Non-Invasive Peptide Delivery, Reformulation, and the Pharma Finance Environment

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Introduction

Since the birth of biotechnology in the mid-1970s more than 200 peptide or protein therapeutics have been created through recombinant DNA technology resulting in annual sales in excess of US$50 billion. While these new peptide and protein drugs demonstrate high potency and high selectivity, and at the same time exhibit essentially no chemical toxicity, their inherent susceptibility to denaturation, poor transmucosal absorption and hydrolysis in the gastrointestinal tract make them unsuitable for oral administration. Most protein or peptide therapeutics are therefore administered by injection – an inconvenient and expensive delivery mode – and patient acceptance of injectable therapeutics in situations where the medical consequences may not be immediate or life threatening, and especially in cases where administration must be frequent and chronic, is a serious issue. As a result, while the range of potential clinical indications for therapeutic proteins and peptides is quite broad, the actual number of such therapeutics in general use is small compared to the number of approved chemically synthesised and orally active drugs.

Intranasal delivery of peptides has proven to be moderately effective for small peptides such as calcitonin (less than 4 kDa), which must be administered chronically on a daily basis for the treatment of osteoporosis, but the actual bioavailability is only about 3% on average (Novartis Pharma, 2003). For larger peptides, intranasal bioavailability has historically been essentially non-existent, although significant advances in this area have recently been made as will be described below. Nevertheless, where adequate bioavailability is attainable, the advantages of intranasal administration in terms of greater patient comfort, convenience and the elimination of needlestick injuries and the concerns about syringe disposal associated with daily injections make this an attractive mode of delivery and one that is well accepted by patients. This is clearly evidenced by the fact that total annual sales for nasally delivered products exceed US$6 billion. More to the point, sales of nasally delivered versions of previously injectable-only therapeutics have demonstrated up to a 33-fold increase in annual sales compared to the original injectable formulations.

Pharma Industry and Finance Market Dynamics: Shortened ROI Horizons Now Demanded by Investors

Since the mid-1990s, investors have become increasingly disenchanted with the long development timeframes and high risk of failure associated with the development of so-called 'NCEs' (New Chemical Entities) – novel drugs that offer the potential for new therapeutic modalities. This is reflected in the dramatic change in the investment philosophy of many venture capitalists since the mid-1990s and also in the equity market pressures on public companies to focus on drug development programmes that promise shorter times to market. Reformulation of approved drugs has garnered the attention of both venture investors and pharmaceutical companies as a low-cost/low-risk route to increased product sales and perceived company value in the relatively near term.

A growing number of FDA-approved, off-patent (or soon-to-be off-patent) injectable drugs are directly amenable to 505(b)2 FDA regulatory treatment. The 505(b)2 provisions allow for a new route of administration or a new indication, among other things, for an existing drug (*Table 1*). Depending upon the specific clinical indication, this accelerated approval route can permit an NDA filing in as little as 30 months. The benefits of life cycle management strategies for the innovator company (the original developer of a drug) based on reformulation and new modes of non-invasive drug delivery are high,

Pharmaceutical Product Changes Amenable to 505(b)2 Regulatory Treatment

- New indication
- Route of administration
- Dosage form
- Dosage strength
- Formulation – change in quality or quantity of excipient
- Dosing regimen/frequency
- Combination of two approved drugs
- NCE – (new salts, pro-drugs, active metabolites)
- Changes from monograph

Table 1 – Pharmaceutical Product Changes Amenable to 505(b)2 Regulatory Treatment

and the expense is minimal compared to that of creating NCEs. When an innovator fails to exploit opportunities to replace suboptimal formulations using newly enabling technologies, or fails to take advantage of their existing branding efforts through reformulation opportunities, this provides prime opportunities for small specialty pharma companies to enter and exploit proven markets with dramatically reduced regulatory and business risks (*Tables 2 and 3*). Opportunities for the reformulation of injectable peptide drugs, as well as certain small molecule drugs, abound. Reformulation of non-peptide, small molecule drugs to improve suboptimal pharmacodynamics has been a mainstay of life cycle management for 'big pharmas' and can be a commercially worthwhile endeavour for 'specialty pharmas'. Reformulating an injectable peptide or protein into a non-invasive formulation such as a metered nasal spray is considerably more challenging, because of the inherent shortcomings of peptides mentioned above – denaturation, poor transmucosal absorption, and hydrolysis in the gastrointestinal tract. For this reason, the clinical and commercial upsides are more dramatic and worth the effort.

