V(t)$  is the voltage across the treatment cell and  $I(t)$  is the current through the treatment cell. The total energy losses have been calculated using the experimental voltage and current waveforms for both impulses and these losses are given in Table II.

TABLE II  
TOTAL ENERGY LOSSES PER PULSE

Waveform	Field Strength	Initial Population (CFU/ml)	
		10 <sup>7</sup>	10 <sup>4</sup>
Square	67 kV/cm	394 mJ	327 mJ
	80 kV/cm	641 mJ	459 mJ
Exponential	67 kV/cm	401 mJ	299 mJ
	80 kV/cm	544 mJ	409 mJ

Analysis of Tables I and II shows that the energy losses due to the polarisation process (55-152 μJ) only accounted a very small portion of the total energy loss (which are in the range 299 - 641 mJ). Therefore the energy dissipation during PEF treatment in the test cell with TiO<sub>2</sub>-coated electrodes is mainly due to the ionic conduction through the liquid sample. However, these conduction losses are lower than the conduction energy losses reported in [12] for the treatment cell with uncoated electrodes, which were in the range of 0.6-1.7 J.

Table II also shows that the energy delivered by a single impulse increased with the increase in the initial population of the yeast due to an increase in conductivity of the yeast solution: the conductivity of the 10<sup>7</sup> CFU/ml samples is ~ 182 μS/cm while the conductivity of the 10<sup>4</sup> CFU/ml samples is ~ 124 μS/cm. As a result, the ionic conduction current is higher in the case of 10<sup>7</sup> CFU/ml samples than that of the samples with 10<sup>4</sup> CFU/ml. Therefore, the energy losses due to the conduction current are higher in the case of higher concentration of the yeast samples. Table II demonstrates that the energy losses are higher for higher electric field, also there is a tendency in decrease in the energy losses for exponential impulses as compared with the square impulses.

Comparison of the energy losses in the treatment cell with TiO<sub>2</sub>-coated electrodes obtained in the present study and in the treatment cell with uncoated electrodes has been made. The data for the treatment cell with uncoated electrodes are taken from [12], where the same HV impulses and the same strain of yeast were used. The results of this analysis are shown in Fig. 10 and Fig. 11, which demonstrate a reduction in the surviving population of the yeast for both treatment cells as a function of the specific energy. Figs. 10-11 show similar PEF inactivation

tendencies for both types of electrodes: the surviving population of the yeast reduces as the specific energy increases. However, in the case of TiO<sub>2</sub>-coated electrodes, the energy losses which are associated with the same log<sub>10</sub> reduction in the yeast population are significantly lower as compared with the uncoated electrodes. The main reason for this reduction in the energy losses is lower conduction current though the yeast suspension in the test cell with TiO<sub>2</sub>-coated electrodes. Therefore, this result suggests that the main mechanism of PEF inactivation is a non-thermal process, which potentially involves electro-mechanical disruption of the yeast membrane. Potentially further reduction in the specific energy consumption could be achieved if completely non-conductive electrodes are used. Such design of the PEF treatment cell was implemented in [17], where only displacement current during the PEF process was observed.

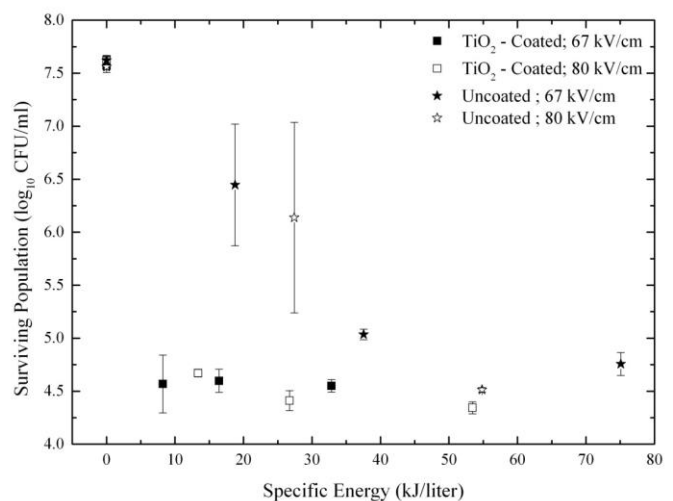


Fig. 10. Energy efficacy of the PEF treatment using the square waveform. Squares, TiO<sub>2</sub>-coated electrodes; stars uncoated electrodes (data from [12]). Solid points, 67 kV/cm impulses; open points, 80 kV/cm impulses. Initial population of yeast was 10<sup>7</sup> CFU/ml. Error bars show standard deviation (n=3).

