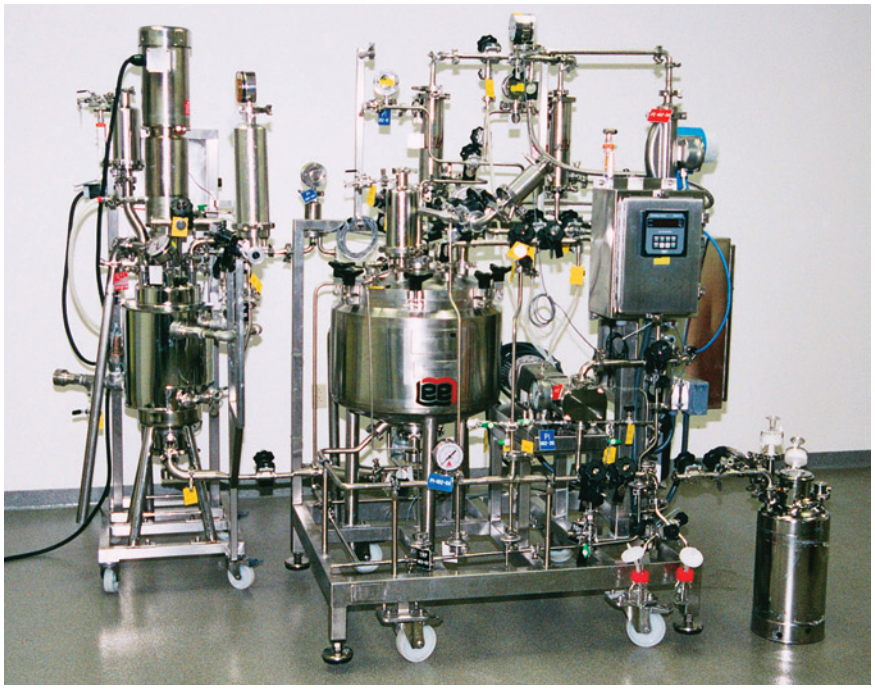


ProMaxx CGMP equipment suite for water-based formulation of microspheres.

Epic Microsphere Technology Good Match For Baxter

At the heart of Baxter Healthcare Corp.'s acquisition of Epic Therapeutics Inc. is the latter's microsphere formulation technology. Epic's "ProMaxx" technology provides the perfect complement to Baxter's own NanoEdge formulation technology.

Baxter's NanoEdge provides for the formulation and delivery of poorly soluble, low-molecular-weight drugs while allowing for sustained release. Epic's ProMaxx technology, by contrast, allows the creation of uniform microspheres for formulating such molecules as proteins and peptides for immediate or controlled release.

Baxter sees ProMaxx as a key component in building its drug-delivery business. Already established in i.v. bags, the company has been working hard the past few years to expand beyond that niche into high-end, high-tech drug delivery with NanoEdge.

"ProMaxx and NanoEdge formulation technologies are complementary in that they enable our pharmaceutical and biotechnology clients to develop engineered drug particles out of small and large molecules that have challenging formulation or delivery needs," says David F. Drohan, senior vice-president and president of Baxter's medication delivery business at the time of the acquisition announcement.

ProMaxx is a proprietary water-based delivery technology that can be used for a variety of pharmaceutical agents, including proteins, peptides, small molecules, and DNA. The platform produces bioerodible protein microspheres to deliver both small and macromolecular drugs. Both microsphere size and drug release characteristics can be tailored to the desired therapeutic application. The microspheres can be used for

Microsphere *continued on page 22*

Micro- and Nanoparticle Systems Show Promise for Oral Dosage Forms

Researchers at the University Henri Poincaré (UHP, Nancy, France) are working on an innovative microparticle-based technology for improving the oral absorption of heparin, a drug commonly used for the treatment of venous thrombosis. The study is part of an effort for developing injectable macromolecular drugs into oral dosage forms, thereby avoiding the disadvantages typically associated with parenteral administration.

