“Injectable Drug Delivery 2011: Formulations Focus”

This edition is one in the ONdrugDelivery series of publications from Frederick Furness Publishing. Each issue focuses on a specific topic within the field of drug delivery, and is supported by industry leaders in that field.

EDITORIAL CALENDAR 2011:
September: Prefilled Syringes
October: Oral Drug Delivery
November: Pulmonary & Nasal Drug Delivery (OINDP)
December: Delivering Biotherapeutics

SUBSCRIPTIONS:
To arrange your FREE subscription (pdf or print) to ONdrugDelivery, contact:
Guy Furness, Publisher
T: +44 (0) 1273 78 24 24
E: guy.furness@ondrugdelivery.com

SPONSORSHIP/ADVERTISING:
To feature your company in ONdrugDelivery, contact:
Guy Furness, Publisher
T: +44 (0) 1273 78 24 24
E: guy.furness@ondrugdelivery.com

MAILING ADDRESS:
Frederick Furness Publishing
48, Albany Villas, Hove, East Sussex, BN3 2RW
United Kingdom

PRODUCTION/DESIGN:
Mark Frost
www.frostmark.co.uk

“Injectable Drug Delivery 2011: Formulations Focus” is published by Frederick Furness Publishing.

Copyright © 2011 Frederick Furness Publishing.
All rights reserved

www.ondrugdelivery.com

Contents

Medusa: an Innovative Formulation Approach to Improve Pharmacokinetic & Safety Profiles of Biotherapeutics
Dr You-Ping Chan, Director of Chemistry & Analytical Research,
Dr Rémi Meyreux, Director of Formulation of Injectable Products
Platform, Camille Rivail, Business Development Analyst &
Dr Jean Chatellier, Vice-President Alliance Management
Flamel Technologies, SA

4-6

The IVECT Method: Intezyne’s Versatile Encapsulation & Crosslinking Technology as a Platform for Drug Delivery
Dr Adam Carie, Senior Scientist, Preclinical Development,
Dr Jonathan Rios-Doria, Manager, Preclinical Studies,
Dr Habib Skaff, CEO, & Dr Kevin Sill, CSO
Intezyne, Inc

8-11

Enhanced Drug Delivery to the Brain: Safe & Versatile
Mr Willem van Weperen, CEO & Dr Pieter Gaillard, CSO
to-BBB technologies, BV

13-15

Novel Polymers for Enhancing Therapeutic Half-life & Drug Targeting
Dr Jill Ogden, Vice-President of Business Development &
Dr Richard Palmer, Executive Chairman
Warwick Effect Polymers, Ltd

16-19

www.ondrugdelivery.com
Copyright © 2011 Frederick Furness Publishing
Maximizing value in your drug delivery development pipeline through profitable drug delivery partnerships

100+ Drug Formulation & Drug Delivery Decision Makers
100+ Latest formulations and Delivery Technologies
30+ Expert Speakers
3 Days of Learning & Networking

ASIA’S MOST INFLUENTIAL DRUG FORMULATION & DELIVERY GATHERING

MULTINATIONAL PHARMA SPEAKERS

Ursula Redeker
Head Technical Development Biologics
ROCHE
Switzerland

Keith Horspool
VP Pharmaceuticals
BOEHRINGER INGELHEIM
USA

Peter Hoffmann
VP New Technology Development
GENZYME
USA

David Monteith
Head, Pharmaceutical Development, Emerging Markets
MERCK, USA

ASIA PACIFIC PHARMASPEAKERS

Kennie Dee
VP R & D
UNILAB
Philippines

Jack Aurora
CSO
HISUN
China

Henry Sun
Vice-President
TASLY
China

Chang Shan Hsu
President
INNOPHARMAX
Taiwan

Register by 2 September and save SGD 200!

Register at www.imapac.com or call +65 6493 1597 for details

ASIA PACIFIC’S ONE AND ONLY INDUSTRY-LED INTERNATIONAL CONFERENCE THAT BRINGS TOGETHER FORMULATION AND DELIVERY DECISION MAKERS FROM PHARMAS, TECHNOLOGY COMPANIES, ACADEMICS TO SHARE MARKET INTELLIGENCE, BEST PRACTICES AND EXPLORE PROFITABLE PARTNERSHIPS

Media Partner:
Proteins and peptides are an important class of drugs as they allow for better treatment of many diseases by having higher specificity and activity. However, many biotherapeutics face numerous formulation hurdles such as poor stability, aggregation, and short half-life, which may in turn cause such promising molecules to be shelved. Alternatively, if developed, they may have limited commercial attractiveness due to sub-optimal safety, efficacy, or convenience. In addition, development and manufacturing costs associated with this class of drugs are significantly higher than those of small molecules.

The use of traditional sustained-release systems for proteins and peptides can solve some of these problems but also creates a number of challenges. These include: complexity of formulation processes under aseptic conditions; stability of the drug, especially when organic solvents are involved; burst effect; local tolerability; and low bioavailability. The use of polymeric systems, such as PLA/PLGA, has been studied for more than 25 years but still requires the use of organic solvents and is therefore not adapted to large and fragile protein formulation.

Other approaches used to prolong the half-life of biologics, such as protein engineering (for example, PEGylation, albumin fusion and
Fc fusion) necessitate the development of a new biological entity (associated with higher clinical development risks and costs) and generate drastic decrease of the biological activity due to detrimental conjugation and/or steric hindrance issues.

The Medusa® drug delivery platform has been designed to address the challenges related to the formulation, pharmacokinetics and safety profile of biologics. Medusa enables the creation of precise pharmacokinetics of biotherapeutics over time, leading to a reduction of side-effects and better treatment compliance.

The amphiphilic Medusa polymer is based on a hydrophilic biodegradable polyglutamate chain grafted with hydrophobic Vitamin E. The polymer self-assembles in aqueous medium to form a stable solution of nano-sized hydrogels comprising multiple polymer chains and 95% water. The hydrogel is formed by hydrophobic interactions only, no chemical cross linking agent is required. A diagramatic overview of how the polymer chain and the drug self-assemble in water to give the hydrogel, and the resulting PK profile after administration, is given in Figure 1.

