Effect of temperature, oil content and high power ultrasound on the supercritical CO_2 pasteurization of lipid emulsions

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Supercritical carbon dioxide (SC-CO_2) is a novel and effective pasteurization method, which preserves the nutrients and physicochemical properties of the treated products. Its effectiveness can be improved with the combination of other techniques, such us high power ultrasound (HPU). The pasteurization of lipid emulsions has gained interest due to its application in food, pharmaceutical and cosmetic industries. Several authors observed a strong protective effect against external stress when the microbial cells were in complex physicochemical systems, compared to simple solutions. Although non-thermal pasteurization of liquid products has been extensively studied, the combination of SC-CO_2 and HPU has only been tested in fat-free media. Therefore, in this work, the feasibility of the pasteurization of lipid emulsions using a combination of SC-CO_2 and HPU was studied and compared to the SC-CO_2 treatment alone. For that purpose, *E. coli* inactivation at two different temperatures (35 and 50°C) and 350 bar was considered. Inactivation experiments were conducted in different soybean oil-in-water emulsions (10, 20 and 30 %) and distilled water (0%) as control, with an initial microbial load of 10^7- 10^8 CFU/mL. Treatments were carried out in a supercritical CO_2 plant with an embedded ultrasound system (65 ± 5 W; 30 kHz). For the experiments with HPU, ultrasound was turned on when the desired pressure in the vessel was reached. Samples were extracted at different times and analysed in triplicate by the plate count method. The results were expressed as $\log_{10}(N/N_0)$ versus time and the inactivation kinetics were mathematically modelled by the Weibull model.
The application of ultrasound reduced the process time to completely inactivate the *E.coli* population in approximately 90%, both, for water and lipid emulsions. *E. coli* inactivation rates increased with temperature in the SC-CO_2 treatments. For the emulsions, reductions of 3.4-5.2 log-cycles were achieved in 50 min at 35°C, while 7.8-8.1 log-cycles of inactivation were achieved in 50 min at 50°C. Temperature also had a significant ($p<0.05$) effect on the inactivation with SC-CO_2 + HPU treatments. At 35°C and 5 min, reductions of 6.2-7.0 log-cycles were achieved while, at 50°C, the complete inactivation (7.0-8.0 log-cycles) was achieved in only 3 min for 0 and 10% oil emulsions and, in 5 min for the 20 and 30 % emulsions. At 35°C and 50 min of SC-CO_2 treatment, 3.4, 4.3, 5.2 and 7.4 log-cycles inactivation were obtained for the 30, 20, 10 % emulsions and for water, respectively , which shows the protective effect of the oil in the emulsions. At 50°C only differences were found between water and the emulsions. When HPU was combined with SC-CO_2, the protective effect of the oil content was reduced and found to be significant ($p<0.05$) only at 50°C.
These results show the potential that the SC-CO_2 + HPU technique has for the pasteurization of liquid products. The increase of the oil content in the emulsions increases the protection of microorganisms against the SC-CO_2 treatments. However, the protective effect of the oil can be significantly reduced when high temperatures or HPU are used.

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