The Regulatory Risks for Reformulated Drugs are Minimised

- The APIs have proven clinical utility and safety via the injection route.
- A new route of administration for approved, off-patent drugs is amenable to 505(b)2 FDA regulation.
- The pharmacokinetics and pharmacodynamics goals are defined.
- Clinical end-points – typically bioavailability, non-inferiority – are defined.
- New indications for already approved, off-patent drugs are also amenable to 505(b)2 FDA regulation.

Table 2 – The Regulatory Risks for Reformulated Drugs are Minimised

The Business Risks for Reformulated Drugs are Well Defined and Understood

- Market size
- Physician and patient expectations
- Clinical development time
- Clinical difficulty – On time/On budget probability
- Estimated clinical cost
- Time to market
- Competition and market trends
- Number of potential partners
- Required marketing/sales force size
- Manufacturing difficulty/cost
- Potential sales forecast
- Competitive advantage
- Patient convenience/Who will pay/How much

Table 3 – The Business Risks for Reformulated Drugs are Well Defined and Understood

Key Issues in Peptide Reformulation

Three key technical hurdles specifically associated with reformulation of peptide drugs for intranasal administration need to be addressed in order to reduce the associated technical and financial risks to satisfactory levels. These are (i) the achievement of adequate bioavailability, (ii) the prevention of peptide aggregation and (iii) the minimisation of immunogenicity.

Achieving Adequate Bioavailability

Since the mid-1980s, a large number of molecules have been screened for the ability to enhance transmucosal delivery of peptides with limited success. These have included surfactants, cyclodextrins, charge-modifying agents, mucolytic or mucus-clearing agents, ciliostatic agents, liposomes, vasodilator agents, selective transport-enhancing agents, and modulators of epithelial junction physiology, among others. For the most part, these agents have provided bioavailabilities in the single-digit percentage range for peptide or protein drugs.

In recent years, the development of a large class of alkylsaccharide delivery enhancement agents – molecules that provide unsurpassed intranasal bioavailabilities, comparable to those achieved by injection – was pioneered by Elias Meezan and Dennis Pillion at the **University of Alabama Medical Center**, Department of Pharmacology and Toxicology (Pillion *et al.* 1994, 1995). The resulting families of alkylsaccharide molecules are collectively designated as Intravail® absorption enhancement agents. A large number of molecules are included in this structural class, and the scope and properties in the broader class are quite diverse. Intravail agents allow intranasal delivery – or more broadly, transmucosal delivery (Ahsan *et al.* 2003, Yang *et al.* 2005) – of peptide, protein and non-protein macromolecular therapeutics (Arnold *et al.* 2002) having molecular weights up to and in excess of 20 kDa. They are

chemically synthesised molecules that are metabolised to CO_2 and H_2O and provide controlled transient permeation of the mucosal barrier. They are non-irritating to mucosal tissue and have been designated as Generally Recognised As Safe (GRAS) substances by the **FDA**. They are highly effective, in some cases being fully active at 1/15,000 of the prescribed **WHO**'s ADI (allowable daily intake). The permeation effectiveness of the Intravail excipients has been demonstrated to be largely a function of molecular weight. An example of enhanced transmucosal absorption of a 4 kDa peptide drug demonstrated in a three-way cross-over study conducted with 10 normal human female patients using an Intravail excipient is shown in Figure 1. The Intravail excipient is seen to increase intranasal bioavailability to 36% from 7% for peptide without the excipient. Figure 2 demonstrates the equivalent glucose-lowering pharmacodynamics of an intranasally administered GLP-1 peptidic agonist (Exendin-4 or Byetta® (exenatide)), using Intravail as a transmucosal absorption enhancer, compared to the currently approved subcutaneous injectable in an ob/ob mouse model of diabetes. In direct contrast, in the absence of an Intravail excipient, no glucose-lowering effect is seen upon nasal administration of Exendin-4.