Formulated with Medusa, the biological entity will spontaneously associate with the hydrogel through reversible hydrophobic and/or electrostatic interactions (Figure 2). The solution containing non-covalently bound hydrogel/biologics complexes is ready to be administered.

Based on hydrophobic and/or electrostatic interactions, this association step is performed conveniently in water at room temperature, without the use of any organic solvents, heat or shear mixing. The liquid formulation containing the hydrogel can be sterile filtered, then filled in vials or cartridges or freeze-dried and reconstituted in water.

The scale-up and transfer of the formulation process are easy and do not require sophisticated handling or reactors (see Figure 3). The mild conditions used allow the proteins or peptides to maintain their complete structural and functional integrities, as proved by a panel of analytical methods such as RP-HPLC, SEC, Western Blot and bioactivity studies following extraction from the Medusa hydrogel.

After subcutaneous injection, a depot is formed at the injection site and the fully active biologic is slowly released in vivo over one to fourteen days in humans, as it is displaced by competition with physiological proteins (such as human serum albumin) available in high concentrations in the subcutaneous tissue.

By adjusting the hydrogel/biologic ratio, the length of the polyglutamate chain, and the amount of Vitamin E, the association of the formulated biologic with the polymer can be finely tuned, and so accommodate any physicochemical and structural features of most biologics, best adapted to the therapeutic objective.1

As an example, interferon alpha-2b (IFN-α2b) has been formulated with Medusa to extend the release over one week in humans, and is currently in a Phase II clinical study in France (ANRS HC 23 COAT-IFN).2

In vitro, it has been demonstrated that using serum bovine albumin (BSA), the interferon is released upon increasing albumin concentration (Figure 4), and that the interferon is chemically unchanged as evidenced by RP-HPLC. The release is complete with excess BSA and, when tested for in vitro activity, 100% activity is observed. Indeed, the Medusa/interferon association is physical in nature and fully reversible, enabling the controlled delivery of non-denatured, fully-active proteins within the appropriate therapeutic window.

In summary, Medusa is well suited to meet challenges in developing and manufacturing formulation of biologics. It allows not only the sustained delivery of biological drugs, from one day to up to two weeks in humans, but also the solubilisation of poorly soluble molecules (for example, solubilisation of IL-2 (Proleukin®), which has been enhanced by 35 times using Medusa). Improvement of the safety profile is also achieved. Since no chemical modification is required, the application of Medusa to the lifecycle management of existing biological drugs is straightforward from a clinical and regulatory point of view.

**BENEFITS OF MEDUSA**

The advantages of Medusa over other drug delivery systems are summarised in Figure 5.

- Applicable to a wide range of peptide and protein drugs as well as small molecules
- Sustained delivery from 1 to 14 days in humans
- Combination of several different drugs in the same formulation
- Remarkable increase of solubility of poorly soluble biologics
- Full activity of biologics maintained
- Improved safety profile / good local tolerance
- Safe, non-immunogenic, fully biocompatible and biodegradable hydrogels: glutamic acid and Vitamin E are GRAS
- GMP manufacturing of the polymer at commercial scale
- Bio-friendly, water-based process
- Formulation process is cost effective, easy to implement, to scale-up and to transfer
- Strong intellectual property position and Freedom to Operate (no third party licensing obligations).

![Figure 2: Efficient Loading of Various Proteins](Image)

![Figure 3: A “Bio-Friendly”, Rapid and Easy-to-Scale-Up Formulation Process.](Image)
ABOUT FLAMEL TECHNOLOGIES

Flamel Technologies SA (NASDAQ: FLML) is a leading drug delivery company dedicated to develop safer, more efficacious formulations of drugs addressing unmet medical needs. Its product development pipeline includes biological and chemical drugs formulated with the Medusa® and Micropump® proprietary platforms.

The Medusa platform (injectable drugs, described here) for the formulation and/or the extended release of biologics (including proteins, antibodies, and peptides) as well as small molecules.

- DeliVax™, Medusa’s vaccine application, permits the efficient formulation of antigens.

The Micropump micro-encapsulation platform (oral drugs) for the formulation and the controlled release of chemical drugs, is designed to increase the absorption time of drugs, particularly for drugs only absorbed in the small intestine and to deliver the drug in specific sites in the gastrointestinal tract. Micropump allows tailoring the exact kinetics required to optimize the final product and offers the advantage to easily and accurately mix microparticles with different release kinetics, in different ratios, every single particle performing independently. Micropump can be presented in various dosage forms such as capsule, tablet, sachet or oral suspensions (LiquiTime™) without modifying the release rate.

The company’s has developed products approved in the USA and in Europe and manufacture Micropump-based microparticles.

- LiquiTime™, allowing stable and controlled release ready-to-use liquid oral suspensions of one or several combined drugs over time.
- Trigger Lock™ for the controlled release of narcotic and opioid analgesics while deterring tampering (particles cannot be crushed to extract the active).

These versatile drug delivery platforms may be used to address threshold formulation problems such as poor solubility, aggregation and instability for both chemical and biological drugs. The company’s innovative delivery platforms are used for the LCM of marketed products, including Biobetters, and the development of new compounds with many unique competitive advantages:

- Improvement of drug characteristics such as efficacy, bioavailability and pharmacokinetics
- Improvement of the drug safety profile with a noticeable diminution of peak dose concentrations, which in turn allows administration of higher effective doses and potentially greater efficacy
- Potential improvement of patient compliance due to reduced side-effects and greater convenience
- Protection of market position through patent extension and/or product differentiation
- Extension of market to new indications and new patient populations.

Flamel Technologies has collaborations with a number of leading pharmaceutical and biotechnology companies, including Baxter, GlaxoSmithKline (Coreg CR®, carvedilol phosphate), Merck Serono and Pfizer.

Medusa® and Micropump® are registered trademarks of Flamel Technologies SA.

