# NanoBiotech News

*The global nanobiotechnology intelligence source*

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## Advanced Magnetics' Combidex scheduled for ODAC review

*By Steve Lewis*

The U.S. Food and Drug Administration's Oncologic Drugs Advisory Committee (ODAC) is scheduled to review the regulatory filing for Combidex, Advanced Magnetics' (AMEX:AVM) investigational molecular imaging agent, on March 3, 2005.

Advanced Magnetics, based in Cambridge, MA, is jointly preparing for the upcoming ODAC meeting with Princeton, NJ-based Cytogen Corporation (NASDAQ:CYTO), which holds the U.S. marketing rights to Combidex.

Observers are divided over whether this is a good or a bad sign for the ultimate FDA approval of Combidex, concedes Lisa Gordon, director of investor relations for Advanced Magnetics.

"An advisory panel meeting is often part of the regulatory process," she observes. "The significance [of this meeting] is up to interpretation -- some say it's a good sign, some say it isn't."

During the ODAC meeting, clinical data will be

*continued on page 4*

## Drug delivery innovation may soften pharma's pending patent woes

*By Russell A. Jackson*

The development of nanotech-based drug delivery mechanisms will have a significant impact on the pharmaceutical sector's financial future. About $60 billion worth of branded products will lose their U.S. patent protection in the next five years. That has big pharma scrambling for ways to protect its aging giants from those made by smaller, often more nimble competitors.

One of the most effective ways to build a fort around them is to find new, cheaper, less-invasive ways to get them to their targets in patients' bodies.

"The pharma market has evolved considerably over the past decade," says S. Ravi Shankar, a research analyst in Frost & Sullivan's Technical Insights Group. "Because the drug delivery sector is linked directly to the pharma sector, the challenges faced by the pharmaceutical industry give the drug delivery firms an opportunity to develop technolo-

*continued on page 4*

*A more targeted therapy*

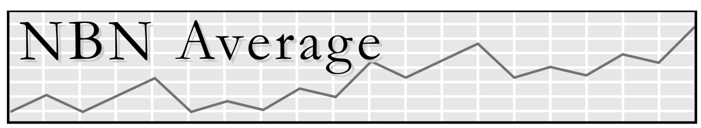
## Nanotechnology enhances gene therapy for prostate cancer

*By Judith Nemes*

A collaborative team of U.S. researchers has successfully completed a preclinical trial study that used nanotechnology to improve delivery of gene therapy for prostate cancer treatment.

The team, composed of chemical engineers at the Massachusetts Institute of Technology (MIT) in Cambridge, MA, and cancer researchers at the Lankenau Institute for Medical Research in Wynnewood, PA, employed nanotechnology to deliver therapeutic DNA to prostate tumor cells in mice to make the tumors shrink, says Janet Sawicki, PhD, a senior investigator at Lankenau, which is part of the Main Line Health System in Wynnewood, PA.

*continued on page 6*




| Company | Symbol | Close 02/01 | Close 02/08 | % Change |
|---|---|---|---|---|
| Acacia Research Corporation | ACTG | $ 5.80 | $ 5.68 | -2.07% |
| Accelr8 Technology | AXK | $ 2.00 | $ 2.85 | 42.50% |
| Advanced Magnetics | AVX.V | $ 16.80 | $ 19.98 | 18.93% |
| Advectus Life Sciences | AVXSF.PK | $ 0.03 | $ 0.03 | 0.00% |
| Affymetrix | AFFX | $ 43.13 | $ 42.28 | -1.97% |
| Agilent Technologies | A | $ 22.52 | $ 23.75 | 5.46% |
| Altair Nanotechnologies | ALTI | $ 2.40 | $ 2.21 | -7.92% |
| American Pharmaceutical Partner | APPX | $ 49.39 | $ 49.11 | -0.57% |
| Biophan Technologies | BIPH.OB | $ 1.18 | $ 1.23 | 4.24% |
| Biosante Pharmaceuticals | BPA | $ 5.69 | $ 5.45 | -4.22% |
| Caliper Life Sciences | CALP | $ 7.60 | $ 8.03 | 5.66% |
| CombiMatrix | CBMX | $ 3.17 | $ 3.12 | -1.58% |
| Flamel Technologies | FLML | $ 17.40 | $ 16.78 | -3.56% |
| Nanobac Pharmaceuticals | NNBP.OB | $ 0.12 | $ 0.13 | 8.33% |
| Nanogen | NGEN | $ 5.07 | $ 5.00 | -1.38% |
| Novavax | NVAX | $ 2.50 | $ 2.41 | -3.60% |
| pSivida | PSDV | $ 8.71 | $ 8.75 | 0.46% |
| SkyePharma | SKYE | $ 8.71 | $ 11.86 | 36.17% |
| Starpharma Holdings Limited | SPHRY.PK | $ 6.80 | $ 6.60 | -2.94% |
| TOTAL | | 209.02 | 215.25 | ▲ 2.98% |

# Five Star nanoemulsions enable nutraceuticals as first commercial platform

*By Marie Powers*

Five Star Technologies, a privately held advanced materials and process technology company based in Cleveland, OH, has introduced the initial in-house commercial platform for its controlled flow cavitation (CFC) technology in a nutraceutical product line developed by Improvita Health Products, Inc., also based in Cleveland.

