Effect of Low-Intensity Pulsed Electric Field Treatment on Lycopene, Ascorbic acid, and Color of Tomato Juice

ABSTRACT

Aims: This study aimed to evaluate the effect of low-intensity pulsed electric field (LIPEF) treatment on the lycopene content, ascorbic acid retention, and color enhancement of juice prepared by using Roma variety tomatoes.

Study design: An experimental one-way ANOVA research design was employed, involving the random selection of Roma variety tomato samples from the regional vegetable research station for LIPEF treatment. The treated tomatoes were then analyzed for lycopene, ascorbic acid, and color.

Place and Duration of Study: Department of Food Processing Technology and Department of Food Safety and Quality Assurance, College of Food Processing Technology and Bio-energy, Anand Agricultural University, Anand, Gujarat, India between September 2022 and March 2023.

Methodology: The tomato slices were kept in the PEF treatment chamber of multipulse mode PEF treatment. They were subjected to electric field strength ranges of 2, 4, 6, and 8 kV/cm and pulse number 10, 30, 50, and 70 with a monopolar pulse of 1 Hz frequency having a pulse on/off times respectively of 10 µs. The tomato slices and water were placed together in the PEF chamber connected to the electrodes. A uniform sample of 300 g with the addition of 300 mL water was kept in a batch for treatment. After the PEF treatment, the slices were processed to extract the juice for further analysis. Then the slices are subjected to a single screw fruit juice extractor and homogenized in a food processor for uniform sampling. Fruit juice was collected in an amber-colored glass bottle to avoid loss of lycopene and color degradation and juice was analyzed to determine different physico-chemical characteristics

Results: The study revealed significant findings regarding the effect of LIPEF treatment on Roma variety tomatoes. Treatment with 4 kV/cm and 50 pulses resulted in a remarkable 69.11% increase in lycopene content compared to untreated tomato juice. Additionally, there was a remarkable retention rate of 99.84% for ascorbic acid in the treated samples compared to the control. Furthermore, LIPEF treatment led to a substantial increase of 40.78% in the a* value, indicating enhanced red coloration of the extracted juice compared to the control.

Conclusion: The findings of this study demonstrate the effectiveness of low-intensity pulsed electric field (LIPEF) treatment in enhancing the lycopene content, retaining ascorbic acid, and improving the red coloration of Roma variety tomatoes. LIPEF treatment has the potential to be utilized in food processing industries to improve the quality and nutritional value of tomato-based products. Further research and optimization of LIPEF parameters are warranted for practical application in tomato processing industries.

Keywords: Tomato, Lycopene, LIPEF, Electric field, Pulse number

1. INTRODUCTION

Tomato (*Solanum lycopersicum*) is a major vegetable consumed around the world, either fresh or processed in various forms including juices, pastes, purees, sauces, soups, and beverages [1]. They can either used in dried form using dehydration systems or can be used to extract color and lycopene which can be incorporated either for color as replacement in soft serve ice cream-like products. Lycopene is said to be a protective food due to its extensive production as well as its unique nutritional significance. It is known to be a valuable source of nutrients and bioactive substances, including vitamins, carotenoids, and phenolic compounds, that are helpful for human health [2]. Lycopene, the principal coloring substance in tomato, is a potent antioxidant and can quench singlet oxygen is twice that of β-carotene. It has been suggested that consuming lycopene-rich products may protect against cardiovascular diseases and cancer [3]. In several studies, bioactive compounds, including ascorbic acid and lycopene, were found to be degraded during processing, lowering their nutritional value [4-5]. Additionally, the perception and acceptance of consumers may be impacted by the color changes that take place during the manufacturing and storage process of tomato-based products. The researchers have been investigating the application of different emerging technologies in the food industry to enhance the nutritional value and quality of tomato-based products.

There are several extraction methods used to extract lycopene, including supercritical fluid extraction, microwave-assisted extraction, pressurized liquid extraction, and ultrasound-assisted extraction [6-8]. While these methods have their own advantages and limitations, pulsed electric field (PEF) has several unique benefits that make it a promising option for enhancing the extraction of lycopene. The food sample is subjected to high or low-intensity electric field pulses throughout this process for a short period in a processing chamber [9]. It has been shown that PEF treatment with different electric conditions, such as the electric field strength and the pulse number, can provide different benefits and can be used for multiple applications. Treating plant tissue with a PEF causes the cell membranes to rupture, resulting in increased permeability of the cell walls. Therefore, bioactive compounds are extracted more effectively [10].