REFERENCES:

1. US DMF Type IV on Medusa Polymer (filed: assigned number 024634).
BIT’s 1st Annual Symposium of
Drug Delivery Systems (SDDS-2011)

Time: November 3-5, 2011 Venue: Shenzhen, China

Highlights of the Conference
• To Organize up to 300+ Oral Presentations on the Latest Tech Trend of Drug Delivery Systems
• To Bring together 100+ Project Posters on New Technologies Implemented in Industrial Manufacturing
• To Network with 500+ Colleagues and Friends in China and Beyond
• To Meet with 100+ Leading Suppliers from all over the World

Programmed Tracks at a Glance:
Track 1: Outlook for Global Market Trend of Drug Delivery Systems
Track 2: Breaking Research and Technologies of DDS
Track 3: Current Innovative DDS for Treating Major Diseases
Track 4: Medical Devices for Effective Drug Delivery
Track 5: Nanotech for Drug Delivery
Track 6: Regulatory Issues, Quality Control, and Business Development
Workshop 1: Key Regulatory and Legal Issues in Improving Bioequivalence or Bioavailability by New DDS
Workshop 2: Strategic Planning for Patient Centered Drug Delivery Product

Contact Information:
Ms. Grace Yang
Organizing Commission of SDDS-2011
28 Gaomeng St., F1
Dalian High-tech Zone
Dalian, LN 116025, China
Tel: 0086-411-84799609-821
Fax: 0086-411-84799629
Email: grace@bitlifesciences.com

http://www.bitlifesciences.com/sdds2011
Advancements in combinatorial chemistry and high-throughput screening have culminated in the production of volumes of chemical libraries for pharmaceutical and biotechnology companies looking to expand product pipelines. However, a major hurdle in the development of new chemical entities from these libraries lies in the intrinsic physiochemical properties of the compounds themselves. Poor aqueous solubility continues to relegate potentially useful drugs to the sidelines as formulation techniques for intravenous and oral delivery lag behind the ability to produce and screen the libraries.

"IN CONTRAST TO SIMPLE BLOCK COPOLYMER MICELLES, THE IVECT™ METHOD UTILISES A MULTI-BLOCK COPOLYMER DESIGN, WHERE EACH SEGMENT (OR BLOCK) OF THE CARRIER HAS BEEN TAILORED TO ADDRESS THE MOST CRITICAL ISSUES FACING TARGETED DRUG DELIVERY."

Medicinal chemistry techniques can be utilised to alter the solubility of a drug by creating different salt forms or prodrugs. However, the pharmaceutical activity may be less potent, they may be slower acting due to the necessity of conversion to the active form, and these strategies may be very costly with long optimisation times.

Several strategies have been employed to formulate active pharmaceutical ingredients (APIs) in nanoparticle drug delivery vehicles to impart aqueous solubility and also have the added benefits of improved circulation time, minimised exposure of the API outside the target area, and have triggered release of the API payload at the target area. Moreover, their multi-functionality permits the incorporation of cell-targeting groups, diagnostic agents, and a multitude of drugs in a single delivery system.

Polymer micelles are particularly attractive due to their ability to deliver hydrophobic therapeutic agents; allowing for systemic delivery of compounds that are completely insoluble without a delivery vehicle. The foundation of Intezyne’s technology is based on the IVECT™ method for the production of stabilised polymer micelles.
Simple diblock copolymer micelles have been and are still being employed for drug delivery applications, but these systems often lack biocompatibility, post-administration stability, and effective strategies to target active cell targeting groups to their surface. Despite these drawbacks, block copolymers and polymer micelles still offer a number of chemically tunable features, such as high drug loading capacities and the ability to encapsulate a variety of therapeutic classes, which are not readily accessible using other technologies (for example, polymer-drug conjugates, dendrimers and liposomes).

In contrast to simple block copolymer micelles, the IVECT™ method utilizes a multiblock copolymer design, where each segment (or block) of the carrier has been tailored to address the most critical issues facing targeted drug delivery. This modular and versatile drug delivery platform can be chemically manipulated to accommodate a wide range of drugs, improve post-administration stability, vary the micelle size, control the release of the therapeutic, and target specific diseased cells.

Using this modular approach, Intezyne has created a cross-platform drug delivery system that has been carefully tuned to encapsulate and deliver small-molecule drugs, oligopeptides and proteins, DNA/RNA, and contrast agents for medical imaging, using only two separate polymers.

Each segment of the IVECT™ system has been constructed from biodegradable and/or bio-compatible building blocks, increasing the likelihood of achieving regulatory approval. Figure 1 shows a schematic of the IVECT™ system components, with hydrophobic amino acid (AA) blocks (yellow) designed to form the encapsulation core where hydrophobic small molecules (red spheres) are stably housed until the micelle is degraded. Stabilising AA blocks (green) keep the micelle intact once the formulation is diluted in the bloodstream. The hydrophilic poly(ethylene glycol) (PEG) block (blue) forms a protective corona around the micelle, giving the delivery system stealth-like properties to avoid protein opsonisation and the reticulo-endothelial system (RES). Additionally, the distal termini of the PEG blocks can be modified with peptide targeting groups that can actively target specific cells and potentially facilitate receptor-mediated endocytosis. Each block of the polymer has been tailored in order to fine-tune the size of the micelle for optimal delivery characteristics, as well as for optimal drug loading.

Initial development efforts for the IVECT™ method are focused on encapsulating chemotherapeutics for the treatment of cancer. One such new formulation utilising the IVECT™ Method is IT-141. The active pharmaceutical agent of IT-141 is SN-38, a topoisomerase I inhibitor, and the active metabolite of Pfizer’s Camptosar (irinotecan).

While SN-38 has exhibited excellent in vitro efficacy, its poor solubility in water (10 μg/mL) has thus far limited its clinical use. Using a proprietary triblock copolymer, Intezyne has successfully encapsulated SN-38 in the IVECT™ DDP at loadings of up to 15% (w/w) of the final formulation.

This formulation, IT-141, is freely soluble in water at concentrations up to 200 mg/mL. At this concentration of IT-141, SN-38 is effectively dissolved in water at 30 mg/mL, resulting in solubility that is more than a thousand times higher than free SN-38. Furthermore, SN-38 possesses a lactone ring that is unstable a physiological pH, rendering it inactive as an anti-cancer agent. Encapsulation of SN-38 within the micelle core shields SN-38 from the bloodstream thus inhibiting degradation of the active compound.