Professors P. Maincent, N. Ubrich, and C. Vigneron of UHP's Pharmacy College in collaboration with Evelyne Vuaridel, PhD, director of the Galenic Unit of Debiopharm S.A. (Switzerland) have filed a patent application on the particle carriers developed to improve

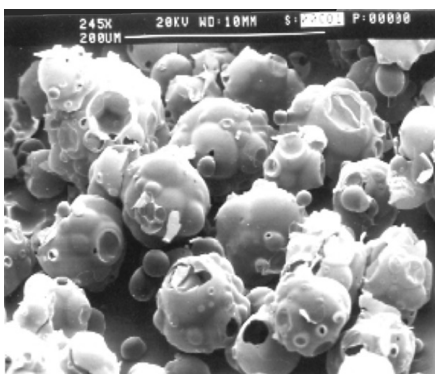


Figure 1: Electron microscopy photograph of heparin-loaded microparticles produced with a blend of PLGA and Eudragit RS (×245).

the oral absorption of large biomolecules such as low molecular weight heparin (LMWH), peptides, and proteins. The group developed various polymeric formulations of microparticles of unfractionated heparin (UFH) and LMWH using blends of biodegradable and polycationic polymers to

examine the feasibility of oral delivery of the macromolecules (see Figure 1).

Under normal circumstances the hemostatis process prevents spontaneous bleeding and blood loss from ruptured blood vessels through clot formation while maintaining the circulating blood in a fluid state. In the absence of hemostatis, clots can form and block a vessel (vein or artery), which can sometimes lead to thrombosis or other more severe complications such as pulmonary embolism.

The major disadvantages associated with the injectable administration of heparins to treat venous thrombosis are well known and have been described extensively in the literature (see *Pharmaceutical Technology*, March 2002, page 38). In contrast, oral formulations can reduce healthcare costs by lessening or eliminating the need for medical assistance or ongoing patient monitoring. In addition, researchers note that "plasmic levels of heparin remain more constant with an oral sustained-release formulation, which could potentially have more sustained efficacy."

A feasibility study performed using rabbit models focused on LMWH to identify the best parameters with regard to oral bioavailability and prolonged release of the active ingredient. Components of the formulation were the polymers poly (epsilon caprolactone) (PCL) and poly DL-lactic-co-glycolic acid (PLGA), which are used for parenteral administration in humans, as well as polymethacrylates (Eudragits), which have established use in conventional tablets as granulating aids and coating agents.

The microparticles containing LMWH were produced with approximately equal parts of the biodegradable and nonbiodegradable polymers. Polymeric microparticulate formulations of LMWH (600 IU/kg) then were orally administered. The researchers obtained high absolute and relative bioavailabilities with microparticles prepared with Eudragit (RS/RL) and PLGA.

According to the researchers, "the structure of the microparticles allows the active principle to be protected from degradation until its release at the site of absorption. The powdered formulation is homogeneously dispersed in the gastrointestinal tract, allowing close contact with the mucus layer." Results also showed an increased duration of antithrombotic activity and "excellent" bioavailability after administration of LMWH to rabbits.

This study of heparin-loaded microparticles follows a successful experiment in which researchers tested an insulin nanoparticle system composed of PCL and Eudragit RS in diabetic rats. In this work, PCL–Eudragit RS insulin-loaded nanoparticles and empty nanoparticles (for control experiments) were formed by a water–oil–water solvent evaporation technique. The nanoparticles were administered by gavage in streptozotocin-induced diabetic rats, and blood glucose and insulin levels were measured for as long as 24 hours after gavage. Oral glucose tolerance tests were performed 4 and 8 hours after gavage. Results showed that the size of the insulin-loaded nanoparticles was 358 ± 11 nm; the yield of encapsulation of insulin was superior to 98%; insulin nanoparticles (100 U/kg body weight) administered by gavage to fasted diabetic rats significantly reduced blood glucose levels; insulinemia was increased; and the peroral insulin nanoparticles lowered the glycemic response to the oral glucose challenge.

Debiopharm has licensed the microparticle technology for heparin delivery, which, according to the company, will cover all the technical activities required for developing an encapsulated polymeric system from laboratory scale to pilot batches for clinical trials and full-scale industrial manufacturing. Meanwhile, the research group's next step is to test other animal models to obtain additional data. In the future, it is considering working with peptides, insulin, and other proteins.

Maribel Rios

DSC Process Accelerates Protein Screening

Protein formulation for the development of biopharmaceuticals has put new demands on the pharma industry to accelerate screening processes and develop ways to keep the substances stable and biologically active during storage. To address this imperative, researchers at MicroCal, LLC (Northampton, MA), have developed a differential scanning calorimetry (DSC) process that is designed to streamline the screening of liquid formulations for thermostability through increased sample throughput and automated multiple-sample screening.

