Peptide drugs, overcoming the challenges, a growing business

INTRODUCTION

Due to the advances achieved in the peptide field the last few years, this class of therapeutics has continued gaining more interest. Thus peptide drugs became a focus for several pharmaceutical and biotech companies. They are used in different therapeutic areas like allergy, anti-infection, diagnostics, oncology, diabetes, obesity, cardiovascular, arthritis, etc. Beside others, Peptide drugs have the following advantages:

– they have high activity and specificity (little unspecific binding to molecular structures other than desired target)
– they are often very potent
– they have minimized drug-drug interactions
– they are less accumulating in tissues and therefore have a low toxicity
– they offer a biological and chemical diversity, which is also easy to investigate from the therapeutic point of view

Peptides have also disadvantages which limited their success until last few years. These limitations are mainly related to their stability in the body resulting in:

– short half-life and low bioavailability. Peptides are cleared very quickly from the body.
– formulation challenges. Progresses in drug delivery technologies have substantially contributed to newly considering peptides as an exciting therapeutic indication
– challenging and costly synthesis. Some of the problems encountered during synthesis like solubility and aggregation constitute hurdles during manufacturing.

The overall average success rate from clinical trials in phase I to drug approval for a peptide is in the range of 11 per cent. This percentage depends on the therapeutic area. It is much higher (almost doubled) in the cardiovascular class and lower for peptide drugs in the oncology field.

WHAT ABOUT THE PEPTIDE DRUGS MARKET?

In 2003 the classical peptide drugs market reached approx. 6.4 billion $. This numbers are not including the peptidomimetics where the only angiotensin converting enzyme (ACE) inhibitors and HIV classes exceed the 5 billion $ sales. There were more than 720 peptide drugs/candidates in several therapeutic areas in 2004. Among them 5 per cent on the market and the rest in clinical trials 38 per cent (phase I, II and III) and advanced pre-clinical stages 56 per cent. These numbers reflect the growth in the peptide field and show a sustained future business.

Assuming the expected growth (8-10 per cent), the peptide drugs market should cross the 10 billion $ barrier in 2010.

OVERCOMING THE CHALLENGES AND ACHIEVING SUCCESS

The overall timeline for developing a drug through the commercialization is in the range of 7 to 12 years. Small and mid-sized companies have often a shorter process driven by their finances. The main hurdle in developing peptide drugs is their stability. Solubility and pH (this later affects the bioavailability) represents important characteristics to be considered during drug formulation.

Peptide sequence modifications

To overcome the short half-life and low bioavailability several technologies have been investigated. Among these tools to enhance the stability, we find peptide backbone modification, like introducing unnatural amino acids or D-amino acids, peptide bond modification (b-amino acids, reducedamide bonds...) constraining the backbone by introducing cyclizations or constrained amino acids resulting in less flexibility and enzyme digestion. In some cases, conjugation like attaching a fatty acid or PEG-derivative represents good tools to achieve acceptable peptide stability. In these last two examples the modified peptide drug substance constitutes a new chemical entity that needs additional toxicological investigation. Attaching a fatty acid to a peptide may not only result in increased half-life and bioavailability (1) but can also lead to more specificity binding to a target and therefore fewer side effects.
Drug delivery technologies (DDT)

Other techniques to improve the pharmacokinetics (PK) are drug delivery technologies (DDT) and the recent advances in this field in order to modulate the in vivo stability. Since most peptide therapeutics are exerting their action on the cell surface, by binding to the membrane proteins, various techniques have been developed for intracellular delivery. Basically this can be achieved using two main approaches:

1) liposomes or nanoparticles fusing with or penetrating the cell membrane to release their content into the intracellular domain.

2) short peptide sequences called protein transduction domain (PTD) allowing delivery of peptides or proteins they are fused with into the cell.

The liposomal formulation often suffers from the active pharmaceutical ingredient (API) encapsulation efficiency into liposomes. However, this technology still represents, in some cases, the best solution for certain drugs formulation. The recent approval of inhaled insulin, Exubera (Pfizer), will add some excitement in the field of DDT hopefully leading to more success for peptide formulation and open more room for further market growth.

