Toward the Establishment of Standardized *In Vitro* Tests for Lipid-Based Formulations, Part 1: Method Parameterization and Comparison of *In Vitro* Digestion Profiles Across a Range of Representative Formulations

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ABSTRACT: The Lipid Formulation Classification System Consortium is an industry–academia collaboration, established to develop standardized *in vitro* methods for the assessment of lipid-based formulations (LBFs). In this first publication, baseline conditions for the conduct of digestion tests are suggested and a series of eight model LBFs are described to probe test performance across different formulation types. Digestion experiments were performed *in vitro* using a pH-stat apparatus and danazol employed as a model poorly water-soluble drug. LBF digestion (rate and extent) and drug solubilization patterns on digestion were examined. To evaluate cross-site reproducibility, experiments were conducted at two sites and highly consistent results were obtained. In a further refinement, bench-top centrifugation was explored as a higher throughput approach to separation of the products of digestion (and compared with ultracentrifugation), and conditions under which this method was acceptable were defined. Drug solubilization was highly dependent on LBF composition, but poorly correlated with simple performance indicators such as dispersion efficiency, confirming the utility of the digestion model as a means of formulation differentiation. © 2012 Wiley Periodicals, Inc. and the American Pharmacists Association J Pharm Sci 101:3360–3380, 2012

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Additional Supporting Information may be found in the online version of this article. Supporting Information

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INTRODUCTION

The issue of low aqueous drug solubility continues to hinder the robust testing of new chemical entities during drug discovery and development. Of the many formulation strategies that have been used to address the obstacles associated with low solubility, lipid-based formulations (LBFs) have generated significant interest.[1] The composition of LBFs can vary widely, although common design features include the presence of molecularly dispersed drug within a blend of various polar and nonpolar oils with/without surfactant and cosolvent.[2,3] As the drug is presented to the gastrointestinal (GI) tract in solution, although in an oily liquid, the use of LBFs circumvents the limitations to solubility introduced by solute–solute interactions in the crystalline solid.[4] LBFs also promote drug solubilization in the GI fluids via the provision of surfactants and lipids (and their digestion products) that collectively supplement the inherent solubilization capacity of the endogenous GI fluids (i.e., that provided by bile salts, phospholipids, and cholesterol secreted in bile).

Unlike many traditional formulations, the physical and chemical nature of most LBFs is dramatically changed after oral administration by the interaction with biliary and pancreatic secretions in the small intestine, in a process analogous to the digestion of food-based lipids.[5] The major mechanism of chemical change is that of lipid digestion. Lipid digestion is mediated by pancreatic lipases and esterases that are secreted into the upper small intestine in response to the ingestion of exogenous lipid, and to a lesser extent by acid-stable lipases in the stomach.[6] The lipid digestion products generated are subsequently solubilized by bile salt–phospholipid–cholesterol-mixed micelles secreted in bile, resulting in the formation of a range of colloidal species in the GI fluids. The resulting colloidal structures support the solubilization of exogenously administered lipids and coadministered poorly water-soluble drugs. LBFs, therefore, exploit the lipid digestion and absorption cascade, such that drug incorporated into the administered lipid vehicle is transferred into the colloidal phases produced during lipid digestion and these species shuttle digestion products and drug from the lipid substrate to the intestinal wall for absorption.[5,7–10] Current understanding suggests that a critical aspect of this process is the avoidance of drug precipitation during processing of lipid formulations because regeneration of the solid state results in a reversion to a situation consistent with administration of a crystalline drug suspension (where dissolution rate is typically a limitation to drug absorption for poorly water-soluble drugs). One caveat to this overarching suggestion is the recent realization that drug precipitation from LBFs may, for some compounds, result in the amorphous drug being formed.[11] In this case, the process of redissolution of precipitated drug is expected to be faster than if the drug were to precipitate in the crystalline form.

The basic mechanisms by which LBFs promote drug absorption are therefore reasonably well developed. The specific determinants of *in vivo* performance, however, are not fully understood, and robust approaches to probe *in vivo* performance using *in vitro* tests remain poorly defined. The determinants of *in vitro* and *in vivo* performance are likely to include (1) the capacity of the formulation to maintain solvent capacity on dilution and digestion, (2) the rate of formulation digestion [which dictates the rate and quantity of lipid (or surfactant) digestion products and drug that partitions from the oil reservoir into the intestinal milieu], and (3) the solubilization capacity of the localized GI environment when enriched with the products of the digested formulation.[12–15] These determinants are applicable to a wide range of LBF as digestion will inevitably impact on the solubilization capacity of all formulations that contain digestible lipids or surfactants. Digestion of poorly dispersed lipid-rich LBF (such as simple lipid solutions) is typically beneficial to drug absorption because digestion leads to the generation of more amphiphilic lipid digestion products that are more readily incorporated into bile salt–phospholipid-mixed micelles. Digestion therefore catalyzes the *in situ* assembly of highly dispersed colloidal phases with high drug solubilization capacities. In contrast, self-emulsifying formulations and formulations with high proportions of surfactant and cosolvent do not require digestion to reduce particle size because initial dispersion of the formulation commonly leads to the production of nanometer-sized colloidal droplets. Formulation digestion remains unavoidable, however, and may be detrimental to absorption if digestion leads to a decrease in solubilization capacity and drug precipitation.

In an attempt to better understand the performance of a wide range of LBFs, *in vitro* lipid digestion models have emerged as a possible mechanism by which the complex series of *in vivo* interactions that underpin utility may be modeled and predicted *in vitro*.[16–24] In these models, the lipid formulation (containing incorporated drug) is dispersed in a digestion medium that is broadly representative of the contents of the upper small intestine, and digestion of the formulation is initiated by addition of a porcine-derived pancreatic extract containing pancreatic lipase and other pancreatic enzymes. Formulation digestion results in the liberation of fatty acid (FA) from either glyceride lipids or surfactant FA esters, and this leads to a drop in the pH of the digest. By conducting digests in a pH-stat titrator, the transient drop in