FDA identifies proteins as drugs and as such requires that all biopharmaceuticals and their formulations undergo stability tests, both real-time and accelerated. In the testing process, the biopharmaceutical is stored and then assayed at various intervals during the course of several months and for as long as two years. As a result, biopharmaceutical production companies are on the

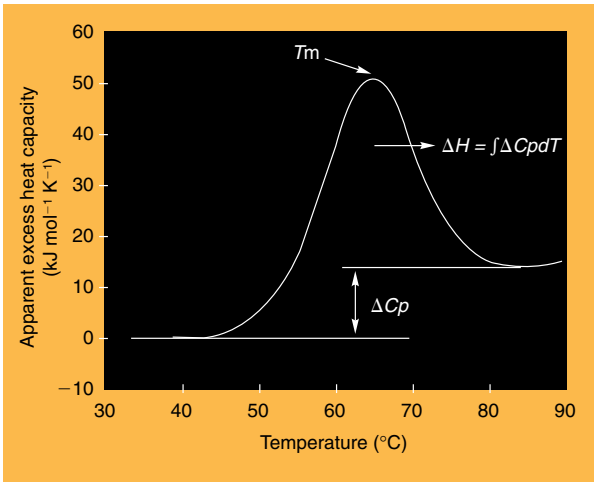


Figure 1: A typical DSC thermogram. This DSC scan was used on a dilute protein solution in which the protein underwent a transition from a compact, native state at low temperature to an unfolded, denatured state at high temperature.

lookout for methods to increase throughput during the testing processes to speed a drug's time to market.

Protein formulations that are supplied as liquids must maintain stability in solution for a period of time that meets FDA's requirements for shelf life. Proteins unfold when they are heated or when denaturing chemicals such as buffers and solvents are added to the solution, making the protein more likely to aggregate and therefore destabilize and break down. Determining a formulation's ability to remain stable and bioactive during production, packaging, storage, and shipping is crucial to the success of the product.

DSC is currently the only way to measure thermodynamic parameters of proteins as well as other substances such as lipids and nucleic acids. Using DSC, a formulations scientist can prepare a protein in various solutions. All the proteins will unfold during a DSC experiment because the protein is being heated. As shown in Figure 1, a DSC trace indicates the temperature midpoint of the transition for the enthalpy change (T_m), the point at which half the protein is native form, half is unfolded. The higher the T_m, the more stable the protein should be at the storage temperature. A high T_m also indicates that the protein is less susceptible to unfolding and denaturing at low temperatures.

Although DSC is not a replacement for stability studies, it does serve as a screening tool that enables formulation scientists to focus on three or four of the most promising formulations instead of having to test 30 or 40 different combinations to arrive at a final formulation. MicroCal's VP Capillary DSC platform "is very sensitive, especially for formulations screening," said Dr. Verna Frasca, technical services scientist at MicroCal. "A difference in transition midpoint of 0.5 or 1 °C is significant."

Considerably less than one milligram of protein is all that is needed to obtain reasonable data per scan using MicroCal's instrument. Various buffers are then tested along with the protein to see which one gives the highest T_m. Additives are also tested to see which excipients can increase the T_m, followed by preservatives, which are often the cause of protein aggregation. DSC is used for all the T_m screening.

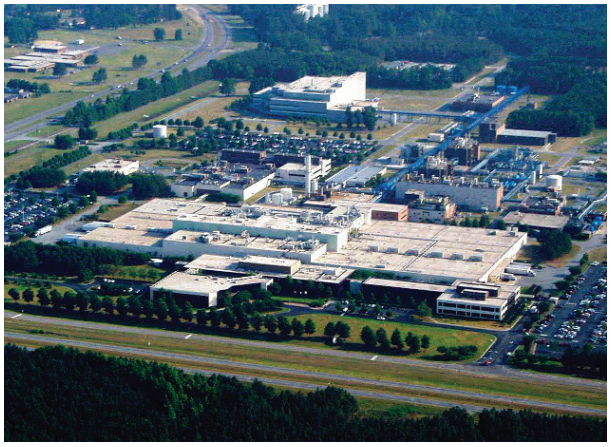
MicroCal's DSC platform uses a capillary-cell design to minimize the thermal fluctuations resulting from aggregation. Although multiple-cell instruments can increase throughput, Frasca notes they also have limitations for sensitivity. "With the current version of electronics, we didn't feel [multiple cells] would be the best solution." Instead, MicroCal chose a single-channel, single-cell instrument that can be used 24 hours a day, seven days a week.