API MANUFACTURING

One of the main tasks in drug development is adjusting the costs according to the risks and probability of achieving success. However a certain development level (processes for both manufacturing drug substance and drug product) is highly recommended since implementing changes and potential optimizations during clinical trials will impact the global costs allocated to a project. As an API manufacturer we see virtual and small companies with limited resources to address these needs. Therefore, these companies are not only outsourcing the API manufacture but the documentation and regulatory activities as well. Larger pharmaceutical and biotech companies perform some of these activities in house. The contract manufacturing organizations (CMOs) are therefore a long term partner, for several years, during the clinical trials by supplying “right the first time” materials, services and responses to the authorities in order to support the projects. The increasing regulatory requirements have reduced the Far East competition and the peptide supply chain remained at amino acids derivatives and to certain extent peptide fragments. The majority of the peptide API manufacturing remains under control of both European and American companies.

Process development

Appropriate process and process development support good documentation and filing. Several components must be considered for a cost effective synthetic strategy for an API production. Listed are points we consider important:

- Customer requirements or specifications, may affect significantly manufacturing costs and delivery time. The process should deliver the expected purity and yields and allow identification of the critical process parameters, proven acceptable ranges (PAR) to support the most efficient way the documentation and the definition of the design space (6).
- Stage of development: an important point, especially for quality and compliance requirements. The efforts/costs related to regulatory activities increases during drug development.
- Targeted volumes at commercial scale. This is the driver for the choice of a synthetic strategy. API costs saving starts at this stage and all consequences resulting from implementing optimizations or changes in later stages generates much higher costs because of their impact on different areas of development.

Supply chain

By choosing or developing a process, one should consider the supply chain early, especially when targeting high API volumes. A fragment condensation strategy for example could involve, for large scale, cost effective outsourcing of early steps for achieving better economy of scale. Whereas a straightforward solid phase strategy will have its supply chain management stopped at the amino acids level. We know from the recent Trimeris T-20 project that the amino acids derivatives have reached their plateau prices and further optimization is very unlikely to occur. Many of these amino acids (and also the 2-chlorotriethylchloride resin) have seen their prices dropped by a factor 8 to 10 in the last 8 years. These are key considerations in designing a process for better success.

Solid phase or solution phase peptide synthesis?

With the above mentioned components one should pick the most appropriate approach to the project. It can be solid phase peptide synthesis (SPPS) or liquid phase peptide synthesis (LPPS) or even a combination of both. Some CMOs have more affinity with SPPS, others prefer LPPS. Only few have good experience in both areas. How does the synthetic approach impact costs? Of course, the costs structures in different CMOs may advantage one or the other strategy. However the following should apply independently on how the costing is structured. The purification remains the bottle neck in peptide manufacture. The crude peptide purity (before potential preparative HPLC purification) is the most critical parameter in the process. Thus, the down stream processing can be the most expensive part because of yield losses during purification to reach the targeted specifications.

- LPPS might be more suitable for a short peptide sequence (for example <15 residues) especially if large volumes are targeted later on. Through subsequent isolation of the intermediates their purity is enhanced and the final resulting API has usually higher purity compared to material obtained by SPPS. The final preparative HPLC purification, when needed, is less extensive and the desired quality can be potentially obtained by one single pass. However, even if certain steps can be performed in parallel, the time required for an LPPS process is often longer than for a SPPS process. Nevertheless the added value is high, especially when the volumes exceed the 5-10kg. This strategy offers fragment synthesis possibilities at large scale (supply chain considerations), they can be stored shortening therefore the delivery time and allow responding quickly to the market demand. These intermediates can be well characterised and considered as starting materials for future process validation. As an example from our experience the LHRH analogues are typically obtained in good yields by LPPS and >97% per cent purity undergoing a high throughput during purification achieving very cost effective overall process.
- For long peptide sequences (for example >20 residues) SPPS remains the most appropriate even...
some longer peptides have been achieved by LPPS. Peptide fragments can be produced by SPPS and further coupled either using SPPS or LPPS. One of the limitations in SPPS is the high raw material costs, solvents and waste volumes. Typically 3-6000kg solvents and waste are handled during the synthesis of 1kg 20-25-mer peptide API (excluding the final purification and down stream processing). Extensive HPLC purification is required. But SPPS has shown its practicability at very large scale and the CMOs have overcome these related logistical issues.