One of the unique aspects of polymer micelles is that they can effectively bypass renal clearance, and avoid the reticulo-endothelial system and subsequent uptake by the liver due to their inherent physical dimensions. In addition, the micelle’s particle size of approximately 50-150 nm allows for preferential accumulation in a solid tumour based upon the enhanced permeation and retention effect (EPR).1

Since solid tumours grow much more rapidly than their healthy counterparts, the blood vessels supplying the tumour are often ill-defined and possess a large number of pores, typically in the range of 200 nm. When introduced into this porous section of blood vessels, particles of 20-200 nm can readily enter into the tumour environment. However, these same particles have a difficult time diffusing from the tumour environment back into the bloodstream through another pore, and are essentially trapped in the tumour environment. Due to the design of the IVECT™ platform, IT-141 also possesses this advantageous particle size. Measurements made by dynamic light scattering (DLS) show IT-141’s particle size to be roughly 130 nm with a standard deviation of ±10 nm. Thus, the EPR effect allows for tumour targeting based solely upon particle size.

Herein lies a significant advantage of IVECT™ over of traditional polymer micelles. If the micelle is rapidly diluted to below its critical micelle concentration (CMC, or the concentration above which micelle formation is favourable), as occurs after a therapeutic is injected into a patient, it will degrade into a mixture of polymer and drug, and will not be able to utilise the EPR effect. Notably, IT-141 has shown micelle stability of up to 24 hours at biologically relevant concentrations in plasma, as characterised by in vitro DLS experiments. This greatly enhanced micelle stability is a result of the IVECT™ stabilisation block and translates to a longer circulation time, allowing the delivery vehicle a greater chance of localising at the site of disease.

IN VIVO EFFICACY OF IT-141

Initial efficacy studies focused on the comparison between Camptosar and IT-141 in human colorectal cancer using a HT-29 mice xenograft study. The study was performed with Figure 1: Pharmacokinetic Profile of SN-38 from IT-141 and Camptosar.
equimolar doses of both Camptosar and IVECT-encapsulated SN-38, then compared with a control group consisting of the IVECT™ polymer alone. As shown in Figure 2, IT-141 exhibits tumour inhibition of 107% (53% regression) opposed to 21% inhibition by Camptosar. Furthermore, only 25% of the tumours treated with Camptosar responded to treatment, while IT-141 elicited a response in 100% of the tumours. All mice remained healthy with stable body weight throughout the study.

Furthermore, a study was performed to explore the dose response of IT-141 on HT-29 tumour xenografts in nude mice and is represented in Figure 3. Doses of 5, 15, 30, and 45 mg of SN-38 were administered per kg of animal body mass (mg/kg). Animals were dosed by tail vein injection on day zero, four, and eight. While the 5 mg/kg dose exhibited no statistical deviation from the control, inhibition of 61% was observed for the 15 mg/kg group. Higher doses of 30 mg/kg exhibited tumour inhibition of 108%, with 111% inhibition for the 45 mg/kg dosage group. These results correspond to an average tumour regression of 61% and 88% respectively.

In addition, four out of seven mice treated with 45 mg/kg of IT-141 had no discernible tumour on day twenty of the study. Again, all mice remained healthy with stable body weight throughout the study and 100% of the mice responded to the treatment.

**PHARMACOKINETIC AND BIODISTRIBUTION PROFILE**

Pharmacokinetic and biodistribution data was generated from both healthy CD-1 mice and mice with HT-29 xenografts. IT-141 was administered by a fast IV bolus into the tail vein and plasma and organs were collected by cardiac puncture at times of 5 and 15 minutes, 1, 4, 12, 24, 48, and 72 hours with three mice utilised for each time point. Figure 4 shows the SN-38 concentration in plasma collected from HT-29 tumour-bearing mice over 72 hours.

Analysis of the plasma concentration versus time curve resulted in the following pharmacokinetic parameters: an area under the curve of 28.0 (hours x μg SN-38 per mL plasma) overall half-life of 4.1 hours, and a terminal half-life of 15.6 hours. This data exhibits marked improvements over Camptosar, which has an area under the curve of 3.2 and a terminal half-life of 3.5 hours.

**SAFETY AND TOLERABILITY**

The maximum tolerated dose (MTD) was determined for a single tail vein injection of IT-141 in both HT-29 tumour-bearing mice and healthy CD-1 mice. The mice’s body weight and overall health were monitored for seven days after injection. Mice that were administered with higher doses of IT-141 exhibited gastro-intestinal toxicity (swollen and/or necrotic large intestine and reduced spleen size), a routinely observed adverse side effect of camptothecin derivatives. Based upon survival rates and overall mouse health after one week, the single dose MTD was determined to be 60 mg/kg and the multiple dose MTD (3xQ4D) was determined to be 45 mg/kg in HT-29 tumour-bearing animals.

To further understand the potential toxicity from IT-141, histopathology was performed on HT-29 tumour bearing mice following three administrations of IT-141. Once tumour xenografts reached approximately 100 mm³ the mice were randomised into groups of 6-8 mice per group and treated with saline, IT-141 at 60 mg/kg, or IVECT™ polymer control (empty micelles). The sample was administered by a fast IV bolus into the tail vein once every fourth day for a total of three treatments. Vital organs and tumour tissues were collected on
day 18 of the study for histological processing by haematoxylin and eosin staining. Pathological analysis revealed the major toxicity of treatment to be neutropenia as determined by the presence of extramedullary hematopoiesis in the spleen. This was expected, as neutropenia is a known dose-limiting toxicity for SN-38. Slight liver toxicity was seen in some treatment groups, as evident by oval progenitor cell proliferation in the liver samples. No drug-related toxicities were observed in the heart, lungs, kidney or brain.

CONCLUSION

IT-141 represents an important advancement in the treatment of colorectal cancer by enlarging the therapeutic window, reducing toxicity, and greatly increasing efficacy when compared with current chemotherapeutic options.

Likewise, Intezyne has employed the IVECT™ method to encapsulate over 20 distinct chemotherapeutics, including taxanes, anthracyclines, topoisomerase II inhibitors, and kinase inhibitors, among others. Ultimately, the goal of Intezyne is to treat cancer better by enlarging the therapeutic window of cancer drugs by increasing the local drug concentration in the tumour environment and decreasing the toxicities associated by minimising the exposure of the free drug to the rest of the body.

