HOW TO INJECT SUPERCRITICAL CO2 IN A PHARMACEUTICAL PROJECT ?

LOCHARD Hubert, MARCIACQ Florence, FREISS Bernard
Pierre Fabre Médicament, CDFSC, 16 rue Jean ROSTAND, BP 92, 81600 Gaillac Cedex
hubert.lochard@pierre-fabre.com

The use of high-throughput screening and similar techniques in drug discovery provides new candidates that present sometimes high molecular weight and low water solubility. Therefore, researchers have to find new techniques that permit to formulate and to increase the bioavailability of these active pharmaceutical ingredients.
Among different technologies, supercritical fluids technology has already permitted to enhance bioavailability of active substance, either by increasing the solubilisation kinetic or by modifying the apparent solubility of the molecule.
The Supercritical Fluids Division of Pierre Fabre Laboratories, has been involved in supercritical fluids for nine years. Most of our works has been carried out on supercritical CO_2.

Complexation with cyclodextrins

Cyclodextrins, a family of oligosaccharides, are commonly used to form inclusion complexes with hydrophobic organic molecules in order to enhance the Active Pharmaceutical Ingredient (API) apparent solubility.

Because the outside surface of these molecules is hydrophilic and the inside surface hydrophobic, they are able to include, fully or partially, in their cavity large organic molecules by non-covalent bonds. Physical and chemical properties of the included molecules may thus be favourably modified, and in particular the physical stability and the aqueous solubility can be improved.

Several techniques have been used to prepare inclusion complexes, like kneading, freeze-drying, grinding, co-precipitation and melting. However, due to the poor aqueous solubility of many non-polar drugs, the energy or process cost, they seem less interesting than supercritical CO_2 complexation.

Pierre Fabre Medicament has developed an extensive expertise in Supercritical Fluid technology, focused on the development of new tools for the formulation of active ingredients. Among the processes developed, FORMULPLEX® is a patented complexation process with cyclodextrin in a powder form to improve bioavailability of poorly soluble drugs.

Because of physical properties of supercritical fluids (density, viscosity, diffusion constant, mass transfer…), thermodynamical constants and reaction kinetic of the complexation are different than those observed with other complexation processes. Therefore complexes obtained in supercritical CO_2 present structure characteristics different from those obtained with other techniques. An interesting property is that in many cases, the interactions between the active ingredient and the cyclodextrins are more efficient, resulting in a higher apparent solubility of the complex obtained.

Another interesting feature is that, in some cases, a complexation that is not obtained by conventional methods occurs in a supercritical medium. API/Cyclodextrin complexation process is equilibrium, which is dependent on the medium. The process leading to an inclusion complex is totally “green” using only carbon dioxide and a catalytic amount of water.

In certain cases, an enhancement of the stability of APIs in aqueous media has been observed. Furthermore, this technology does not require any mechanical mixing. This is an important issue to avoid any local over-heating of the material and thus a potential degradation of temperature-sensitive molecules.

Formulplex® is a single-step complexation process. Adding supercritical carbon dioxide on a mixture containing the API, the cyclodextrin and a catalytic amount of water enables to perform the complexation. The operating conditions to be set up are the raw material ratios, pressure, temperature and reaction time. At the end of the step, a depressurisation permits to obtain the complex in solid form without any residual solvent.

This simple and reproducible reaction allows us to achieve a rapid and reliable scale-up.

The results gathered in the table underline the effectiveness of the technology Formulplex ® for various types of drugs. It presents aqueous solubility after 15 min in a 37°C aqueous solution. The cyclodextrins used for the trials are α, β, γ, hydroxypropyl-β and methyl-β-cyclodextrins.

The water-solubility of the complexes obtained is presented in comparison with the “physical mixture” that is the complex obtained in water.

Product	Cyclodextrin	Molar Ratio (API / Cyclo)	API Solubility (µg / mL)	Physical Mixture (µg / mL)	API solubility after Formulplex1 (µg / mL)	Complexation Ratio	Formulation
Antiseptic	Methyl - β	1/2	0	2	400	nd	Lotion
NSAID	β	1/2	200	542	5000	100%	Capsule
Drug in development	β	1/2	0	8	170	nd	Capsule
Anti-Histaminic	β	1/2	0	500	1320	95%	Tablet
Anti-cancerous	Methyl - β	1/5	0	0	184	100%	Capsule
NSAID	β	1/1	8	10	870	100%	Tablet
NSAID	HP - β	1/2	157	1338	4183	100%	Tablet
Anti-cancerous	HP - β	1/2	250	327	739	nd	Oral Form
Cox2	Methyl - β	1/2	134	157	824	100%	Tablet
Statin	β	1/2	1	1	300	100%	Tablet

Figure 2: API content – Dissolution tests conditions: buffer 6,8 – 37°C – solubility after 15 minutes.