The lollipop, gel, and jelly bean/chew delivery modalities from Improvita's Well Kids Zone line use nanoformulations that allow slow release of active ingredients that help to alleviate cold and flu symptoms, boost immune systems, and shorten the duration of illness. The CFC process represents "an elegant, precise mechanism to deagglomerate and mill down particle sizes" to the nanoscale, according to Timothy E. Fahey, the company's vice president of business development.

Hydrodynamic cavitation is the formation, growth, and implosive collapse of vapor bubbles in a liquid created by fluctuations in fluid pressure. Long known to engineers as a powerful mechanical force, uncontrolled cavitation can cause the molecule-by-molecule erosion of turbine blades, submarine propellers, pipes, and valves, Fahey explains. Five Star's patented CFC technology allows the company to control the location, size, density, and intensity of the implosion of bubbles to produce particles in high volume with custom-selected characteristics, such as uniform particle size and crystalline structure.

At Five Star's manufacturing facility in Cleveland, which opened in August 2004, millions of cavitation bubbles are created in a flow-through reaction chamber, Fahey explains. When a bubble forms in the low-pressure zone, it pulsates, creating Bjerknes Force, which draws surrounding particles to the bubble surface. An increase in the surrounding pressure induces bubble collapse, usually in the form of a microjet in one part of the bubble. The liquid microjet, which can reach speeds of 3,000 ft/sec to 4,000 ft/sec, is hurled against the opposing bubble wall, creating a shock wave that causes pressure impulses that can reach 150,000 psi. As the microjet continues through the opposing bubble wall, it penetrates the surrounding media, creating an ultra-sheer layer between the jet and the adjacent liquid.

By controlling the bubble dynamics, Five Star creates a variety of advanced nanostructured materials and a wide range of emulsion, dispersion, and reaction enhancement processes. The CFC process technology is broadly applicable, robust, scalable, and a particularly cost-effective mechanism for producing nanoparticles, which can be efficiently packed with customized formulations.

### *Speeding the mode of action*

Improvita, for instance, offers an active formula of chelated vitamin C with zinc designed to boost the immune system and relieve cough and cold symptoms; a specially formulated lollipop, lozenge, and gel that coats the mouth and throat to relieve sore throat pain and alleviate bronchial stuffiness or congestion; and an antacid and anti-nausea formula -- also in lollipop, chew, and gel forms -- designed to relieve nausea and symptoms associated with stomach flu or intestinal distress.

All of the products contain CFC-formulated nanoparticles that speed the mode of action, Fahey says.

"Instead of having to drink cough syrup from a spoon or a cup, the cough and cold gel provides a shot of the active ingredient directly into the back of the throat, where it can go to work immediately," he explains. "The nausea relief products provide the same burst of activity."

The Improvita product line also includes a nanoparticle-formulated nasal spray designed to moisturize and gently open a child's nasal passages, and Five Star is working with Improvita on an adult throat spray that would work by the same mechanism to relieve sore throat symptoms.

"Improvita is excited about the implementation of nanotechnology in the foundation of the Well Kids Zone products," says Tom Klamet, the company's president, noting that nanoparticulate technology allows for better control of dosing, potency, uniformity, and stability of the all-natural formulas of Well Kids Zone products.

*continued on page 3*

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## Five Star Technologies *from Page 2*

"Nanoparticles are a perfect size for targeting bacteria and virus proteins with a high level of precision," he points out.

To generate revenues for the short term, Five Star is "heavily focused" on nutraceuticals, Fahey says. While Improvita is its lead customer, Five Star is "down the road" with other companies in applications such as appetite suppression and nutritional supplements, and additional health and personal care products using the CFC technology are expected to roll out by mid-year. From there, "the CFC technology has good applications in topicals, and we think we can build a very nice core business from this combination," Fahey tells *NanoBiotech News*, predicting the company will break even next year.

But the CFC process lends itself to a wide array of nanoparticle management platforms, from non-contact precision milling to high bioequivalence emulsion systems. In pharmaceutical applications, the company's nano-crystallization formulations could enable novel dosage forms, expand applications for existing active pharmaceutical ingredients, enhance targeting and controlled release, and resurrect shelved drug candidates.