REFERENCE:

UBM Live is gearing up for what promises to be the largest year ever of their flagship pharmaceutical ingredients event, CPhI Worldwide 2011, being held from 25-27 October 2011 at the Messe Frankfurt, Germany. Alongside co-located events, ICSE for contract and outsourced services, P-MEC Europe for pharmaceutical machinery and equipment, and the newly introduced InnoPack for pharmaceutical packaging solutions, UBM Live is projecting a large increase in attendance over the 2010 events held in Paris. Last year saw more than 28,500 attendees from over 140 countries visit the combined events, with over 1,900 exhibitors participating across the events.

“CPhI Worldwide and co-located events will return to Frankfurt in 2011 for good reason. The city is not only one of the most important business and financial hubs in Europe, the region is also one of the oldest and most established pharmaceutical markets in the world, serving as a home to branches of many major players in the pharmaceutical industry,” noted Annemieke Timmers, Global Brand Director, CPhI. “Frankfurt also provides easily accessible transportation and accommodation options for visitors and is a central location to travel to, both domestically and internationally. The city always receives positive feedback and is popular with attendees. We are delighted to be hosting the events here once again.”

New for 2011, UBM Live is introducing the CPhI Pre-Connect Virtual Event. On 8 September 2011, this event will be easily accessible from a PC or laptop anywhere in the world. CPhI Pre-Connect is not only an opportunity for companies who will be on-site in Frankfurt to get a head start with networking and lead generation prior to the show, but also allows companies that may not be able to attend the Frankfurt events the opportunity to promote their products and services to a targeted market.

As UBM Live’s events continue to evolve and grow in tandem with industry developments, the 2011 events in Frankfurt will see other new introductions and innovations. The success of the ICSE Packaging Zone in 2010 served as a platform for UBM Live to introduce InnoPack as a stand-alone event this year. InnoPack, focused on innovative packaging concepts in the pharmaceutical packaging market, is expected to host over 80 exhibitors and will be further complemented by the return of the ‘Packaging Trail’ to provide attendees with access to specific packaging needs within the other events being hosted. Also new to the events, LABWorld will make a debut as a pavilion within P-MEC to offer resources for instrumental analysis, measuring and testing technologies, materials testing, quality control and laboratory equipment for smaller scale environments.

Building on the success of the zoning that was introduced in 2010, CPhI Worldwide will host two new zones, Generic APIs and Finished Dosage, for attendees with specific interests in these areas. In addition to the General Floor, ICSE will host dedicated zones for Contract Research and Clinical Trials, New Exhibitors, the USA, and the new Logistics and Supply Chain Zone for logistics, cold transport and supply chain solutions for the pharmaceutical industry. ICSE will also feature the return of BioPh as a pavilion, in order to offer access to resources in the biopharma sector.

“The focus of these events is to continue to offer focused and comprehensive resources to our visitors,” commented Haf Cennydd, Global Brand Director ICSE, P-MEC, and InnoPack. “We conduct in-depth research around our events to garner feedback from exhibitors and attendees alike. New introductions such as InnoPack and the LABWorld pavilion reflect our ongoing commitment to keeping our events in step with the industry and providing highly relevant content.”

The Frankfurt events will also provide many opportunities for targeted learning and the discovery of new technological resources for guests to review in a hands-on environment. On Monday, 24 October, an educational ‘Pre-show Conference Series’ will be hosted that will cover a range of relevant topics and provide insights into new products and technologies. A ‘Lunch-time Educational’ series will also be hosted on each day of the show with a different topic for each day, including expectations for the future, strategies for success and compliance in emerging regions. Additionally, the CPhI, ICSE and InnoPack Speakers’ Corners and the CPhI Innovations Awards will both return in 2011. The CPhI, ICSE and InnoPack Speakers’ Corners allow exhibitors to present their company and services to a captive, qualified and highly interested audience while the CPhI Innovation Awards return for the 8th year to spotlight new and industry changing innovations from companies and organisations that are breaking new ground in the pharmaceutical, contract services, packaging and biopharmaceutical sectors.


The UBM Live annual schedule of pharma events also includes South America (21-23 March, 2012, Big Sight Exhibition Centre, Tokyo, Japan) and China (26-28 June, 2012, SNIEC, Shanghai, China).
When it comes to treatment of brain-related illnesses, the central nervous system (CNS) is one of the most difficult organs to reach. This unmet medical market is very large as it affects two billion people worldwide, a number which is expected to grow with increasing life expectancy and expanding global population. Although most drugs currently in development against neurological targets show high efficacy for the target, they often cannot reach the brain in sufficient amounts to exert an effect due to presence of the neuroprotective blood-brain barrier. This results in cessation of their development due to, for example, dose-limiting toxicity outside the CNS, or off-target effects. This includes the majority of the small molecules and nearly all of the large biotechnology drugs, such as recombinant proteins or gene-based medicine.

**THE BLOOD-BRAIN BARRIER: GATEKEEPER OF THE CNS**

The blood-brain barrier dynamically regulates the entry of compounds into the brain to meet the intrinsic requirements of the CNS. Neurotransmitters, nutrients and certain ions are able to enter, while potential neurotoxic compounds (including plasma proteins, cytokines, antibodies, drugs, bacteria and viruses) are actively excluded, effluxed and metabolised. Since the blood-brain barrier also keeps out most therapeutic compounds, there is unprecedented demand for new methods to deliver these potential new drugs into the brain safely. Currently, there are only few approaches for brain drug delivery and these are limited in their application, highly invasive, or disruptive to the neuroprotective blood-brain barrier.

**THE G-TECHNOLOGY**

The G-Technology is to-BBB’s core platform; it is a pegylated liposomal drug delivery technology with glutathione as targeting ligand at the tips of the PEG molecules. This provides a safe method to enhance drug delivery to the brain, as both components are already on the market; pegylated liposomes encapsulating chemotherapeutics, fungal

---

**ENHANCED DRUG DELIVERY TO THE BRAIN: SAFE AND VERSATILE**

Here, Willem van Weperen, MSc, MBA, Chief Executive Officer, and Pieter Gaillard, PhD, Chief Scientific Officer, both of to-BBB, present the G-Technology®, a glutathione-PEG liposome technology for the delivery of therapeutic molecules across the blood-brain barrier and to the brain.