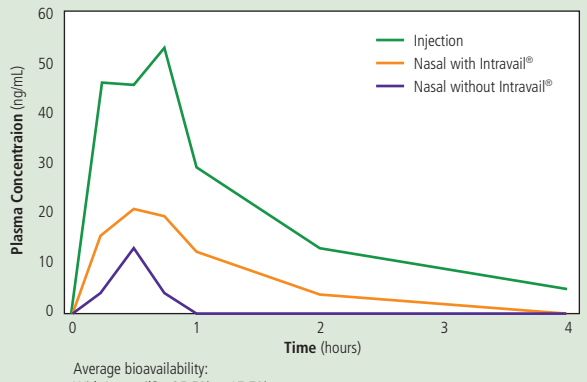

Figure 1 – A ten-patient three-way cross-over study demonstrates enhanced intranasal bioavailability with Intravail excipient (36% vs. 7%) for a 4 kDa peptide.

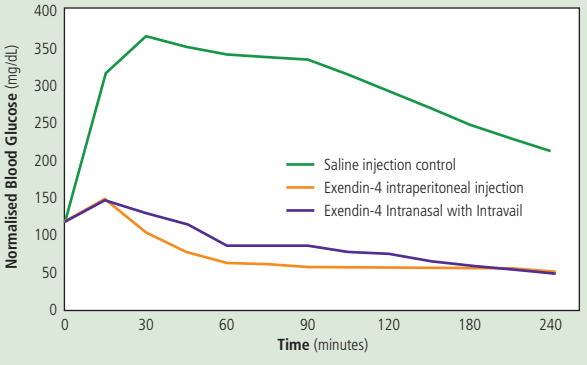

Figure 2 – Pharmacodynamics of intranasally administered Exendin-4 using 0.1% Intravail excipient and currentsubcutaneous injectable in the diabetic ob/ob mouse model.

Intravail Increases Oral Cmax

Administered Dosage*	Serum C_{max}
25 mg sumatriptan dose – without Intravail	104 ng/mL
25 mg sumatriptan dose – with Intravail	700 ng/mL
Change in C_{max} due to Intravail excipient	6.7-fold increase

* in canines.

Table 4 – Intravail Increases Oral Cmax

The use of various Intravail excipients has recently been extended beyond intranasal administration of peptides and proteins to include oral, flash-dissolve buccal, and paediatric rectal suppository applications for small molecule drugs as well as peptides with dramatic results. For example, in a canine study comparing oral absorption of sumatriptan, the C_{max} in serum is seen to increase sevenfold in the presence of an Intravail excipient (*Table 4*).

Preventing Peptide Aggregation

Proteins undergo numerous physical and chemical changes that affect potency and safety. Among these are aggregation, which includes dimerisation, trimerisation and higher-order aggregates. Aggregation is rapidly emerging as a key issue underlying multiple deleterious effects for peptide or protein-based therapeutics, including loss of efficacy, altered pharmacokinetics, reduced stability or product shelf life. Moreover, as described below, aggregation can stimulate induction of unwanted immunogenicity. Hydrophobic aggregation mediated by seemingly innocuous solution formulation conditions can have a dramatic effect on the subcutaneous bioavailability and pharmacokinetics of a therapeutic peptide and, in the extreme, can totally preclude its absorption (Clodfelter *et al.* 1998). During the course of the manufacturing and formulation process, proteins are purified and concentrated using a variety of means. These means include ultrafiltration, affinity chromatography, selective absorption chromatography, ion exchange chromatography, lyophilisation, dialysis and precipitation. Such concentration can lead to aggregation, which in turn can increase the immunogenicity of the protein therapeutic. It was recently discovered that addition of small amounts of certain alkylglycosides, either in the final formulation or during the course of purification and concentration, reduces or eliminates peptide or protein aggregation. This has the added benefit of conveying remarkable stability to those peptide or protein drugs that are susceptible to loss of activity by aggregation such as insulin, human growth hormone, and erythropoietin (EPO), to cite a few examples. While some of the same alkylsaccharides that confer increased transmucosal absorption also confer