To continue to refine its metal oxide materials applications, Five Star is collaborating on research catalysis and electronics in project-specific, co-funded work. The company is likely to take a similar approach in developing drug delivery applications, Fahey says, noting that in the past Five Star has licensed or sold technology to global players such as Merck & Co., Inc. (NYSE:MRK), Schering-Plough, Inc. (NYSE:SGP), Eli Lilly and Company (NYSE:LLY), and others, which now serve as validation partners.

"We don't have the in-house expertise to build out a full drug delivery program," he concedes. "We're building more IP and developing proof of concept with existing compounds. Eventually, we plan to develop a scientific advisory board that would provide guidance in developing this product stream."

### *VCs like the company*

Five Star has been a hit with top-tier venture capital firms. Relying solely on private investments from its 1995 start-up until attracting a small VC round in 2002, the company raised $4.5 million in an October 2003 financing round led by Morgenthaler Ventures, a $2 billion venture firm based in Menlo Park, CA. Other investors include Industrial Technology Ventures, LP, an affiliate fund of Cordova Ventures in Atlanta, Chevron Technology Ventures in San Francisco, and Early Stage Partners LP, based in Cleveland. Five Star expects to solicit a final round of venture financing this year, Fahey says.

Last week, Bob Pavey, general partner at Morgenthaler Ventures, joined Fahey and company founder and CEO Jim Mazzella in a featured presentation at the International Business Forum's Nanotech Investing Forum in Palm Springs, where they discussed Five Star's ability to move from platform technology to product. In a bit of serendipitous timing, Five Star also appeared in the online edition of *BusinessWeek's* Feb. 14, 2005, cover story on nanotechnology.

"In one sense, Five Star is a nontraditional venture investment because they didn't have a product ready for market, though they do now," Pavey tells *NanoBiotech News*. "But our firm has been looking at a lot of next-generation materials companies -- next-generation materials applied in very interesting technologies. Five Star has developed a platform technology that's capable of changing a lot of traditional industries, from drug delivery to catalytic converters. They have the ability to produce very fine and precise particles in very high quantities."

### *IP portfolio is strong*

Morgenthaler, which typically invests $5 million to $40 million over several rounds in each portfolio company, was attracted to Five Star not only for the versatility of its nanomaterials but on the strength of its intellectual property (IP), which includes 11 issued U.S. patents,[1] four PCTs, and more than a dozen additional U.S. patent applications. In its early years, Five Star operated in "stealth mode," Fahey admits, perfecting the CFC technology invented by Ukranian scientist Oleg V. Kozyuk, PhD -- now the company's chief scientist and director of research and development -- and securing the IP.

"They're a very creative and effective group," according to Morgenthaler's Pavey. "They know how to run a company. They have Midwestern ethics -- no fancy offices, just a focus on developing the technology and getting it into products."

Five Star could someday be a good candidate for a public offering, "but it's way too early to say," Pavey cautions. "We never finance a company if we don't believe a public offering could occur at some point, but with Five Star we're talking years down the road, not months."

In the meantime, Five Star will continue to validate its technology by producing not only nanoparticles but also the nanoemulsions that

*continued on page 7*

## Advanced Magnetics *from Page 1*

presented in a briefing package, and the committee with then give a recommendation to FDA on the Combidex submission. "A positive vote, however, is not a guarantee of approval; nor is a negative vote a guarantee of non-approval. Besides, we still have a PDUFA (Prescription Drug User Fee Act) date of March 30."

Advanced Magnetics has reported a number of clinical results in the past year. Highlights include:

• In a study of 21 patients with pathologically confirmed node-positive disease, scientists identified 66 nodes by a combination of MRI and Combidex. (See *NanoBiotech News*, Oct. 13, 2004, p. 1.)

• A study of 12 patients scheduled for renal biopsy for suspicion of proliferative glomerulopathy or renal graft rejection found that Combidex imaging can help identify those conditions. (See *NanoBiotech News*, Oct. 13, 2004, p. 1.)

• An efficacy study in the November 2004 issue of *Radiology* reported that MRI and Combidex used together improved the sensitivity of detecting metastases to lymph nodes in patients with urinary bladder cancer. (See *NanoBiotech News*, Oct. 27, 2004, p. 1.)

• Most recently, Harvard researchers reported the ability to 'predict' metastases using Combidex and MRI. (See *NanoBiotech News*, Jan. 19, 2005, p. 1.)

Advanced Magnetics received an approvable letter in June 2000 from the FDA for use of Combidex as a magnetic resonance imaging (MRI) agent for diagnosing lymph node disease. The company submitted a response to the agency's request to provide additional details in mid-2004. (See NanoBiotech News, July 7, 2004, p. 1.)