PEF involves subjecting the food sample to a high or low-intensity PEF for a short period, typically microseconds. The application of high intensity pulsed electric field (HIPEF) to tomato-based products can improve their nutritional value, extend their shelf life, and also increases the juice yield, whereas low-intensity pulsed electric field (LIPEF) provides enhanced extraction of bioactive compounds, such as lycopene, from tomato-based products [11]. It can also help to preserve the color of tomato-based products by reducing the degradation of carotenoids during processing. This technology serves as a pre-treatment method in several food processes, including food dehydration, extraction, enzyme inactivation, and reduction of pesticide residues [12-13]. Hence, the aim of this study was to assess the impact of LIPEF strength and pulse number on tomato juice of locally available tomato variety (Anand Roma).

2. MATERIAL AND METHODS

2.1 Raw material

Fresh and ripe tomatoes of the Anand Roma variety were procured from the Vegetable Research Station at Anand Agricultural University, Anand, India. The tomatoes were washed with tap water to eliminate surface dirt from tomatoes, and surface was wiped to remove water. The washed tomatoes were kept in a crate and stored at a temperature of 7 ± 2 ˚C in a temperature controlled deep freezer (Electroquip, OhmkarEquipments, India).

2.2 Equipment

The EPULSES®- LBM3A-20 pulse generator from Energy Pulse Systems, Portugal which utilizes a voltage of 400 V (three phases) to power a 9000 W has an efficiency of over 90% (Fig 1). The pulse generator is connected to a treatment chamber measuring 10 cm x 10 cm x 10 cm, and fitted with two electrodes. The treatment chamber consisted of two parallel plates of rectangular electrodes with electrode gap varying 2.5-10 cm in a batch treatment chamber.

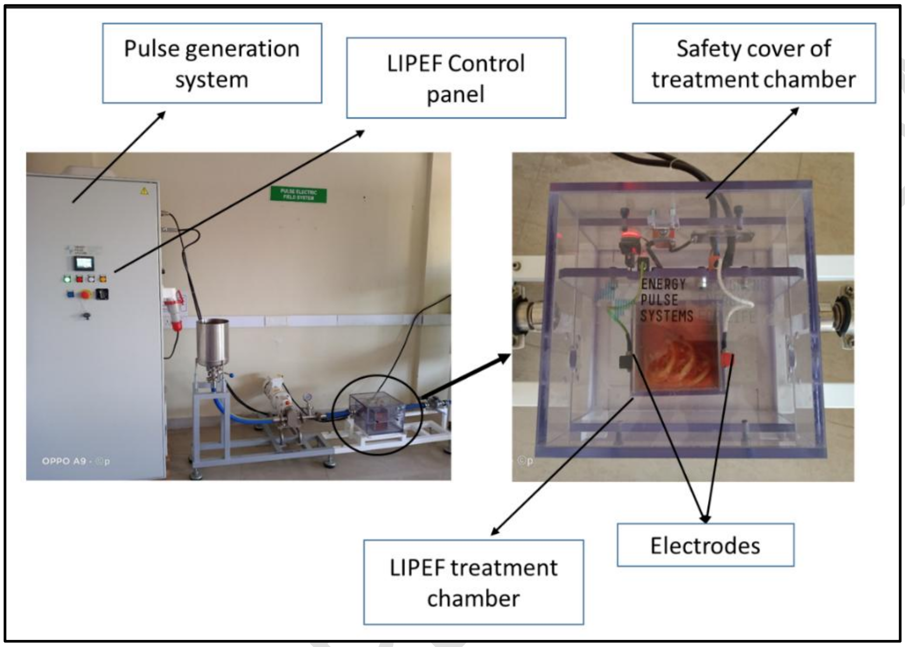

Fig. 1. Low intensity pulsed electric field system

2.3 PEF Treatment

The tomatoes were cut using cutter (J23A model Fruit cutter, Khera Equipment) to obtain uniform slices of 1 cm thickness. The cut tomato slices were kept in the LIPEF treatment chamber of multipulse mode LIPEF treatment. They were subjected to electric field strength ranges of 2, 4, 6, and 8 kV/cm and pulse number 10, 30, 50, and 70 with a monopolar pulse of 1 Hz frequency having a pulse on/off time respectively of 10 µs. The tomato slices and water were placed together in the LIPEF chamber connected to the electrodes. A uniform sample of 300 g with the addition of 300 mL water was kept in a batch for treatment. After the LIPEF treatment, the slices are subjected to a single screw fruit juice extractor and homogenized in a food processor for uniform sampling. Fruit juice was collected in an amber-colored glass bottle to avoid loss of lycopene, and color degradation and juice was analyzed to determine different physico chemical characteristics.