The G-Technology is to-BBB’s core platform; it is a pegylated liposomal drug delivery technology with glutathione as targeting ligand at the tips of the PEG molecules. This provides a safe method to enhance drug delivery to the brain, as both components are already on the market; pegylated liposomes encapsulating chemotherapeutics, fungal

---

Mr Willem van Weperen
Chief Executive Officer
T: +31 71 3322255
F: +31 84 8313409
E: vanweperen@toBBB.com
to-BBB technologies BV
BioScience Park Leiden
Bio Partner Center II
Niels Bohrweg 11
2333 CA Leiden
The Netherlands
www.toBBB.com

Dr Pieter Gaillard
Chief Scientific Officer
T: +31 71 3322255
F: +31 84 8313409
E: gaillard@toBBB.com
investigate therapeutics ranging from small molecules safely enhanced the delivery of drugs to the brain, thereby exerting an effect in models for pain, brain tumors, viral encephalitis, and neuroinflammation. Furthermore, a technologic and mechanistic validation assay has shown that the free drug was delivered to the extracellular fluid of the brain. Increasing the amount of glutathione on the outside of the liposomes resulted in a free-drug concentration up to five times higher compared with non-targeted liposomes. Although the ultimate brain uptake and efficacy of any encapsulated compound will depend on the compound as well as the disease, to-BBB will be able to test and optimise the G-Technology almost for every specific situation.

STRENGTHENING THE PLATFORM

The versatility of the G-Technology comes from the liposomes, i.e. vesicles existing of a lipid bilayer with an aqueous core in which both lipophilic and hydrophilic molecules can be entrapped (see Figure 1). Furthermore, the addition of PEG gives the liposomes stealth-like properties as it minimizes scavenging of the liposomes by the body’s defence system, thus enabling a long circulation time in blood.

In the past year, to-BBB has entered several research collaborations to investigate the possibilities of the G-Technology further. The large research collaborations as well as with its lead technologic and mechanistic validation assay have shown that the free drug was delivered to the extracellular fluid of the brain. Increasing the amount of glutathione on the outside of the liposomes resulted in a free-drug concentration up to five times higher compared with non-targeted liposomes. Although the ultimate brain uptake and efficacy of any encapsulated compound will depend on the compound as well as the disease, to-BBB will be able to test and optimise the G-Technology for almost every specific situation.

<table>
<thead>
<tr>
<th>Targeting the blood-brain barrier</th>
<th>Drug carriers</th>
<th>Drug development from lab to clinic</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Proven inherently safe receptor biology in humans</td>
<td>6. No modification of active ingredient</td>
<td>8. Low costs &amp; straightforward manufacturing</td>
</tr>
<tr>
<td>2. Safe and human applicable ligand</td>
<td>7. Able to carry various classes of molecules</td>
<td>9. Activity in all animal models</td>
</tr>
<tr>
<td>3. Receptor specific binding</td>
<td>10. Strong IP protection</td>
<td></td>
</tr>
<tr>
<td>4. Applicable for acute and chronic indications</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5. Favorable pharmacokinetics</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Figure 2: Ten key development criteria for safe and efficacious drug delivery to the brain with all of its research collaborations as well as with its lead compound (see Figure 2).

These criteria are related to targeting the blood-brain barrier, drug carriers, and drug development from laboratory to clinic. Although the G-Technology is adhering to the criteria related to glutathione (as targeting ligand) and the liposomes (drug carriers), adherence to the last three criteria is also dependent on the compound that will be included.

To bring a drug to the market, straightforward manufacturing and low (or justifiable) costs are favorable, if not essential. Inclusion of recombinant proteins into the liposomes will increase the costs more compared with the inclusion of synthetic small molecules or peptides, but as long as these are justified and/or means of reduction of costs can be found in the production process (e.g. high encapsulation efficiency or recovery of non-encapsulated material) it will be possible to develop these products.
THE G-TECHNOLOGY IN PRACTICE: 2B3-101 AS PRIME EXAMPLE

The lead product of to-BBB, glutathione pegylated liposomal doxorubicin (2B3-101) is based on the marketed pegylated liposomal doxorubicin (Doxil®/Caelyx®). Therefore, this product is adhering to all ten key development criteria listed in the Figure. The active ingredient, doxorubicin, is active in all animal models. Furthermore, the existing production process of Doxil/Caelyx can be easily adapted at minimal costs to include PEG modified with glutathione.

The first clinical trial with 2B3-101 was initiated in June 2011. This trial is primarily designed to evaluate the safety of 2B3-101 in patients with solid tumors with brain metastases. However, specific efficacy data in patients with brain metastases from breast cancer will be obtained at the latest stage of the trial. Preclinical research with 2B3-101 has shown that 2B3-101 reduces brain tumor growth more efficacious than Doxil/Caelyx. Furthermore, 2B3-101 prolongs survival of mice with experimental brain tumors up to 60% when given at the maximum tolerated dose. Extensive GLP toxicity, safety and toxicokinetic evaluations have shown equal or less severe findings for 2B3-101 compared with Doxil/Caelyx. For the development of 2B3-101, pegylated liposomal doxorubicin (2B3-101) is produced according to cGMP standards by TTY Biopharm (Taiwan). The lead product has recently entered clinical trials. Preclinical research has shown that 2B3-201 prolongs half-life and increases brain uptake of methylprednisolone compared with the free compound, thereby increasing its efficacy at a much lower dose. Simultaneously, to-BBB will continue to work together with other pharmaceutical biotechnical companies to optimise the G-Technology for active compounds to be able to meet the unmet need of patients with devastating brain diseases.

