A Lipid Based System for the Oral Delivery of Growth Hormone

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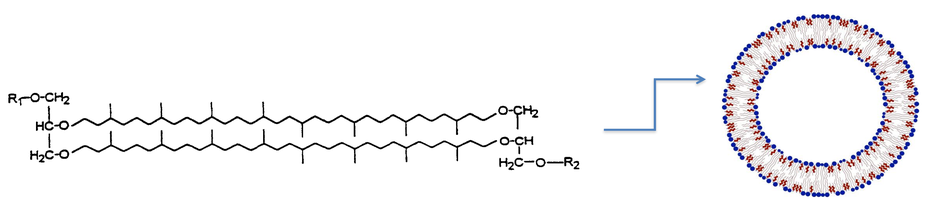
Introduction

Bioavailability of peptides and proteins is extremely low after peroral administration due to their instability in the GI tract and a very poor absorption across the small intestinal wall.
Therefore, we developed a lipid based liposomal carrier system for peroral delivery of peptides. This carrier system is based on the incorporation of tetraether lipids (TEL) into the liposomal lipid bilayer. These are membrane spanning lipids resulting in a significant improvement of liposomal stability.



These liposomes were characterized with respect to size, release properties and in vivo efficacy in rats using human growth hormone (hGH), having a molecular weight of 23.000 Da, as model peptide.

Methods

Liposome preparation:
Liposomes were prepared by the film method followed by extrusion through a 200nm polycarbonate membrane using a LiposoFast extruder (Avestin, Ludwigshafen, Germany).

Lipid bilayer consists of egg-phosphatidylcholin and cholesterol stabilized by up to 30 % tetraether lipids from archeabacteria. Liposomes were loaded with either carboxyfluoresceine (CF) as a model compound, or human growth hormone.

Freeze fracture electron microscopy was applied to study the shape of the liposomes after incorporation of tetraether lipids.

Size distribution:
Liposome dispersions were analyzed by dynamic light scattering (DLS) using a Zetasizer Nano ZS (Malvern, Worcester, UK).

Carboxyfluorescein release:
Release of carboxyfluoresceine (CF) in artificial gastric and intestinal juice at 37° C was measured at different time points using a Fluoroskan Ascent (Thermo Fischer Scientific, Waltham, USA).

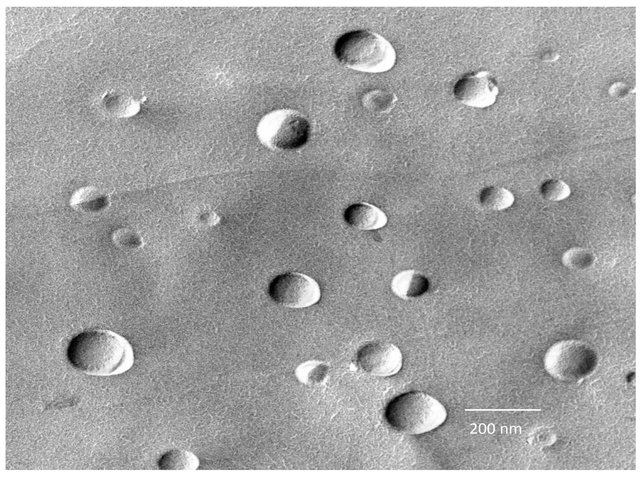
In vivo efficacy:
Liposomes were loaded with human growth hormone and administered to male wistar rats (appr. 200g body weight). Blood samples were taken and growth hormone concentration in plasma was determined by a specific ELISA.

Determination of bioavailability:
Bioavailability was determined by the ratio of AUC values after peroral (p.o.) and subcutaneous (s.c.) administration.

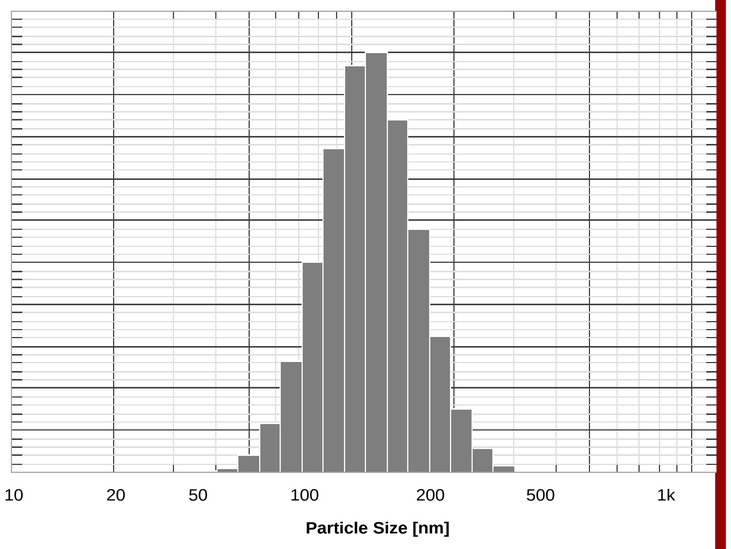
$$\frac{AUC_{po} * dose_{s.c.}}{AUC_{s.c.} * dose_{po}} * 100\% = \%bioavailability$$

