

### Obtention of zinc-protoporphyrin from porcine liver pre-treated with pulsed electric field

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**Introduction:** The porcine liver is commonly used to elaborate foie gras and sausages, but a considerable number of consumers reject them because of their low sensory quality (López-Pedrouso et al., 2021). Although porcine liver is rich in essential amino acids, vitamins and minerals, is also an attractive source for zinc-protoporphyrin (ZnPP) that can be used as a natural colouring to improve the food's appearance. Interest is based on metalloporphyrin complex stability against light and heat providing a broad application in the food industry. On the other hand, pulsed electric field (PEF) has been used to enhance the extraction of bioactive compounds from fish by-products (Franco et al., 2020), but its use is still limited. Therefore, this study aimed to optimize the extraction of ZnPP from porcine livers pretreated with PEF by applying different voltages.

**Materials and methods:** Liver samples were homogenized with water in the proportion 1:1. The mixture was intensively crushed and vortexed until complete homogenization. Then, the homogenates were placed between two electrodes separated by 5 cm, reaching 1.8 cm of height. PEF was generated by using a semiconductor-based positive Marx modulator Epulsus-PM1-10 equipped with a batch treatment chamber (EnergyPulse Systems, Portugal). Liver homogenates were treated with voltages ranging from 3000 to 8000 V. Once the PEF treatment was applied, samples were incubated employing a mix of antibiotics. A control batch (without previous PEF treatment) was incubated. The ZnPP (mg/Kg) determination was performed according to Bou et al. (2020). Data were submitted for one-way analysis of variance (ANOVA) with a 95% confidence interval ( $p < 0.05$ ) followed by Duncan's test to separate mean values.

**Results:** The previous PEF treatment allowed an improvement in the extraction of the ZnPP, especially when applied a voltage in the range between 4000 to 6000V. However, there were significant ( $P < 0.05$ ) increments in the concentration of ZnPP for all voltages assayed in the experiment. The highest ZnPP concentration (96.69 mg/Kg) was achieved after using 5000V, which corresponds to a notable increase of 173.7% regarding control batch. Slight voltages during the PEF treatment (3000V) only produced yield extraction of 12.3%, meanwhile stronger ones (8000V) raised this yield to 67.6% with respect to control which showed a final concentration of 35.3 mg/Kg liver. Our data could suggest that a slight PEF treatment has little effect on ZnPP extraction (less increment on the ZnPP concentration concerning other treatments), but on the contrary, an excessive voltage during PEF treatment could cause ZnPP degradation that could affect either pigment or/and its extraction. Thus, the application of intermediate voltages (4000 to 6000 V) is enough to have a positive effect in the extraction of the pigment achieving great amounts of the analyte of interest. Indeed, it has been demonstrated that PEF treatment can produce hydrolysis of macromolecules as proteins (López-Pedrouso et al., 2019).

**Conclusions:** Our results confirm that PEF technology could be useful to improve the ZnPP extraction from liver samples. However, further studies are necessary to confirm our results, employing a greater number of samples of the same and different nature. As PEF assisted extraction is an environmentally and economic choice in terms of operating cost, further research studies should be done to confirm this preliminary study.

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