FUTURE CHALLENGES

While to-BBB, together with its partners, has gained much experience about the possibilities of the G-Technology in the past years, many more challenges lie ahead. 2B3-101 as a lead product has recently entered clinical trials. However, to-BBB will continue to build the case by extending preclinical research. Furthermore, to-BBB is continuing its efforts for the treatment of devastating brain diseases with its second lead, 2B3-201, glutathione pegylated liposomal methylprednisolone, for the treatment of neuro-inflammation. Initial preclinical studies have shown that 2B3-201 prolongs half-life and increases brain uptake of methylprednisolone compared with the free compound, thereby increasing its efficacy at a much lower dose.

Simultaneously, to-BBB will continue to work together with other pharmaceutical companies to optimise the G-Technology for active compounds to be able to meet the unmet need of patients with devastating brain diseases.

The authors would like to acknowledge Corine Visser, Publication Program Manager at to-BBB, for writing and editorial assistance.

ABOUT THE AUTHORS:

Willem van Weperen has more than 15 years’ experience as a pharma/biotech leader. Before becoming to-BBB’s CEO, he held several global commercial, clinical and general management positions mostly at Genzyme. Pieter Gaillard is an entrepreneur and scientist who co-founded to-BBB and currently holds the position of CSO. With his extensive pharmacology and CNS experience he is globally recognised as an expert in the blood-brain barrier research field.

REFERENCES:

In this article, Jill Ogden, PhD, Vice-President of Business Development, and Richard Palmer, PhD, Executive Chairman, both of Warwick Effect Polymers, describe the company’s novel PEG and sugar-containing polymers, PolyPEG® and GlycoPol®, and their application to the challenges of half-life extension and drug delivery to and into cells.

Polymers are being used increasingly to modify and enhance the properties of both biological and small-molecule drugs. There is significant pharmaceutical R&D activity in these areas and half-life extension and efficient targeting and delivery of therapeutics are widely sought-after goals in drug development.

Although numerous injected peptide and protein therapeutics have been developed successfully over the past 25 years or so, several pharmacokinetic and immunological challenges are frequently encountered that can limit the efficacy of both novel and established biotech molecules. These molecules often have very short circulating half-lives, which limits them to frequent dosing schedules or prevents them from ever becoming useful therapeutic products. Conjugation of drug molecules to poly(ethylene glycol) (PEG), referred to as PEGylation, is a popular approach to addressing such issues.

PEGs are inert water-soluble polymers that can be conjugated to proteins and other therapeutic cargos by a variety of chemistries to increase the hydrodynamic volume of the cargo. As the kidney will efficiently remove most molecules that are less than approximately 20 kDa, the attachment of a large and inert polymer that increases the size of the therapeutic molecule will reduce and even avoid renal clearance.

In addition, PEGs can “shield” biologicals from interactions with enzymes and from inac-
tivation by the immune system. As a result, PEGylated drugs can exhibit prolonged half-life, higher stability, increased water solubility, and reduced immunogenicity. Therapeutic entities that can benefit from PEGylation include proteins, peptides, antibody fragments, oligonucleotides and nanoparticulate drug carriers. PEGylation of small molecules may also improve their pharmacokinetic and/or toxicological profile.

There is a range of PEGylated products on the market including Neulasta® (pegfilgrastim for the treatment of neutropenia; Amgen), Pegasys® and PegIntron® (both PEGylated versions of interferon-alpha for hepatitis C; Roche and Merck, respectively) and Cimzia® (certolizumab pegol for Crohn’s disease and Rheumatoid Arthritis; UCB). In 2010, global sales of Neulasta® alone were US$3.6 billion and combined sales of Pegasys® and PegIntron® were over US$2.6 billion; 2010 global sales of Cimzia®, approved in 2008, were US$280 million. The polymers used in these first-generation PEGylated products are linear and branched PEG species.

Over the years, PEG technology has steadily developed in respect of the chemistries for conjugating the PEGs to their therapeutic cargos, the polymer chain length and molecular weight, and the purity of the conjugates. In general, the polymers are conjugated to free amines on lysine residues, to thiols on free cysteine residues, or to the N-terminus of the protein, using succinimide, maleimide or aldehyde chemistry respectively.

Novel conjugation chemistries to attach PEGs to their therapeutic cargos have emerged as many researchers are looking to improve the efficiency of conjugation. For example, there are approaches focusing on site selectivity, use of potentially reversible linking technologies, different amino acid sites for reaction and the bridging of disulphide bonds in proteins. Most of this chemistry naturally extends from proteins and peptides to all types of RNA/DNA conjugations.

It is not clear how broadly new conjugation chemistries are being taken up and this will most likely be on a molecule-by-molecule basis, and probably where the tried-and-tested chemistries that are familiar to regulators have failed.

Some of the early PEGylated drugs were based on multiple attachment of small (<10 kDa) PEG chains, although this leads to heterogeneity amongst conjugates in the number of PEG chains attached. Such multiple site conjugation can also lead to loss of biological activity of the cargo.

In order to overcome these issues, more recent approaches to PEGylation now focus on attachment of a single larger PEG molecule to a region away from the critical areas of the therapeutic cargo. Such PEGs include larger linear and branched PEGs (e.g. the two arm 40 kDa PEG which is approved in Macugen®) and WEP’s proprietary comb-shaped PEGs, PolyPEGs®.

**POLYPEG®**

WEP’s next generation PolyPEGs® differ from linear or branched PEGs in that they comprise a poly(methacrylate) backbone with short pendent PEG teeth attached. This comb copolymer structure (Figure 1) also differentiates PolyPEG® from conventional PEGs, so that it is recognised as a completely different molecule from the patent perspective.

A considerable degree of structural diversity in PolyPEGs® can be achieved by varying the length of the poly(methacrylate) backbone and the size of the PEG teeth. Such flexibility in “polymer architecture” ensures the optimal PolyPEG® can be produced for the purpose. The PolyPEG® technology also has the advantage that larger molecular weight polymers can be produced than can be achieved with linear or branched PEG, while maintaining single site conjugation to the therapeutic cargo.

One of the key benefits of PolyPEGs®, compared with linear and branched PEGs, is their ultra-low viscosity, which is due to the ‘comb’ structure (see Figure 2). Viscosity can be a significant limitation of linear and branched PEGs when formulated at modest-to-high concentrations. Indeed, high-concentration formulations of biologicals are frequently required in order to administer sufficient drug for clinical benefit, and as this may
require large volumes or frequent dosing, this can limit patient compliance.