2.4 Determination of lycopene content

The lycopene content of the samples was estimated using the following protocol [14]. Initially, a sample weighing 5-10 g was extracted several times using acetone in a pestle and mortar. Subsequently, the acetone extract was gently mixed with petroleum ether (10-15 mL) in a separating funnel. The addition of distilled water to the acetone layer facilitated the transfer of the carotenoid pigment to the petroleum layer. The petroleum layer was then separated and transferred to a 50 mL volumetric flask and filled with petroleum ether until the mark was reached. A 5 mL aliquot of this solution was diluted with petroleum ether to 50 mL, and its absorbance was analyzed at 503 nm using petroleum ether as a blank in a Systronics UV-Vis spectrophotometer 119 and the results were expressed in mg/100g

$$mglycopeneper\ 100\ g = \frac{3.1206 * \text{OD of sample} * \text{Volume made up} * \text{Dilution} * 100}{1 * \text{Weight of sample} * 1000}$$

2.5 Determination of color

The visual color of the tomato juice samples was assessed using the Lovibond RT850i CREISS (Cyber Chrome, Inc. Stone Ridge, NY) in terms of L* (lightness), a* (redness), and b* (yellowness and blueness). With the help of a white and black standard, the device was calibrated. The color L*, a*, and b* values were recorded by placing the tomato juice samples in a glass cuvette against the light source.

2.6 Determination of ascorbic acid

The titration technique, as described by Oliveira et al [15], was used to determine the amount of ascorbic acid (vitamin C) in the sample. A sample of around 5 g was weighed, made up to 100 mL with 3% metaphosphoric acid, and filtered. To get a bright pink color, a 10 mL aliquot was added to a titration flask and titrated against 2, 6-dichlorophenol indophenol. Results of ascorbic acid content were calculated by using the below equation and the results were expressed as mg/100 g.

$$\text{mg}\ of ascorbicacidper 100 g = \frac{\text{Titre value} * \text{dye factor} * \text{volume made up} * 100}{\text{mL of filtrate taken} * \text{volume of a sample taken for estimation}}$$

2.7 Statistical analysis

Experimental values were determined by conducting all experiments and analyses in triplicate and calculating their mean and standard deviation (SD). To assess statistically significant differences (P = 0.05) among the means, two-way analysis of variance (ANOVA) and Tukey test (P = 0.05) were employed. The statistical analysis was conducted using IBM SPSS Statistics 21 software (SPSS Inc., Chicago, USA).

3. RESULTS AND DISCUSSION

The effects of different electric fields (2, 4, 6, and 8 kV/cm) and pulse numbers (10, 30, 50, and 70) on lycopene, ascorbic acid, and color values were analyzed and the results were presented in Table 1.

3.1 Effects of LIPEF treatment condition on lycopene content in tomato juice

The lycopene content in untreated tomato juice was found to be 6.68 mg/100 mL. However, in LIPEF treated tomato juice, the lycopene content was found in the range of 7.41 to 11.30mg/100 mL. The maximum and minimum values of 11.30 and 7.41 mg/100 mL respectively, lycopene were found in the treatment at 4 kV/cm for 50 pulse number and 8 kV/cm for 10 pulse number (Table 1).

Table 1: Physicochemical properties of tomato juice as affected by low-intensity pulsed electric field