"One of the factors with DSC is that it provides fairly low throughput relative to other techniques," says Frasca. With the old instruments, "each scan takes an hour and a half to two hours, so in an eight-hour period you can complete four to six scans from a single instrument."

The capillary-cell design reduces the sample volume requirement approximately threefold. "This means the scan rate is about three times faster compared with other DSCs," says Frasca. Depending on the scan rate, the MicroCal DSC system can perform as many as 50 scans in a 24-hour period.

The VP Capillary DSC platform is integrated with an autosampling system that includes a moving injection needle for sample loading directly to the cell by means of a distributor valve with very low dead volume and sample dilution. Identical sample and reference cells are constructed from 1.5-mm metal capillary tubing, and the functional volume of each capillary cell is 125 µL. The capillary cells have a high cell surface–sample volume ratio for improved thermal equilibration of sample and allow for a scan rate as fast as 250 °C/h. The capillary cells have separate inlet and outlet tubes that "allow sufficient cell cleaning with the autosampler before each sample is loaded," according to Frasca. Sample and reference buffers are stored in 96-well plates in thermostatted drawers.

Cheryl Mikkola



DSM Pharmaceuticals To Double Sterile Manufacturing Capacity

In response to steadily increasing demand, DSM Pharmaceuticals (Greenville, NC) is investing $62 million in the expansion of its sterile manufacturing capacity. By adding five new, state-of-the-art freeze dryers and one new filling line to its Greenville site, the company expects to more than double its current lyophilization capacity by 2004.

Kevin Lesnewski, DSM's senior manager of marketing and business intelligence, sees the market for sterile products growing at a double-digit rate, a figure that doesn't include potential biological products already in the drug pipeline. By adding more capacity, DSM hopes to stay ahead of this demand. Says Lesnewski, "We're anticipating the services our customers will require and building accordingly. We don't want to put any constraints on them."

The five new 300-ft^2 freeze dryers, manufactured by BOC Edwards (Wilmington, MA), will bring DSM's total sterile manufacturing capacity to more than 4,000 ft^2 of shelf area. Two units will be added to an existing filling line with the remaining three supplied as part of a new filling line from Bosch Packaging Technology (Minneapolis, MN). In total, this will give the facility 14 freeze dryers. All the new dryers

incorporate automatic loading and unloading systems. With the added filling line, the facility is expected to be capable of manufacturing more than 24,000 vials/hour or 400 vials/minute of sterile product.

Custom designed for the manufacture of biological products, the new freeze dryers all contain liquid nitrogen cooling systems. According to Joe Wong, DSM project manager, the aggressive cooling cycle supplied by the liquid nitrogen systems "makes them very good for the difficult process of manufacturing biological drugs."

To better monitor and control the new equipment's operation, DSM has designed a custom in-house control program. Wong says,"The program allows us to fine-tune the system. We have better control of the process, better monitoring, and better response."

The first two freeze dryers are expected to be ready for use by the first quarter of 2003, with the remaining three units operating by the end of 2004. DSM's Greenville expansion plans also include a new area for bulk formulation.

Ronelle Russell

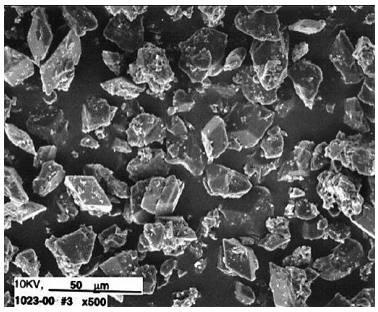
Microsphere *continued from page 17*

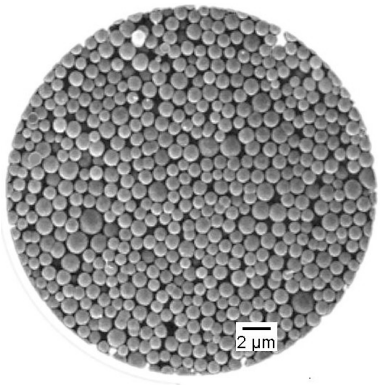
immediate and sustained-release delivery and can be administered by injectable, pulmonary, nasal, topical, and ophthalmic means.