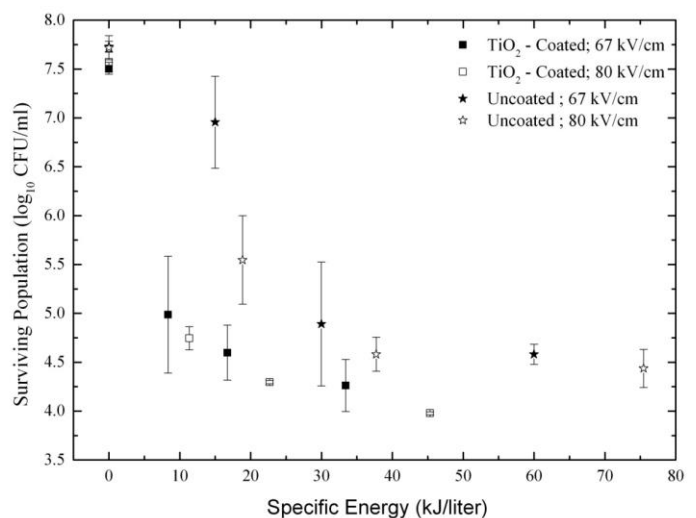


Fig. 11. Energy efficacy of the PEF treatment using the exponential waveform. Squares, TiO<sub>2</sub>-coated electrodes; stars uncoated electrodes (data from [12]). Solid points, 67 kV/cm impulses; open points, 80 kV/cm impulses. Initial population of yeast was 10<sup>7</sup> CFU/ml. Error bars show standard deviation (n=3).

## IV. CONCLUSION

The present study investigated the PEF inactivation of the yeast *S. cerevisiae*, using a treatment cell with TiO<sub>2</sub>-coated electrodes. The TiO<sub>2</sub> coating acts as a barrier, reducing the ionic conduction current in the system. Two types of impulsive waveform, square and exponential, with peak electrical-field magnitudes of 67 kV/cm and 80 kV/cm, were used. The results obtained in this work show that a treatment cell with TiO<sub>2</sub>-coated electrodes provides similar or better PEF inactivation performance as compared with uncoated, conductive electrodes. Moreover, it was demonstrated that TiO<sub>2</sub>-coated electrodes result in a significant reduction in the energy losses during the treatment. Thus, this paper demonstrates that there are clear advantages in the use of electrodes coated with a thin less-conductive film in the PEF process.

It was also found that the tailing effect (reduction in the efficacy of the PEF treatment with an increase in number of the applied impulses), which was evident in the initial inactivation kinetics, can be minimised by mixing the yeast suspension after each 100 HV impulses. A flowing system could be employed to eliminate this 'tailing' effect in future system design. These results are similar to the results obtained in [12] for the PEF process in a treatment cell with uncoated, conductive electrodes. The PEF performances of square and exponential waveforms were investigated for comparison, and it was shown that the exponential waveform, which has a longer effective duration, provides a better PEF performance as compared with the square waveform. This result confirms a similar finding reported in [12], for PEF treatment with uncoated electrodes.

The results of the PEF treatment of yeast in a treatment cell with TiO<sub>2</sub>-coated electrodes demonstrate that such electrodes can be successfully used in the design of PEF reactors for practical applications. Electrodes coated with a thin less-conductive film lead to substantial reduction in the energy losses due to Joule heating, and provide similar PEF inactivation performance as compared with uncoated, conductive electrodes. Therefore, further reduction in energy consumption could potentially be achieved if completely non-conductive electrodes are used. Further investigation of the PEF process using this novel approach is required in order to establish the PEF efficacy for other types of microorganism and energisation regimes.

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