increased stability, the group of stabilising alkylsaccharides are designated ProTek® excipients. The extended room temperature stability for insulin and human growth hormone conferred by ProTek excipients allows for more favourable 'cold chain' handling requirements, greater patient convenience in daily use and, in the case of wearable insulin pumps, less frequent need to discard and replace the insulin reservoirs. Stabilisation of insulin and human growth hormone, by ProTek excipients for extended periods of time upon continuous agitation at 150 rpm, 37 °C, as measured by light scattering, is shown in Figures 3–5. Insulin stability has been independently confirmed by bioassay at day 60.

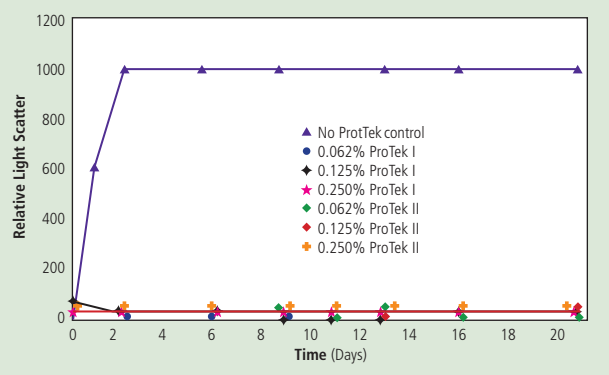

Figure 3 – Dramatic stabilisation of human insulin (pH 6.5) using ProTek alkylsaccharide excipients (at 37 °C, 150 rpm).

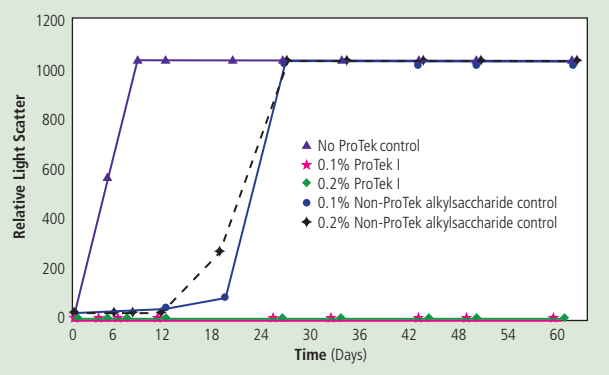

Figure 4 – Extended 60-day stability of human insulin (pH 7.5) with ProTek I compared with a non-ProTek alkylsaccharide control (at 37 °C, 150 rpm).

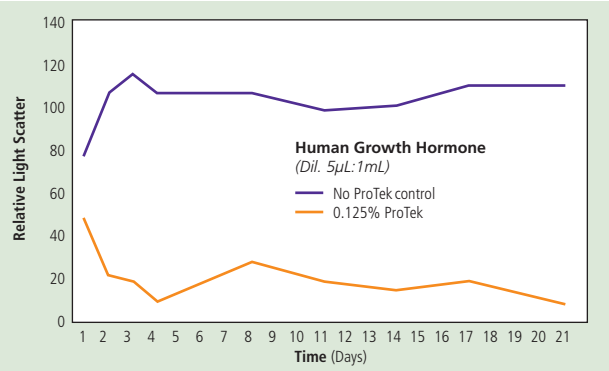

Figure 5 – Stabilisation of human growth hormone (Dil. 5µL:1mL) using ProTek alkylsaccharide excipient (at 37 °C).