There is also an increasing need to formulate biological drugs for self-administration by patients, which requires devices with very small gauge needles and this can limit the potential of conventional linear and branched PEG products due to their viscosity.

A further advantage of PolyPEG® is that, unlike linear and branched PEGs, it does not accumulate in vacuoles in the liver and kidney when administered repeatedly. This could have significant advantages when regulators assess the potential toxicities for therapeutics that require frequent administration over long periods.

**GLYCOPOL™**

WEP’s proprietary glycopolymers, GlycoPols™, are designed to mimic the actions of complex polysaccharide molecules, such as glycans on glycoproteins, for example in their interactions with specific receptors on cells.2,3 This enables the targeting and delivery of a wide range of drugs to specific cells and tissues. GlycoPols™ can be conjugated to therapeutic entities, such as proteins, peptides, oligonucleotides, nanoparticles and small molecule drugs using standard conjugation chemistries.

The GlycoPol™ technology is based upon the facile reaction (click) of an azo-sugar on to a pre-formed polyalkyne polymer scaffold or backbone (shown in Figure 3). The scaffold is produced using living radical polymerisation techniques. Sugars are clicked to the scaffold via an azide typically in the anomeric (natural) position.4 As all sugars contain an anomeric position, it is possible to click mono-, di-, tri-saccharides and polysaccharides, as well as sialic acids and other branched sugars on to the scaffold to produce a wide range of glycopolymers. GlycoPols™ containing mixtures of sugars can also be formed.

Figure 3 illustrates the wide variety of structurally diverse GlycoPol™ molecules that can be synthesised that vary in backbone length, sugar composition and density.

WEP’s approach to producing glycopolymers is simple, straightforward and cost effective as it circumvents the synthesis of glycomonomers that are difficult to synthesise generically and are also prone to polymerisation problems. Glycopolymer libraries differing by sugar composition can be manufactured easily from polymer scaffolds and structure/activity relationships determined in relevant biological assays. This approach has been used successfully to prepare a number of “glyco-libraries”; the recognition of and binding of the glycopolymers to specific lectins (sugar binding proteins) demonstrates that sugars carried on the polymer backbone retain their structure and function.5

WEP’s living radical polymerisation process builds polymers in a highly controlled manner from a functional initiator containing the conjugating group. This allows glycopolymers to be made easily with a high level of control over their molecular architecture. It is also possible to produce block copolymers containing, for example, a glycopolymer block and a PolyPEG® block, or two different glycopolymers blocks – the permutations and combinations are endless.

**DRUG TARGETING AND DELIVERY**

In order to evaluate the potential of GlycoPols™ to target drugs to tissues, WEP has synthesised a range of prototype GlycoPol™ structures containing different backbone lengths, different sugars and mixtures of sugars. These were conjugated to biotin and screened for specific binding in human tissue microarrays against 27 normal tissues and to inflamed lung and liver tissues. Certain GlycoPols™ were shown to bind selectively to several tissues, including epithelial cells in breast, lung and prostate tissues and in the...
liver (see Figure 4). The binding was specific, as indicated by the observation that binding could be blocked by pre-incubation with non-biotinylated GlycoPols™.

In a separate study, binding of biotin-labelled GlycoPols™ to ten different normal and matched cancer tissues was assessed. This showed selective binding of certain GlycoPols™ to some cancer tissues with more intense binding being observed as compared with the corresponding normal tissues. Figure 4 illustrates the differential and selective binding to Non-Small Cell Lung Cancer (NSCLC) tissues compared with normal lung.

These results demonstrate the potential of GlycoPol™ for targeting and delivery of therapeutics to specific cell and tissue types. Several parameters, such as length of the backbone, sugar content and composition, can all be modified to fine tune a GlycoPol™ and optimise it for a specific target.

WEP is also exploiting GlycoPol™ for the delivery of RNAi-based therapeutics. RNAi has been seen as a breakthrough technology in selective and novel therapies but is still hampered by the continued challenge of delivering the active molecule into the target cell. Polymer delivery systems are being actively explored, however, the combination of polymer properties required, together with pharmaceutically appropriate standards of manufacture, requires a more sophisticated and controlled polymer technology.

WEP’s GlycoPol™ technology can meet these needs through controlled synthesis of block copolymers and the diversity of molecular features that can be incorporated into a single copolymer element. To illustrate this potential, a specific GlycoPol™ conjugated to an siRNA has been shown to bind selectively to cultured cells and to be internalised (Figure 5) resulting in target gene knockdown.

In summary, GlycoPol™ offers an innovative approach to the targeting and delivery of a wide range of therapeutic cargos from proteins to oligonucleotide and RNAi therapeutics and even small molecules. The ability to target drug cargos to the optimal site of action based on the specific and selective interaction of polymer-borne sugars with carbohydrate receptors on cells and tissues opens up exciting possibilities.

Advances in molecular therapeutics continue to generate many new opportunities for novel treatments. An alignment of polymer chemistry with biology offers great potential in addressing some of the complex issues associated with the formulation and delivery of these innovative agents leading to a new generation of successful therapeutics.

ABOUT WEP

Warwick Effect Polymers Limited (WEP) is a developer and manufacturer of high-value, novel functional polymers for enhancing therapeutics. The company is responding to the increasing need in the pharma industry for solutions to the complex delivery and formulation issues associated with new generations of biotherapeutics. It combines its proprietary polymer chemistry catalyst systems and considerable expertise to produce a range of novel polymers focused on addressing these needs.

REFERENCES:

Experience Makes the Difference
With nearly 80 years of parenteral expertise, we can help to navigate the pathway of success for your molecule. We can help you overcome technical challenges, including process and formulation development, technology transfer, and analytical methods development. Our scientific teams are skilled in the development of solutions, suspensions, and freeze-dried dosage forms to help optimize product quality throughout manufacturing. BioPharma Solutions provides our clients with confidence of delivery, service, and integrity — we know the work we do is ultimately vital to the patients you serve.

For more information, visit baxterbiopharmasolutions.com or visit us at ICSE booth # 41F04