"ProMaxx transforms proteins and other molecules into 0.5- to 3-μm microspheres, which are ideal for delivery to the deep lung. Systemic absorption of drugs occurs from the large and permeable surface area of the deep lung," says Epic chief technology officer Larry Brown.

The ProMaxx process was actually the result of "a fortuitous lab discovery at Epic," recalls Brown. "We were working with water-soluble polymers. You can take proteins out in the presence of water and precipitate them. But we found that if you control pH, ionic strength, and other factors, you can produce uniform microspheres rather than just an amorphous precipitate."

Epic's lead product based on the ProMaxx platform is LeuProMaxx, a sustained-release injectable medication for prostate cancer. The 28-day treatment LeuProMaxx formulation is about to enter Phase III clinical trials, and Epic has developed an 84-day formulation that is now in Phase II trials.





Insulin crystals, before and after microsphere formulation.

In addition, the company has produced microspheres having the ideal size, shape, and aerodynamic properties for the pulmonary delivery of insulin. ProMaxx insulin microspheres have so far been demonstrated compatible with dry-powder inhalers and metered-dose inhalers.

"ProMaxx is a straightforward manufacturing process that requires a modest capital investment compared with other macromolecular delivery systems," says Anthony Garramone, Epic president. "It's a CGMP equipment suite with significant capacity, so if you have the facility, ProMaxx requires little capital to get it up and running." (See photo.)

Also, the water-based process uses no organic solvents. "It requires no specialized facility, no EPA-regulated processes," says Brown. It also can be performed by technicians trained in basic manufacturing skills.

The company has "run the process in different locations with different technicians," according to Garramone.

"A modest-size facility produces commercially relevant dose numbers, partially due to the narrow particle size, which eliminates the need for particle sieving after production. Yields are large. Everything we produce, we package," added Brown. Drug loadings can typically exceed 90% in microspheres—much higher than in traditional formulations. For example, the insulin microspheres are 97% insulin.

Baxter's business strategy is to offer an array of services to pharmaceutical clients. The company pursues partnerships, working with proprietary molecules from pharmaceutical companies, then improving formulation for delivery. Also, internally, the company develops off-patent drug molecules and puts them into its proprietary drug-delivery systems to generate formulations with improved characteristics, in some cases reformulating a drug that's already FDA approved. LeuProMaxx is one example. Although FDA approval is still required for the new formulation, there may be some economies of scale in the reformulation of drugs (e.g., some data from the earlier application filing may be referenced).

George Miller

FDA Provides New Formulation Tool

FDA has given the pharmaceutical industry a new tool to help with the formulation of new drug products. The Inactive Ingredients Database, which was finalized and first made available in late December 2002, contains more than 102,000 listings of inactive ingredients present in FDA-approved drug products. The listings are searchable by ingredient name and can be accessed using a minimum of any three consecutive letters from a name.

Each database listing includes a brief description of an ingredient's dosage form and route of administration, the ingredient's Chemical Abstracts Service number, and the maximum potency for each route and dosage form. According to 21 *CFR* 210.3(b)(8), an inactive ingredient is "any component of a drug product other than the active ingredient." The database does not list contaminants found in approved drug products.

The database arose from the electronically available Inactive Ingredient Guide that has been available since 1996. "The information was old and there's a lot of new inactive ingredients and a lot of [potency] levels approved since 1996," said Greg Davis of FDA's Office of Generic Drugs (OGD) Regulatory Support Branch, "so this was an effort to not only update the information that's available to industry but also make it a little more user friendly."

OGD hired a contractor to evaluate thousands of approved applications to determine the accuracy of the data in the current system and the data that was in the old system, according to Davis. It then developed the database using the *Orange Book* staff. The same group is responsible for quarterly updates to the database, which will be available by the fifth working day of every April, July, October, and January.

The Inactive Ingredients Database can be accessed at http://www.accessdata.fda.gov/scripts/cder/iig/index.cfm. Search results are displayed alphabetically and are sorted first by ingredient. If a particular inactive ingredient cannot be located in the database, questions can be mailed to OGD, Regulatory Support Branch HFD-615, 7500 Standish Place, Rockville, MD 20855 or e-mailed to drugproducts@cder.fda.gov.

Cheryl Mikkola