European Journal of Pharmaceutical Sciences 93 (2016) 21–28

Contents lists available at ScienceDirect

European Journal of Pharmaceutical Sciences

journal homepage: www.elsevier.com/locate/ejps

Taylor Dispersion Analysis as a promising tool for assessment of peptide-peptide interactions

Ulrich B. Høgstedt [a,b], Grégoire Schwach [b,c], Marco van de Weert [a], Jesper Østergaard [a,*]

[a] Department of Pharmacy, Faculty of Health and Medical Sciences, University of Copenhagen, Universitetsparken 2, DK-2100 Copenhagen, Denmark
[b] Early Stage Development, Ferring Pharmaceuticals A/S, Kay Fiskers Plads 11, DK-2300 Copenhagen S, Denmark
[c] F. Hoffman-La Roche Ltd., Pharmaceuticals Division, Grenzacherstr. 124, CH-4070 Basel, Switzerland

ARTICLE INFO

Article history:
Received 22 March 2016
Received in revised form 2 June 2016
Accepted 22 July 2016
Available online 25 July 2016

Keywords:
Taylor Dispersion Analysis
Peptide
Self-interactions
Viscosity
Protein

ABSTRACT

Protein-protein and peptide-peptide (self-)interactions are of key importance in understanding the physiochemical behavior of proteins and peptides in solution. However, due to the small size of peptide molecules, characterization of these interactions is more challenging than for proteins. In this work, we show that protein-protein and peptide-peptide interactions can advantageously be investigated by measurement of the diffusion coefficient using Taylor Dispersion Analysis. Through comparison to Dynamic Light Scattering it was shown that Taylor Dispersion Analysis is well suited for the characterization of protein-protein interactions of solutions of α-lactalbumin and human serum albumin. The peptide-peptide interactions of three selected peptides were then investigated in a concentration range spanning from 0.5 mg/ml up to 80 mg/ml using Taylor Dispersion Analysis. The peptide-peptide interactions determination indicated that multibody interactions significantly affect the PPIs at concentration levels above 25 mg/ml for the two charged peptides. Relative viscosity measurements, performed using the capillary based setup applied for Taylor Dispersion Analysis, showed that the viscosity of the peptide solutions increased with concentration. Our results indicate that a viscosity difference between run buffer and sample in Taylor Dispersion Analysis may result in overestimation of the measured diffusion coefficient. Thus, Taylor Dispersion Analysis provides a practical, but as yet primarily qualitative, approach to assessment of the colloidal stability of both peptide and protein formulations.

1. Introduction

Protein-protein (self-)interactions (PPIs) are of key importance in understanding the physicochemical behavior of proteins in solution. For example, PPI measurements can be used to determine the likelihood of success of protein crystallization experiments, where solution conditions leading to moderately attractive PPIs are most likely to yield crystals (Wilson and Delucas, 2014). Protein solubility is also related to PPIs, with the highest solubility generally observed under conditions resulting in the most repulsive PPIs (Wilson and Delucas, 2014). Finally, PPIs are indicative of the colloidal stability of a protein. Under different solution conditions with highly similar conformational stability, aggregation kinetics are generally faster under conditions that result in attractive PPIs as compared to repulsive PPIs (Chi et al., 2003a). In pharmaceutical development there is a strong focus on preventing aggregation since aggregates may cause severe adverse effects to the patients (Kahn and Rosenthal, 1979; Moore and Leppert, 1980).

The magnitude and nature of PPIs are often evaluated by either the second virial coefficient, B_{22}, or the interaction parameter, k_D. These coefficients describe a solution's deviation from ideality, and are related as follows (Harding and Johnson, 1985; Saluja et al., 2007; Zhang and Liu, 2003):

$$k_D = 2B_{22}M_W - \zeta_1 - 2\nu_{sp} \quad (1)$$

where M_w is the molecular weight, ζ_1 is the first order concentration coefficient in the virial expansion of the frictional coefficient, and ν_{sp} is the protein partial specific volume. The value of B_{22} denotes the deviation from ideality of the solution with a negative value as an indicator of attractive interactions, and a positive value indicating repulsive interactions (Saluja et al., 2007). Likewise k_D can be used to assess the nature of PPIs, but due to the hydrodynamic contributions (ζ_1 and ν_{sp}) in Eq. (1) there may be instances where a slightly negative k_D is not indicative of attractive PPIs (Yadav et al., 2010b). However, k_D is still a useful indicator to rank PPIs in the sense that a more negative k_D value indicates more attractive PPIs, and a more positive k_D value indicates more repulsive PPIs (Saluja et al., 2007; Yadav et al., 2010a, 2010b).

Currently, a number of analytical techniques are commonly used to characterize B_{22} and k_D of protein solutions. B_{22} can be determined

* Corresponding author.
E-mail addresses: ulrich.hogstedt@ferring.com (U.B. Høgstedt), gregoire.schwach@roche.com (G. Schwach), marco.vandeweert@sund.ku.dk (M. van de Weert), jesper.ostergaard@sund.ku.dk (J. Østergaard).

http://dx.doi.org/10.1016/j.ejps.2016.07.015
0928-0987/