Results

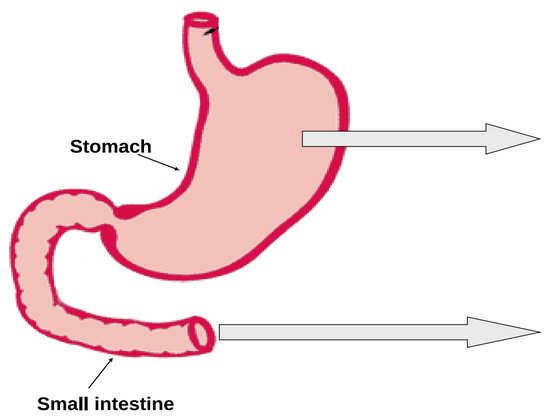
Freeze fracture EM of liposomes:

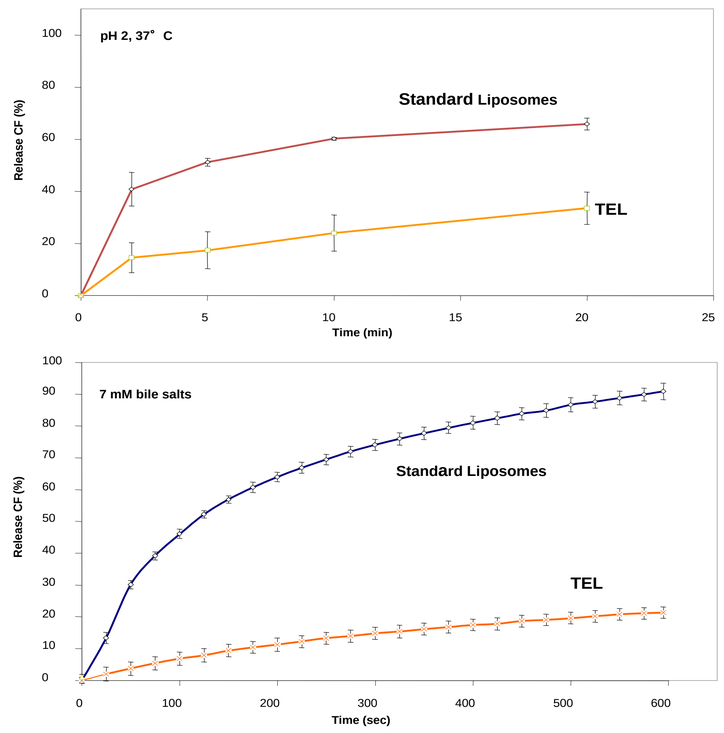


Particle size distribution:



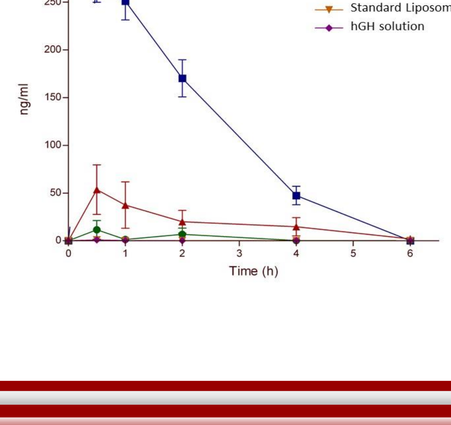
Release Properties:



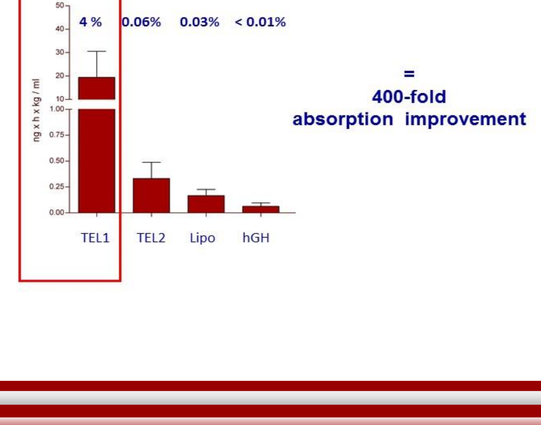


In vivo studies:

hGH Pharmacokinetics



hGH area under the curve



References

•J. Parmentier, M. Becker, U. Heintz, G. Fricker, "Stability of liposomes containing bio-enhancers and tetraether lipids in simulated and gastro-intestinal fluids", Int J Pharm (2011)

• J. Parmentier, G. Hofhaus, S. Thomas, LC Cuesta, F. Gropp, R. Schröder,K. Hartmann,K. Fricker, "Improved oral bioavailability of human growth hormone by a combination of liposomes containing bio-enhancers and tetraether lipids and omeprazole.", J Pharm Sci. 2014;103(12):3985-93
• > all authors do not have any conflict of interest due to this study

Conclusion

- Incorporation of TELs results in a significant improvement of liposome stability in the gastrointestinal tract
- absorption in the small intestine after oral administration is drastically imporved and leads to approximately 3.6% bioavailability of human growth homone.
- Thus, TEL-incorporated liposomes provide a promising tool for the oral delivery of peptides with low bioavailability after oral administration.

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