Advanced Magnetics stock closed at $19.98 on Feb. 8, 2005. The stock had traded at slightly over $17 per share in mid-January, and was trading below $10 a share in July 2004.

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## Drug delivery *from Page 1*

gies that can overcome those challenges." One of them is related to advances in genomics and proteomics, which are yielding more macromolecular drugs. "They need advanced delivery systems to successfully demonstrate their therapeutic benefits."

Also, Shankar tells *NanoBiotech News*, "increasing competition from generic products is putting pressure on the pricing of branded formulations." And with all those billions in products about to become fair game for generic formulations, "that threat is going to be even stronger for the pharmaceutical companies. The majors will have to differentiate their off-patent drugs to ward off that competition, and drug delivery technologies are an important tool that can help them do so." Hence, he adds, "there is an increasing trend in launching off-patent drugs in novel delivery platform."

### *Improving patient comfort*

The goals, Shankar points out, often are providing patients with greater comfort and increasing compliance to prescribed medication regimens. "An example of a technology that provides increased patient comfort is the SonoPrep device, launched by Sontra Corp., for delivering anesthesia through the topical route," he says. "It employs ultrasound energy to deliver lidocaine through the skin, and thus obviates the need for pricking patients with needles to deliver the drug parenterally."

There are other such products undergoing clinical trials. Pfizer, for example, in collaboration with the drug delivery company Nektar Therapeutics, is working on making inhalable insulin a reality. And Exubera is currently in phase III studies. It "would be an easier delivery platform that could replace the syringe," he says, "which is still the premier method for delivering insulin."

Of course, the market won't change overnight. "Certain technologies -- such as those based on nanobiotechnology -- are fairly early in the development process," Shankar explains. "Some of the nanotech-based technologies might take around 10 years to be commercialized successfully." But there are some that will change the way drugs are delivered much sooner than that. Ireland's Elan Corp. used its nanocrystal technology in the reformulation of Wyeth's Rapamune, for example, a drug that was already on the market. "Some of the technologies have already been commercialized," he says, "and some are near commercialization and will be available on the market in two to three years' time."

Those changes in the drug delivery sector are highlighted in "Drug Delivery -- Analysis of Cutting-Edge Technologies and Trends," a new report from Frost & Sullivan's healthcare sector.

### *Pharma companies evolve*

One of the developments covered in the report is drug delivery companies diversifying into pharma manufacturing. "There are compa-

*continued on page 5*

**Drug Delivery** *from Page 4*

nies that earlier were delivery companies," Shankar explains, "but that now have also developed into pharmaceutical companies. They've realized that depending solely on the royalties they receive from their delivery products would not let them grow at a reasonable rate. Instead, they realized they'd have to develop a broad technology platform that can be used for a variety of applications."

Elan, for example, started off as a pure drug delivery company but eventually moved into developing and marketing its own compounds. "It has drug delivery technologies -- including its nanocrystal technology -- and also is developing drugs for neurology, autoimmune diseases and severe pain," he says. "For drug delivery companies to make that transition successfully, they need a robust technology platform, development and commercialization resources and management talent."

It's a tough job, but when a company assembles the right team and the right technology, new drug delivery techniques blossom. Here are a few highlights of some of the technologies -- and the companies behind them -- outlined in the report:

• West Henrietta, NY-based Nanolution LLC, a subsidiary of Biophan Technologies Inc., has developed a nanomagnetic particle technology that promises to "provide precise control of the drug delivery process," Shankar reports. "A variety of electromagnetic properties, including magnetic susceptibility, inductance and capacitance, allow the nanomagnetic particles to be tuned very accurately to a specific wavelength. As a result, particles can be bound to a drug and engineered to respond to particular wavelengths. That provides a higher degree of control compared to other, similar technologies that have been developed or tried over the years."

• Haifa, Israel-based NanoPass Technologies Ltd. was founded in 2000. It is developing a novel drug delivery system based on its MicroPyramid -- its word for "microneedle" -- technology platform, which "addresses safe and efficient transdermal drug delivery of large molecules, including vaccines, genes and therapeutic proteins. The arrays are produced using advanced micro-electromechanical system fabrication techniques." That technology, he notes, "helps make the injection process painless because it utilizes only the outermost layer of the skin, where nerve endings do not normally penetrate."

• Livermore, CA-based Eksigent Technologies has developed the EKPump for protein drug delivery. The device is a single-use, disposable, inexpensive protein drug delivery system, Shankar comments. It's a combination of electrokinetic pumping and gas-free electrodes that provides extremely accurate flow rates. "Its ability to accurately deliver low volumes of highly concentrated protein drugs has led to its miniaturization," he adds. "Its size is one-tenth that of currently available pump systems. It can be surgically implanted or attached to the body with an adhesive."

