# DEVELOPMENT AND CHARACTERIZATION OF BIOACTIVE B-LACTOGLOBULIN NANO-HYDROGELS FOR FOOD APPLICATIONS

Óscar L. Ramos<sup>1,2,3</sup>, Ricardo N. Pereira<sup>1</sup>, Artur J. Martins<sup>1</sup>, F. Xavier Malcata<sup>2,3</sup>, António A. Vicente<sup>1\*</sup>

#### INTRODUCTION

Whey proteins produced during bovine cheese manufacture, or membrane fractionation of bovine milk account for increasingly valuable food ingredient – owing to their nutritionally-balanced composition in amino acid residues coupled with their functional properties (e.g. solubility, and gel-forming, emulsifying and foaming features)<sup>1</sup>.  $\beta$ -Lactoglobulin ( $\beta$ -Lg) is the major fraction of bovine whey proteins (i.e. 50 wt%), and a primary gelling agent; it is stable at low pH and highly resistant to proteolytic degradation in the stomach, besides its ability to act as encapsulating agent<sup>2</sup>.

### **OBJECTIVE**

The objective of this work was to understand the kinetics of aggregation during heat treatment, under narrow pH ranges, that leads to the formation of edible  $\beta$ -Lg nano-hydrogels as well as to rationalize the contribution and importance of covalent and noncovalent interactions to maintain the structure thereof. Furthermore, the ability of said nano-hydrogels to encapsulate bioactive compounds (e.g. vitamins) was also assessed.

### MATERIALS AND METHODS

In this study, aqueous dispersions of  $\beta$ -Lg were accordingly produced, and formation of stable  $\beta$ -Lg nano-aggregates was ascertained after heating at different temperatures (i.e. 75, 80 and 85 °C), for different holding periods (i.e. 5, 10 and 20 min) and under different pH values (i.e. 4, 5, 6 and 7); particular emphasis was on pH 6.0. The protein aggregates formed were characterized for stability (turbidity), role of covalent or non-covalent bonds, surface hydrophobicity, morphology, size, surface charge, and content in accessible thiol groups.

# **RESULTS**

Stable dispersions of  $\beta$ -Lg nano-hydrogels were obtained at pH 6, corresponding to an aggregation yield of ca. 65, 77 and 92%, at 75, 80 and 85 °C, respectively – for a holding period of 20 min. Such structures were characterized by particle size between 100 and 160 nm, low degree of polydispersity (<0.2), and an association efficiency of vitamin  $B_2$  above 50%. Their  $\zeta$ -potential varied from +20 to -40 mV, depending on pH. Nano-hydrogels obtained at pH 5 displayed a lower content of accessible thiol groups as compared to those obtained at pH values above or below. For pH between 4 and 5, large sedimenting protein particulates were obtained, whereas soluble aggregates were formed at pH values outside that range.

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