A significant increase in solubility is observed through the implementation of FORMULPLEX®. These solubility results are usually predictive of the biovailability of the complexed active ingredient.

As case study, complexation of a Non Steroidal Anti Inflammatory Drug (NSAID), ibuprofen, is presented. Different formulations were tested in animals in order to compare ibuprofen treated with Formulplex® and ibuprofen formulated with the same excipients. This pre-clinical tests permit to choose a formulation and to compare it with marketed ibuprofen's formulation on human.

Comparative pharmacokinetic study was performed after oral administration on rats. Plasmatic level of ibuprofen were measured and the maximal concentration (Cmax) and the Area Under the Curve (AUC) were plotted for the different formulations.

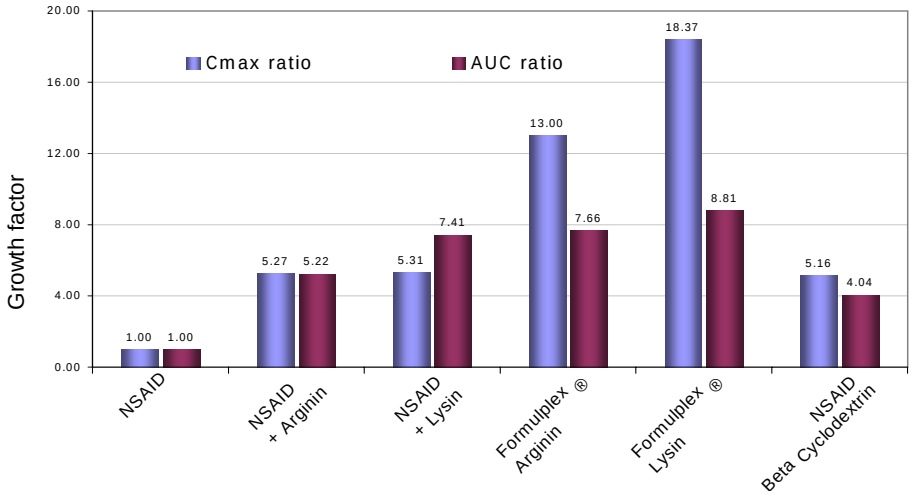

Figure 3. Pre-clinical study on rats: evaluation of different formulations versus non-processed Ibuprofen

Industrialisation of the technology

The Supercritical Fluids (SFC) Unit of Pierre Fabre Medicament is located in Gaillac (France). Equipments are designed to develop Formuplex® processes, to perform the scale-up of the process and to realize the first commercial batches.

The Supercritical Fluids Division has been set up to conform with all the requirements given by ICHQ7A, cGMP and BPF (Bonnes Pratiques de Fabrication, last inspection of the French Health Authority in June 2008).
SCF development area has been designed in order to be the most exhaustive as possible.
We handle powders from a few grams up to hundreds of grams for development phases.
The state-of-the-art equipment allows a large flexibility and adaptability:
- Stirred Autoclave from 250 ml to 50 L
- Pumps from 3kg/h up to 50 kg/h

SFC Production area has been designed in order to fit two main goals:
Manufacture of phase I, II, III clinical studies batches
Manufacture of commercial batches

Figure 8: area dedicated for the production

Conclusions

Through the complexation of a poorly water-soluble API with a cyclodextrin, it is possible to enhance its solubility. A non-water-soluble API can be complexed, resulting in an increase in water solubility by over 300 folds. More recent trials have led to some higher improvements, by up to 900 times.

In achieving this goal in terms of bioavailability enhancement, the supercritical CO_2 technology FORMULPLEX® shows many advantages over standard methods of complexation. In some cases, a stabilisation of the API also has been observed. The cost and reaction time are dramatically reduced and, in some cases, complexation not observed with classical methods occurs in a supercritical medium.

Technologies with Supercritical CO2 don’t use organic solvent and the carbon dioxide is always recycled.

An important point is that if you increase the bioavailability of an active ingredient you can reduce the concentration of this one in each tablets or capsules for the same effect and , in this way, you have reduced the level of active ingredient reject by the body. The effect on environment (concentration of drug in water and effect on fishes for example) could be major.

We develop other applications around the use of supercritical fluids. A specific process to coat small sized particles (less 50µm) without the use of organic solvent and an other to obtain a solid dispersion, always without use of organic solvent.

All the technologies developed around the Supercritical Fluids (especially carbon dioxide) correspond completely to the domain of the green chemistry. From an industrial point of view the scaling-up of these processes is relatively simple and their cost price less important than that could seem.