• Sontra Medical Corp. has developed a device that uses ultrasonic waves to open minute cavities on the skin so that drugs and other macromolecules can be administered into the patient's systemic circulation. It's been approved for delivering topical lidocaine, but the technology "opens up a large platform of applications in the transdermal delivery of pain drugs, vaccines and protein biopharmaceuticals."

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**Prostate cancer** *from Page 1*

One remarkable advance achieved in this study was the finding that nearby normal muscle cells were not harmed by the DNA therapy that was targeted to prostate cancer cells, notes Sawicki, one of the team leaders and an expert in gene therapy research for prostate cancer. Other therapies produce toxic side effects to normal cells, but that did not occur in this study, she says.

Researchers also believe the therapy they have developed could be modified to target treatment of other types of tumors, and it could have potential application for treatment of enlarged prostates, or benign prostatic hyperplasia (BPH). Their findings were published in a recent issue of the *Proceedings of the National Academy of Sciences*.[1] Sawicki also presented the team's results in the past year at the American Association for Cancer Research and at the American Society for Gene Therapy. The two other leaders of the study from MIT are Robert Langer, PhD, and Daniel Anderson, PhD.

"There is a real need for effective treatment for metastatic prostate cancer because nearly 32,000 men die each year in the U.S. from prostate cancer," says Sawicki.

MIT's chemical engineers identified a new class of polymers and one of them -- the C32 polymer -- stood out as the one that could most effectively help deliver DNA to cancer cells growing in cultures, explains Sawicki. When the polymer is mixed with the DNA, the polymer condenses the DNA and enables the nanoparticles to be delivered to tumors in mice as a model system for tumors in people, she says. Researchers knew diphtheria toxin kills prostate cancer cells in cultures, but until this study was done, they didn't have a safe, effective way of delivering toxin DNA to tumors in mice.

When the toxin is made in the cell, the protein synthesis in that cell shuts down and the cell dies, explains Sawicki. She genetically engineered the DNA so the toxin would only be made in prostate cells and not in any other cells.

Although researchers cannot get the DNA to differentiate between healthy prostate cells and cancerous ones, that is not a serious threat to patients because the prostate gland isn't essential for the human body to be healthy, says Sawicki. Besides, she anticipates patients who would potentially be treated with this kind of therapy will already have undergone prostate surgery or radiation therapy.

## *For advanced prostate cancer*

"We're hoping our therapy will eventually be used for patients with advanced prostate cancer where the cancer has metastasized throughout the body," says Sawicki. Prostate cancer usually metastasizes to the bone, lymph nodes and lungs.

In the mouse model, an immuno-suppressed mouse was injected under the skin with human prostate cancer cells. After tumors formed, researchers treated those tumors with localized injections of the DNA nanoparticles and found the tumor growth was suppressed or reversed relative to untreated tumors, says Sawicki.

"What makes this polymer delivery system so interesting is that if we inject the nanoparticles into the muscle near the site of the tumor, we don't get the toxin expressed in the muscle," she says. "That suggests we'll be able to target the treatment to tumor cells and keep healthy neighboring cells unaffected by the treatment. That's an improvement over chemotherapy and radiation therapy. We also found there was no systemic toxicity, which means we were able to successfully reduce the size of the tumor without a detrimental affect on the overall health of the mouse."

Looking forward, Sawicki hopes to advance the gene therapy to enable them to systemically treat the patient so the nanoparticles will target and home in on the tumors throughout the body. The current model only enables researchers to provide local treatment.

MIT has applied for several patents covering the technology of the polymers it developed, and together with the Lankenau Institute has applied for patents on using the nanoparticle delivery of DNA to treat hypertrophic tissues, says Anderson. He says it could take at least two years before testing in human clinical trials can begin. Their application for clinical trials with the U.S. FDA will depend on the outcome of an upcoming large animal research study that will treat dogs with the gene therapy to see if their prostate size can be reduced, says Anderson.

That study, which is scheduled to begin this spring and could take about a year, will be conducted in collaboration with Massachusetts General Hospital in Boston, says Sawicki. Dogs were chosen for the study because, aside from the chimpanzee, the dog prostate is the most similar in structure to the human prostate, she explains. Human clinical trials using the DNA therapy are likely to focus first on treating enlarged prostates before trials on patients with prostate cancer can begin, Sawicki adds.

So far, the polymers MIT developed and the DNA delivery system created by Sawicki and her colleagues aren't being produced commercially, says Anderson. MIT researchers have been approached by a couple of interested venture capital firms, but no relationships have been established.

Funding the research hasn't been a problem to

*continued on page 7*

**Prostate cancer** *from Page 6*

date. MIT received grants for polymer research from the National Institutes of Health (NIH), and Sawicki received her end of the funding from a combination of sources including NIH and the John S. Sharpe Foundation of Bryn Mawr (PA) Hospital, a non-profit research organization.

