PLGA-peptide interactions relevant for octreotide-loaded PLGA microspheres

Avital Beig, Justin Hong, Linglin Feng, Rae Sung Chang, Jia Zhou, Rose Ackermann, and Steven P. Schwendeman
College of Pharmacy, University of Michigan, Ann Arbor, MI 48109, USA

OBJECTIVES

- A significant challenge to develop new and generic controlled release PLGA systems is the instability of peptides.
- According to the literature: (1) the octreotide (OCT) bioavailability is reduced (60-63%) in humans when administered from SLAR relative to immediate release injections, (2) OCT can form acylation products when encapsulated in PLGA-glu microspheres, and (3) the acylation products are triggered by an interaction between peptide and polymer.
- It is reasonably expected that the specific formulation may influence the level of acylation products, which can depend on the microsphere formulation, which causes a concern for bioequivalence of future generic products.
- Therefore, our goal is to develop techniques for assessing peptide-polymer interactions and peptide impurities. We have selected SLAR as a suitable reference product to develop these techniques during the relevant storage and in vitro release periods.

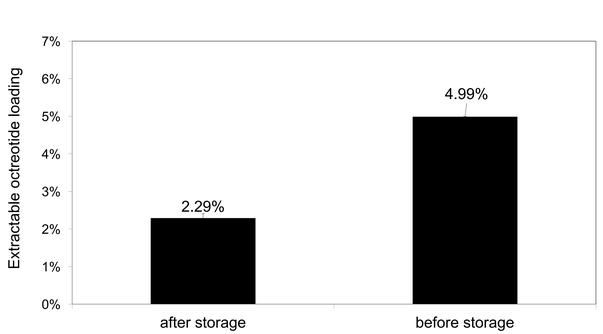

Fig 1. The extracted peptide after storage (6 days, 40ºC/75% RH) implies the covalent octreotide binding in SLAR.

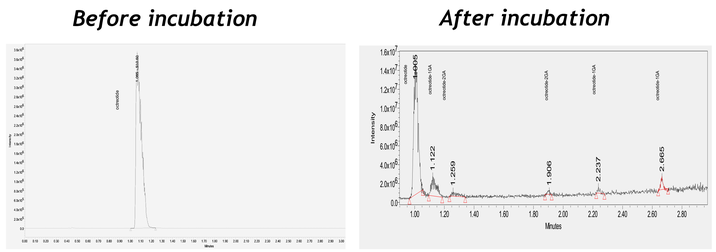

Fig 2. Acylation of octreotide in microspheres before and after incubation at 40°C and 75%RH.

METHODOLOGY

- Microspheres were exposed to storage and release conditions before analysis. In order to estimate covalently bound OCT in SLAR, the microspheres were incubated under storage and release conditions before being analyzed by two-phase extraction/UPLC.
- To extract OCT from incubated SLAR microspheres, microspheres were dissolved in 1 mL of methylene chloride before addition of 4 mL of 50 mM sodium acetate (pH 4), followed by vortexing 1 h. Impurities were identified by UPLC-MS.
- To evaluate the nature of polymer-peptide interaction, SLAR microspheres were incubated with various desorption medias for 24 h at 37 ºC, in the presence of light sonication, and the levels of OCT were measured by UPLC.
- To assess the role of OCT binding after storage and release periods, we monitored the release of OCT in the presence of a PLGA-binding competitor, leuprolide (4 mg/mL in 0.1 M HEPES pH 7.4), in the presence and absence of light sonication help reach encapsulated peptide.

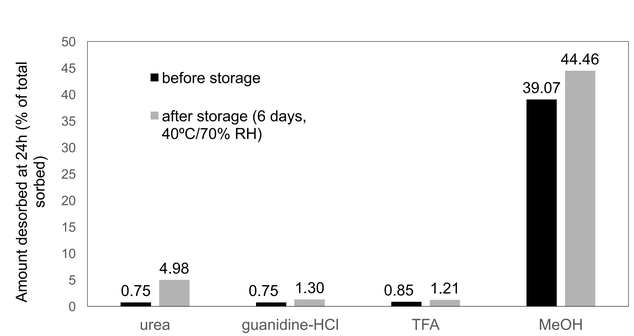

Fig 3. Octreotide-polymer interactions by peptide desorption in decoupling solvents.

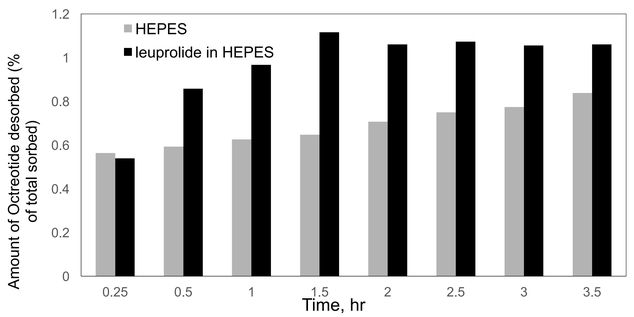

Fig 4. Examination of the octreotide-polymer interactions by competitive binding to leuprolide in presence of light sonication.

RESULTS

- In order to determine the type of interactions OCT was extracted from microspheres. The extracted peptide from fresh SLAR (5.0% w/w) (Fig. 1) was close to theoretical value (5.6%), indicating little or no covalent binding .
- After storage, OCT was not extracted completely from SLAR, suggesting strong and likely covalent PLGA-OCT interactions.
- These interactions also led to significant measurable acylation products of OCT in the microspheres (Fig. 2).
- Various desorption solutions were chosen to selectively disrupt a variety of molecular interactions (Fig. 3). Trifluoroacetic acid (TFA) was selected to potentially compete with PLGA carboxylates for octreotide amino groups. Solutions of 6M urea and 6M guanidine-HCl were used to examine the contribution of hydrophobic interactions and hydrophobic+ionic interactions, respectively.
- The partial desorption of OCT from PLGA while using TFA, urea and guanidine, suggests that ionic interactions do not completely describe the irreversibility of sorption.
- Irrespective of storage only, addition of 50 vol.% methanol in water (decoupling solvent) could release significant amount of OCT from SLAR suggesting excellent peptide encapsulation and some contribution of hydrophobic interactions.
- The presence of the cationic non-peptide competitor for OCT-PLGA interactions, leuprolide, strongly decoupled OCT-PLGA interactions in aqueous media, and the effect was increased with sonication (Fig. 4, 5).

CONCLUSIONS

- OCT binds to PLGA in SLAR under storage conditions, resulting in peptide acylation.
- The binding of OCT and PLGA is likely a combination of both noncovalent and covalent interactions.
- Several methods were successfully applied to quantitate those interactions, which may be useful to assess the peptide-polymer interactions relative to SLAR to assure ultimate bioequivalence of generic PLGA products.

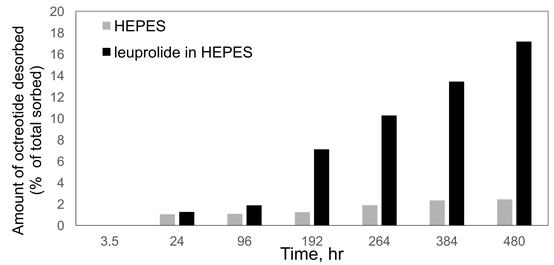

Fig 5. Examination of the octreotide-polymer interactions by competitive binding to leuprolide while shaking.

Funding: This research was funded by FDA contract HHSF223201510170C A0001 BAA.
Disclaimer: This poster reflects the views of the author and should not be construed to represent FDA's views or policies.