This is summarised in the following table:

<table>
<thead>
<tr>
<th>API volumes</th>
<th>Short peptides</th>
<th>Long peptides</th>
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<tbody>
<tr>
<td>High</td>
<td>LPPS</td>
<td>SPPS/LPPS</td>
</tr>
<tr>
<td>Low</td>
<td>SPPS/LPPS</td>
<td>SPPS</td>
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Since the down stream processes such as preparative HPLC represents the capacity bottle neck in the peptide manufacturing, extensive efforts have been employed to increase the crude purity going into the purification step. Several tools like choice of the resin, coupling reagents etc have been used to optimize the processes. The use of pseudoproline building blocks (2, 3) in peptide synthesis have shown substantial advantages. Even their benefit in the synthesis was demonstrated more than 15 years ago by Manfred Mutter; their application in the industrial field is still very limited probably due to their difficult synthesis. Pseudoprolines (shown below) are protected dipeptides where the second amino acid is a serine, threonine or a cysteine residue cyclised between the side chain hydroxyl/thiol group and the amino function. This temporary protection can be easily removed under acidic conditions usually during the final peptide cleavage from the resin. These compounds can be coupled using standard coupling reagents. Their coupling proceeds with high yields and racemisation free (since the oxazolone formation, responsible for epimerization, can be excluded as the amino function is involved in the oxazolidine ring). Since we are producing these derivatives on a regular basis we are using them in our peptide synthesis. We have observed substantial difference in purity (often in the two digits level) between conventional synthesis strategy and use of pseudoprolines confirming the reported literature data (3, 4). Aggregation constitutes a major challenge the peptide chemists are facing during synthesis. Thus some sequences tend to aggregate by self-association (b-sheet formation) for example. The pseudoprolines break these structures (3) as a proline residue do in a peptide sequence, since the amide bond is adopting a cis conformation (5) leading to better solvation (3) and accessibility of the chain for further coupling steps. The cysteine pseudoprolines have shown some limitations in their use in the peptide synthesis due essentially to the high thiazolidine ring stability requiring stronger acidic conditions for deprotection (like trifluoromethane sulfonic acid in trifluoroacetic acid). These deprotection conditions for deprotection (like trifluoromethane sulfonic acid in trifluoroacetic acid). These deprotection conditions are depending on the peptide sequence and should be looked at as a case to case study. It is very important to mention that a certain level of efforts at the API stage (appropriate chemical process) and at the drug product stage (appropriate drug delivery technology) will result finally in time and resources saving during pre-clinical studies. This consideration can be summarised with the following figure. Thus investing in early stage processes for both API and drug product increases the success rate and can result in substantial time and costs saving.

How do the CMOs respond to the peptide market growth?

There are mainly two different reactions in responding or acquiring more peptide business for the CMOs.

- Capacity expansion: By building or acquiring more and larger production capacities. This strategy targets mainly large volumes projects and facilitate business acquisition at later development or even at commercial stage.

- Integration of new knowledge through technology expansion or alliances. This allows the corresponding companies to provide wider services supporting therefore the projects in several areas.

Through acquisition of UCB-Bioproducts early this year, Lonza added LPPS expertise and experience in handling commercial peptide projects to their large SPPS manufacturing capacity. However, the global product portfolio may have been affected since both companies could have been seeking for both large projects. Other companies like Bachem added recently the cytotoxic manufacturing to their capabilities. The company has been successfully looking into new areas providing building blocks and other tools for peptide synthesis complementing nicely its existing business. Some of the mid-sized CMOs like Genzyme Pharmaceuticals are providing both peptides and drug delivery products. The company has signed an alliance with Mimotopes for research peptides material. The other alliances in the drug delivery field with both Pharmidex (CNS delivery) and Brookwood Pharmaceuticals (for specially designed delivery) offers the customers a “one stop shop” and efficiency in the drug development. Thus the company is able to provide from the first unnatural amino acid (usually produced either by asymmetric hydrogenation or enzymatic routes) to the final API and drug product through its partners.

CONCLUSION

The developments in the peptide drugs area in the last decades were concentrated on their chemistry, methodology and their mode of action. On the other hand, chemical modifications and drug delivery technologies have contributed improving the peptides pharmacokinetics and stability resulting in the success of this class of therapeutics. The process development remains an important parameter to be considered for costs and time efficient peptide drug development.

REFERENCES AND NOTES

6. Guidelines ICH Q8

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