"Our hope is to push this research pretty far without having to start courting investors and selling off pieces," notes Anderson.

*Editor's Note: Contact: Janet Sawicki at (610) 645-3123 and Daniel Anderson at (617) 258-6843.*

### Reference

1. Anderson DG, Peng W, Akinc A, Houssain N, Kohn A, Padera R, Langer R, Sawicki JA. A Polymer Library Approach to Suicide Gene Therapy for Cancer. *Proc Natl Acad Sci* USA Vol 101 No.45, 16028-16033. ⊙

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**Five Star Technologies** *from Page 3*

prove their efficacy.

"The real question with Five Star is whether we will grow as one company with multiple business units or develop into multiple companies," Fahey says. "We have a versatile technology that can be used to synthesize metal oxides and to develop ingestible products, so we have an exciting opportunity to build multiple platforms."

*Editor's Note: Contact Timothy E. Fahey at (440) 239-7005.*

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1. U.S. Patent Nos. 6,802,639: Homogenization device and method of using same; 6,589,501 and 6,365,555: Method of preparing metal containing compounds using hydrodynamic cavitation; 6,502,979: Device and method for creating hydrodynamic cavitation in fluids; 6,035,897, 6,012,492, and 5,937,906: Method and apparatus for conducting sonochemical reactions and processes using hydrodynamic cavitation; 5,971,601: 5,969,207: Method for changing the qualitative and quantitative composition of a mixture of liquid hydrocarbons based on the effects of cavitation; 5,931,771: Method and apparatus for producing ultra-thin emulsions and dispersions; 5,810,052: Method of obtaining a free disperse system in liquid and device for effecting the same. ⊙

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Agent may help to diagnose and treat cancer

# UCLA's peptide coating for qdots enables specific molecule targeting

*By Steve Lewis*

An extremely powerful peptide coating for quantum dots (qdots) that disguises them with a protein-like costume -- allowing them to target specific molecules -- has been developed by a team of researchers at the University of California - Los Angeles (UCLA).

The coating, first described in a paper published by the *Journal of the American Chemical Society*,[1] has led to two new advances detailed in the January 2005 edition of *Science*.[2]

In one project, the researchers were able to track receptors on a cell surface and watch them diffuse and interact. The other included proof of principle in animal models that qdots can be used to image processes at the cellular level.

The *Science* paper also reviews the progress made in the use of qdots, both at UCLA and in the scientific literature. "Qdots as biological probes have lived up to the hopes of their initial promoters," notes one of the authors, Shimon Weiss, DSc, UCLA professor of chemistry, biochemistry and physiology, and a researcher at UCLA's Jonsson Cancer Center and a member of the university's California NanoSystems Institute (CNSI).

The most recent UCLA research is the first result of a new joint effort between the Jonsson Cancer Center and the CNSI.

"We have here at UCLA expertise in both nanotechnology and cancer biology, and we'd like to focus and bring together clinicians, life scientists and nanotechnologists to help in cancer research," says Weiss, noting a similar effort announced last fall by the National Cancer Institute (see *NanoBiotech News*, Sept. 15, 2004, p. 1), which included the establishment of Centers of Excellence.

"There is now competition for five major centers around the country," notes Weiss, "But this partnership here will go beyond NIH-supported research. We are planning on long-term support through the Jonsson Comprehensive Cancer Center fund-raising and others. We believe we have the technology and the biological model systems and all the expertise required; we are very well positioned to do work in this area."

### A 'superior' technology

The new coating technology described in both aforementioned papers "Is, we believe, superior to other coatings out there; we believe that it will be applicable for any nanoparticles that need to go into biological materials," says Weiss. Two more papers will be published later this month, he continues, and

*continued on page 8*

**Qdots** *from Page 7*

one of them compares the coating with others already in the market. Additional new results are currently summarized into manuscripts, he adds.

"Our nanoparticles are much smaller; the hydrodynamic radius after the coating is around 10 nm," says Weiss. "QDC's, for example, are around 30. We compared our coatings to lipid coatings, salinization, and so forth, and we come up to be the smallest in size." Other advantages, says Weiss, include a much higher saturation intensity and monodispersivity (lack of aggregation). "Mono-dispersed qdots are more suitable for single-molecule work in the live cell," Weiss emphasizes. "The bio milieu of a cell is quite crowded, so the smaller, less aggregated particles have an easier time finding their target. In addition, we are disguising those particles to look like proteins, and we can manipulate them to work like proteins."

The technology, Weiss says, is licensed to an unidentified holding company.