Parameters	Pulse number / Electric field (kV/cm)	10	30	50	70
Lycopene (mg/100 g)	2	8.34±0.02^{cB}	9.01±0.05^{bC}	10.43±0.10^{aB}	7.75±0.09^{dC}
	4	9.07±.04^{cA}	10.30±0.06^{bA}	11.30±0.10^{aA}	9.23±0.02^{cA}
	6	7.79±0.08^{cC}	9.58±0.04^{bB}	10.11±0.04^{aBC}	7.95±0.24^{cC}
	8	7.41±0.10^{cD}	8.21±0.62^{bD}	9.94±0.21^{aC}	8.55±0.25^{bB}
Ascorbic acid (mg/100 g)	2	24.10±0.75^{aA}	24.19±0.68^{aA}	24.23±1.78^{aA}	23.82±1.14^{aA}
	4	24.19±0.68^{aA}	24.58±0.68^{aA}	24.96±2.27^{aA}	23.43±2.46^{aA}
	6	23.43±0.64^{aA}	23.82±1.14^{aA}	24.19±2.47^{aA}	23.07±0.64^{aA}
	8	23.40±1.81^{aA}	23.37±0.64^{aA}	23.43±2.46^{aA}	23.03±1.80^{aA}
L*	2	19.69±0.58^{aA}	19.76±0.57^{aA}	19.94±0.54^{aA}	19.58±0.57^{aA}
	4	19.59±0.57^{aA}	19.38±0.52^{aA}	19.80±0.38^{aA}	20.06±0.62^{aA}
	6	21.61±0.64^{aA}	21.35±0.62^{aA}	19.90±0.56^{aA}	21.31±0.63^{aA}
	8	21.16±0.65^{Aa}	20.40±0.60^{aA}	19.96±0.56^{aA}	21.25±0.62^{aA}
a*	2	14.84±0.03^{bB}	16.17±0.56^{bA}	18.03±1.17^{aB}	13.65±0.02^{Cb}
	4	15.50±0.07^{bA}	16.39±0.57^{bA}	19.39±0.19^{aA}	16.61±1.19^{bA}
	6	15.36±0.10^{bA}	15.95±0.48^{bA}	18.19±0.59^{aB}	15.50±0.09^{bA}
	8	14.69±0.41^{bB}	15.41±0.03^{bA}	18.11±0.59^{aB}	13.71±0.09^{cB}
b*	2	8.96±0.24^{aA}	9.08±0.22^{aA}	8.62±0.20^{aA}	8.57±0.21^{aA}
	4	9.02±0.21^{aA}	9.86±0.23^{aA}	8.87±0.21^{aA}	8.80±0.20^{aA}
	6	8.90±0.27^{aA}	9.06±0.21^{aA}	8.90±0.20^{aA}	8.96±0.19^{aA}
	8	8.93±0.20^{aA}	9.57±0.23^{aA}	9.50±0.20^{aA}	8.85±0.19^{aA}

(The values are presented as mean ± SD (n=3). Different lowercase letters in the same column indicate significant statistical differences in the effect of pulse number, while different uppercase letters in the same column indicate significant statistical differences in the electric field factor)

The pulse number and electric field both had an impact on the lycopene content of the tomato juice, with the highest lycopene content observed in samples treated with 50 pulses, irrespective of different voltage levels. When subjected to an electric field of 4 kV/cm and 50 pulse number, tomatoes demonstrated a remarkable increase in lycopene content by 69.11% compared to untreated tomato juice. These findings are in line with previous research conducted by Gachovska et al. [16] and Shree et al. [17], where a 68.8% increase in lycopene content was achieved using 16 kV/cm and 50 pulse numbers. The increase in lycopene content observed may be due to the conversion of trans to cis lycopene, resulting from the LIPEF treatment conducted at temperatures lower than 40°C [18].

The lycopene content was found to increase with the increase in the electric field from 2 kV/cm to 4 kV/cm and then decreased with a further increase in the electric field with respect to all pulses. The lycopene content increased by increasing the pulse number from 10 to 50, and decreased with further increase in pulse number regardless of the electric field used as shown in fig. 2. Statistical analysis was carried out to observe the effect of treatment on lycopene and presented in Table 1. It was observed that the lycopene content in tomato juice significantly increased (P = 0.05) through the application of LIPEF treatments and electric field, pulse number and their interaction had significant effect on increase in lycopene content.

