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## Study of the effects of High Hydrostatic Pressure (HHP) and Pulsed Light (PL) on BSA structure and hydrolysis

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## **Abstract**

Non-thermal technologies, such as High Hydrostatic Pressure (HHP) and Pulsed light (PL), affect protein inducing modifications in its conformational structure. For this reason the hydrolysis reaction of the protein can be modulated if it is conducted in combination with these technologies which are able to change the availability of peptide bonds exposed to the enzymatic action.

The aim is to study the effects of HHP and PL on the structure and the extent of hydrolysis reaction of a target protein: the Bovine Serum Albumin (BSA). BSA protein (5 mg/mL) in sodium phosphate buffer (50 mM, pH = 8) were treated with PL and HHP at different processing conditions, namely pressure level and treatment time in the case of HHP and treatment time and energy input in the case of PL. Structural modification of the protein solutions were analyzed by determining the sulphidrilic groups and the changes of the secondary structure.

The effect of the two treatments on the hydrolysis degree (HD) at 37  $^{\circ}$ C was also evaluated by OPA method. Chymotrypsin and trypsin (E/S ratio = 1/10) were used to hydrolyze the BSA protein solutions. The hydrolysis was carried out in HHP assisted or PL assisted conditions or the protein solutions were treated with HHP or PL processes and immediately after hydrolyzed with the enzymes.

Results obtained so far demonstrated that the two technologies tested are able to induce protein modifications and the occurrence and importance of this phenomenon depends on processing parameters causing protein unfolding, namely pressure level and number of pulses. When the maximum protein unfolding is obtained, higher HD values are detected. The highest HD value is obtained in HHP assisted hydrolysis with longer treatment time, and when, before undergoing hydrolysis, the PL treatment is applied to the solution placed at the higher distance from the lamp.

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