### *Tracking and image processing*

Based on the advances in the UCLA laboratories, researchers hope that qdots might be used one day as a one-two punch to both diagnose and treat cancer. The qdot technology will enable researchers to locate a tumor within the body, look at it very precisely at the cellular level to determine what kind of cancer it is and then perhaps arm it with toxic therapies designed to kill the disease.

In the research described in *Science*, Weiss and his colleagues were able to view the cell processes for many minutes using their new technology, shedding light on processes that had been theorized, but not actually seen. Using color encoding, researchers could label many proteins and receptors on a cell surface and monitor in real time how they interact -- and the results of those interactions. For example, different colors can be used to label different cell processes, different cancers or different stages of the same cancer.

In another study, conducted at the UCLA Crump Institute for Molecular Imaging, the qdots were labeled with a positron-emitting isotope and injected into mice. Using PET scanning, researchers were able to watch over time as the qdots made their way through the vascular system and to the liver.

"This was a demonstration of multi-functions," notes Weiss. "It can be radioactive, MRI, or near-infrared imaging; you can perform a real-time optical biopsy. Maybe one day we can also deposit energy specifically into the tumor by excitation of qdots' core electrons. This might afford zooming in to the nano scale for a real-time biopsy for diagnosis and, after that, therapy -- all implemented in a single nanoparticle."

Cautioning that this is at present "Very speculative vision (toxicity, for example, is a major challenge)," the potential of the technology is virtually limitless, says Weiss. "You could do drug discovery, diagnostics, all kinds of bio applications, research -- anywhere in vitro and anywhere in vivo," he says. "It is very, very powerful; the basic notion is that you paint anything you want to detect -- and then you can detect it with single molecule sensitivity."

*Editor's Note: Contact Shimon Weiss at (310) 794-0093.*

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# Japanese researchers' breakthrough could lead to clearer MRI scans

*By Judith Nemes*

Researchers at the Kyoto University Institute for Chemical Research in Japan recently reported their success in trapping a single hydrogen molecule inside a carbon fullerene shaped like a soccer ball that measured only 0.7 nm in diameter.

The study's findings could lead to clearer images in MRI scans within a few years after further chemical modifications are made, says Koichi Komatsu, a professor at Kyoto University and lead researcher on the study. The report was published in *Science*.[1]

Using chemical and thermal reactions, Komatsu and his researchers were able to make a hole in the carbon fullerene, trap a hydrogen molecule inside, and close the opening. That's a dramatic improvement over unstable conventional methods that depend on physical techniques such as high-pressure and high-temperature treatment with gases, he says.

Once the technology is refined, it could lead to the controlled production of endohedral fullerenes, which are nano-sized closed-cage carbon molecules with atoms or a molecule trapped inside. Those types of small structures can enhance MRIs and nuclear magnetic resonance analysis, says Komatsu.

In the report, researchers said a fullerene that contained a hydrogen molecule was as stable as the one without the molecule. They noted the molecule did not escape even when it was heated to 500 degrees centigrade for 10 minutes. The team used fullerenes composed of 60 carbons for the experiment, but noted they can apply the same methodology for fullerenes of 70 carbons and they can insert other types of atoms and molecules.

### *Synthesizing endohedral fullerenes*

"We consider our work to be significant as the first report to show that the organic synthetic method can be successfully applied to the synthesis for the production of an endohedral fullerene, for which only hard-to-control physical methods have been employed so far," says Komatsu.

In the future, when the same method can be applied to encapsulate lanthanoid metal elements, such as gadolinium, it will be useful for improving images in MRI scans, he notes. However, before such advancements can occur, Komatsu and his colleagues are attempting to apply the same method to encapsulate other small atoms and molecules. If they can succeed in that endeavor, they will then turn their attention to working with encapsulation of metals.

The Institute for Chemical Research has applied for a patent of the process developed by Komatsu and his team of researchers. Komatsu estimates it could take at least two to three years before any commercialized product results from the work currently underway in their laboratory. He hasn't signed any agreements yet to work with any companies on developing a marketable product.

*Editor's Note: Contact Koichi Komatsu at 81-0-774-38-3172.*

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# Fullerene-based contrast agents could allow first single-cell imaging

*By Marie Powers*

At the age of 24, Rice University doctoral student Balaji Sitharaman underwent a thyroidectomy as a precautionary measure, since current diagnostic methods could not confirm whether a growth discovered on his thyroid gland was benign or malignant. His personal brush with the limitations of diagnostic imaging led to his PhD dissertation topic on the use of carbon nanostructures as advance contrast agents (CA) for MRI.

In December, the Nanotechnology Foundation of Texas selected Sitharaman as one of two winners of the 2004 George Kozmetsky Award for Outstanding Graduate Research in Nanotechnology for his efforts to create a new class of contrast agents that could potentially enable MRI of individual cells.