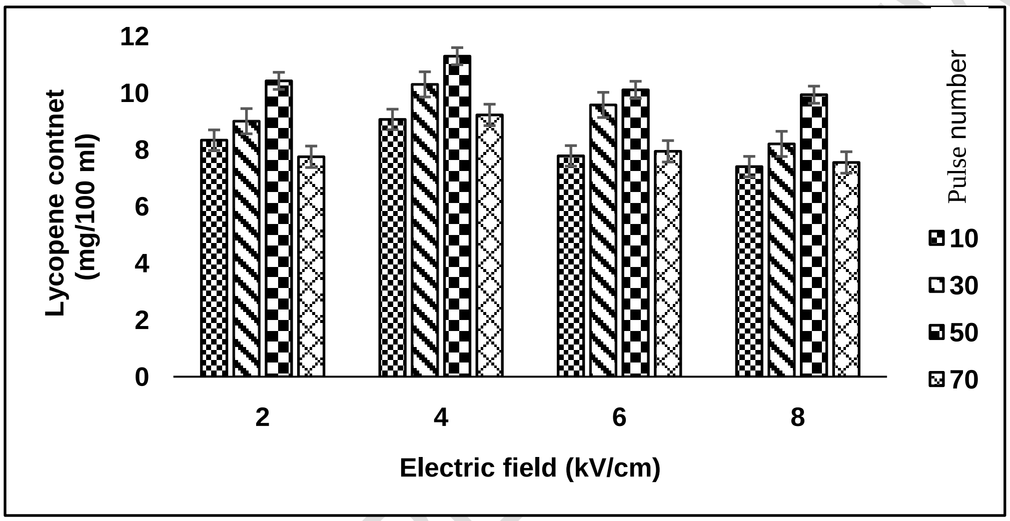

Fig 2. Effect of pulse number and electric field on lycopene content (mg/100 mL)

3.2 Effects of LIPEF treatment condition on Color values retention in Tomato juice

The L*, a*, and b* values of untreated or fresh tomato juice samples were found to be 19.53, 13.65, and 8.95, respectively, which were similar to the values reported by Andreou et al. [19]. The L*, a* and b* values in the LIPEF treated tomato juice was found in the range of 19.38 to 21.35, 13.65 to 19.39 and 8.57 to 9.86 respectively (Table 1). The LIPEF treatment at 6 kV/cm for 30 pulse number gives the maximum L* value of 21.35, while the minimum L* value of 19.38 was observed at 4 kV/cm for a pulse number of 30. The L* values shown that non-significant (P >0.05) effect and the different treatments were not having significant differences. The a* value reached its maximum of 19.39 at 4 kV/cm for a pulse number of 50, whereas the minimum value of 13.65 was obtained at 8 kV/cm for a pulse number of 70. Regarding the b* value, the highest value of 9.86 was observed at 4 kV/cm for a pulse number of 30, while the lowest value of 8.57 was obtained at 2 kV/cm for a pulse number of 70 and it shows significant (P = 0.05) effect. The b* values showed that non-significant (P >0.05) effect and the different treatments were not having significant differences. Treatment of tomato juice at 4 kV/cm for 50 pulse numbers led to an increase in carotenoid content, which was associated with a 40.78% rise in a* value. However, higher electric fields (6 and 8 kV/cm) typically caused a reduction in a* values. The a* value increased with increase in pulse number from 10 to 50 pulse, with further increase in pulse number (Fig 3). That might be due to the result of LIPEF treatments, cell vacuoles may be disrupted, releasing enzymes, substrates, and other substances, causing color degradation, such as lipoxygenase activity, which degrades carotenoid content [20].