Minimising Immunogenicity

In addition to reducing or eliminating transmucosal absorption of peptide and protein drugs, aggregation can stimulate induction of unwanted immunogenicity. Over the last five years, the FDA and other regulatory agencies have increased their scrutiny of aggregation events, and biopharmaceutical companies have therefore increased their efforts to characterise, understand and control or eliminate aggregation. Of particular concern is the induction of unwanted immunogenicity. The immunogenicity of a self-associating peptide can be influenced by the formation of aggregates as a result of non-covalent intermolecular interactions. For example, interferon has been shown to aggregate resulting in an antibody response (Hermeling *et al.* 2006). The antibody response to erythropoietin has been shown to produce 'pure red cell aplasia' in a number of patients receiving recombinant EPO (Casadevall *et al.* 2002), which is potentially a life-threatening side-effect of EPO therapy. As illustrated above, insulin loses activity rapidly as a result of protein aggregation upon agitation at temperatures above those found upon refrigerated storage (Pezron et al. 2002; Sluzky *et al.* 1991). Aggregation of recombinant AAV2 results in reduced yield during purification and has deleterious effects on immunogenicity following *in vivo* administration (Wright *et al.* 2005). Monoclonal, antibody-based therapeutics have also been shown to be subject to inactivation as a result of protein aggregation (King *et al.* 2002). Recombinant human factor VIII (rFVIII), a multidomain glycoprotein, is used in replacement therapy for treatment of haemophilia A. Unfortunately, 15–30% of the patients treated develop inhibitory antibodies. The presence of aggregated protein in formulations is generally believed to enhance the antibody development response (Purohit *et al.* 2006). In addition, protein aggregation can be induced by necessary excipients such as the antimicrobial preservative benzyl alcohol, which is included in some formulations to maintain product sterility (Roy *et al.* 2005). The use of ProTek alkylsaccharides to prevent aggregation as shown above may provide a simple and attractive means of minimising unwanted immunogenicity for some peptide or protein drugs.

Summary and Conclusion

Increasingly, venture capitalists, life science fund managers and the other private and public investors comprising the pharma finance community have become disenchanted with the high cost, long development times, high risk of failure and increasing regulatory barriers associated with the development of new drugs. Reformulation of approved drugs provides a low-cost/low-risk and relatively high-return route to increased product sales in the relatively near term along with a concomitant increase in perceived company value. Reformulation of approved but suboptimally formulated drugs having proven clinical utility and safety, as well as well-defined pharmacokinetics, pharmacodynamics and clinical end-points, minimises

the associated business, regulatory and market risks. Depending upon the specific drug and clinical indication in question, the accelerated approval routes available for reformulated drugs can permit NDA filings in as little as 30 months.

While reformulation of non-peptide, small molecule drugs to improve suboptimal pharmacodynamics has been a mainstay of life cycle management for 'big pharmas' as well as 'specialty pharmas', particularly fruitful but more challenging reformulation opportunities exist in the case of peptide and protein drugs. New technologies to overcome the inherent challenges of achieving adequate bioavailability, preventing peptide aggregation and minimising immunogenicity have been developed. A growing class of alkylsaccharide delivery enhancement and protein stabilisation agents designated Intravail and ProTek excipients, respectively, permit intranasal delivery – or more broadly, transmucosal delivery of peptide and protein drugs having molecular weights up to and in excess of 20 kDa, and long-term room temperature stability and reduced immunogenicity for many peptides and proteins.

Technical developments and evolving financial pressures are fostering a realignment and new emphasis of many drug delivery platform companies to become specialty pharma companies. Reformulation of injectable peptide or protein drugs into more patient-friendly forms such as metered nasal sprays, oral and flash-dissolve sublingual or buccal formulations, offers a particularly large, attractive and as yet untapped opportunity, for all stakeholders in the pharmaceutical field – drug delivery and specialty pharma companies, 'big pharma' companies, physicians, patients and third-party payers.

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