More than 25 million patients in the U.S. undergo MRIs annually, and doctors use contrast agents in almost one-fourth of those procedures to increase the sensitivity of the scans. The most effective and commonly used contrast agent is the toxic metal gadolinium.

In an article published in the *Journal of the American Chemical Society*,[1] Sitharaman and colleagues explain that the lanthanide ion Gd3+ usually is chosen as an MRI contrast agent. To reduce toxicity, the Gd3+ aqua ion is chelated to a ligand.

"These agents are intravenously administered to patients, and by reducing the relaxation time of water protons in the affected tissues, they help produce a higher contrast diagnostic image," the researchers write.

"Although MRI usually provides good spatial resolution, sensitivity is often a problematic issue,

*continued on page 10*

**Drug delivery** *from Page 9*

especially in novel applications such as the targeting of specific cells or tissues," they add. "Since the number of Gd3+ complexes that can be delivered or attached to a specific cell is largely limited by biological constraints, the efficacy of each Gd3+ center must be improved in order to induce sufficient signal intensity to image individual cells. For this reason, the search for more efficient, higher-relaxivity MRI agents represents a key objective in current MRI CA development."

### Reduced toxicity

Sitharaman has addressed the challenge of creating new forms of contrast agents by encasing gadolinium inside fullerenes -- single molecules of carbon atoms arranged in spherical or tube-shaped structures. By enclosing the gadolinium inside the carbon molecules, he has simultaneously reduced the toxicity of the metal to near zero while boosting its effectiveness as a contrast agent.

One of Sitharaman's creations is a buckyball encasing a single atom of gadolinium. In addition, he has discovered a method of encasing as many as 100 atoms of the metal inside a short length of carbon nanotube. The resulting "gadonanotubes" are 100 times more effective as contrast agents than the best forms in clinical use.

The researcher understands well the shortcomings of MRI contrast imaging.

"Most contrast agents that are used in the clinic don't stay in the body long enough to do these detailed studies," he points out. "We're looking for contrast agents with higher retention and the ability to get into cells, and metallofullerenes work quite well for these purposes."

In a related paper published in *Nano Letters*,[2] Sitharaman and colleagues reported that two gadolinium-based metallofullerene compounds aggregated at a pH of 9 to form spherical and irregular clusters measuring 30 nm to 90 nm, with little concentration or temperature dependency. Below a pH of 9, the aggregate sizes increased steadily and dramatically, reaching hydrodynamic diameters of 600 nm to 1,000 nm by a pH of 5.

"We conclude that the tendency of these metallofullerene species to self-assemble into nanoscale aggregates in aqueous solution likely produces their unusually large, outer-sphere, pH-sensitive proton relaxivities," the researchers write. The results suggest that gadofullerenes could serve as pH-sensitive MRI contrast agents for diagnosing abnormal tissue such as tumors and arterial plaques, which are known to possess lower pH values than healthy tissue, they add.

In the JACS paper, the researchers for the first time report relaxivities as a function of magnetic field for water-soluble gadofullerenes, noting, "the pH dependency of the proton relaxivities makes these gadofullerene derivatives prime candidates for pH-responsive MRI contrast agent applications."

### Tissue-specific imaging agents

In future work, Sitharaman plans to use existing methods of attaching antibodies and peptides to fullerenes to try to create a contrast agent that will bind only with diseased cells such as cancer cells. He is hopeful these tissue-specific imaging agents might allow for the first intracellular, individual cell MRIs.

"We have reached a stage where we understand metallofullerenes quite well," Sitharaman tells *NanoBiotech News*. "Now, we will optimize the parameters -- take our experience and apply it to other types of contrast agents in the lab."

To date, Sitharaman's research has attracted funding from the National Institutes of Health, which awarded a Small Business Innovation Research (SBIR) grant to a joint collaboration between Rice and TDA Research Inc. in Houston, and by the Swiss National Science Foundation and the Office of Education and Science (OFES). Sitharaman hopes to form additional collaborations and to attract venture capital that might accelerate the MRI contrast agent research.

Commercialization is a long-range prospect, however.

"I expect the metallofullerenes to move into the clinic in five to 10 years," he says.

In the meantime, Sitharaman expects to publish additional papers this year and will present his findings at the Materials Research Society (MRS) spring meeting in San Francisco. The Kozmetsky Award also enables him to present his research at the Nano Summit Research Conference in Houston in July.

The Nanotechnology Foundation of Texas is an initiative funded by private individuals, corporations, and other foundations to accelerate research in nanotechnology. The Kozmetsky Award is the first of its kind offered to U.S. graduate students working on nanotechnology and carries a $5,000 prize.

*Editor's Note: Contact Balaji Sitharaman at (713) 348-3476.*

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