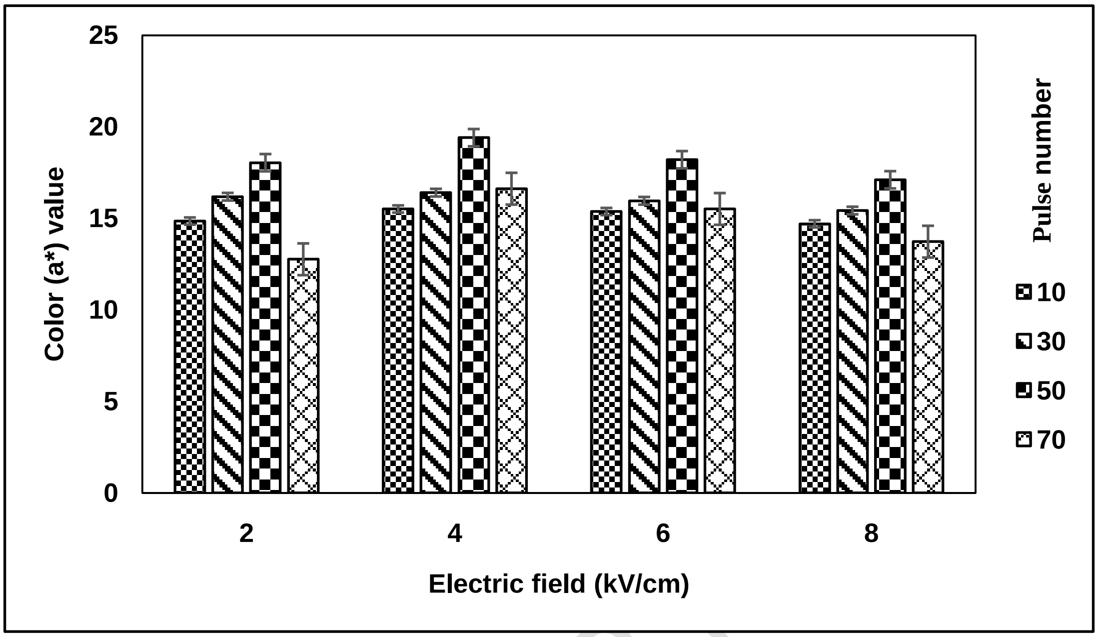

Fig 3. Effect of pulse number and electric field on color a* value

3.3 Effects of LIPEF treatment condition on ascorbic acid retention in tomato juice

The ascorbic acid content of fresh or untreated tomato juice was 24.98 mg/ 100 mL, which is within the range of 7.65 to 59.4 mg/100 mL as reported by Sánchez-Moreno et al. [21]. During the LIPEF treatment, the ascorbic acid content varied from 23.03 to 24.96 mg/100 mL. LIPEF treatment affected the retention of ascorbic acid in tomato juice, with the highest retention of 99.95% observed in juice treated at 4 kV/cm for 50 pulse numbers at 1 Hz, while the highest ascorbic acid loss of about 7.65% was observed at 6 and 8 kV/cm for 70 pulse number at 1 Hz (Table 1). There have been studies showing that orange, orange-carrot and strawberry juices retain more than 80% of ascorbic acid in the juice [22-23]. A slight increase in ascorbic acid retention was observed with increase in pulsed upto 50 pulse numbers, which decreased with further increase in electric field above 4 kV/cm and 50 pulse number (Fig 4). The negligible difference in ascorbic acid content between the control and LIPEF-treated tomato juice may be due to the low processing temperature [12]. LIPEF processing can also stabilize ascorbic acid by inactivating ascorbate oxidase, an enzyme that catalyzes ascorbic acid oxidation in fruit juices [24]. It was observed that electric field, pulse number and their interaction were non-significant effect on ascorbic acid content (Table 1).

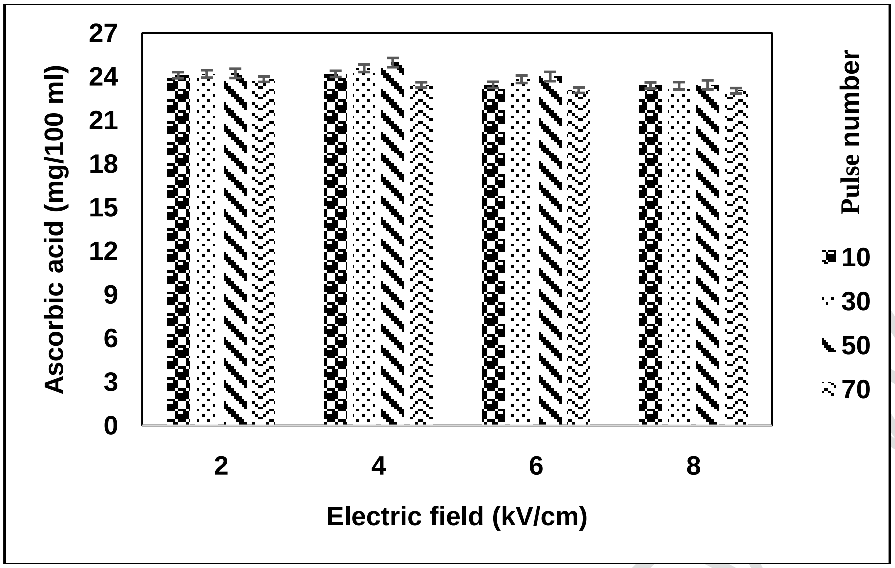

Fig. 4. Effect of pulse number and electric field on ascorbic acid content (mg/100 mL)

4. CONCLUSION

The study showed that LIPEF significantly enhanced the lycopene content in tomato juice of the Roma variety. The higher content of lycopene in comparison to the control demonstrates the potential of PEF as a method to increase the lycopene in the product. However, the effect of bipolar pulses, as well as the shape of the pulse also needs to be investigated in future research work to validate it further. Also, pilot-scale trials of the present research need to be conducted from an economical point of view.

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