newandnoteworthy

Glucans: Augmenting antibodies

In recent years, clinicians have begun to prescribe monoclonal antibodies (mAbs) such as Herceptin, Rituxan, and Campath-1H to treat various cancers. Recent findings by scientists at the University of Louisville School of Medicine, Victoria University of Technology, and Memorial Sloan-Kettering Cancer Center suggest that a natural carbohydrate might dramatically enhance the efficacy of these drugs (*J. Immunol.* **2004,** *173,* 797–806). In particular, they examined the immunostimulatory activity of β-1,3-glucans from yeast and barley.

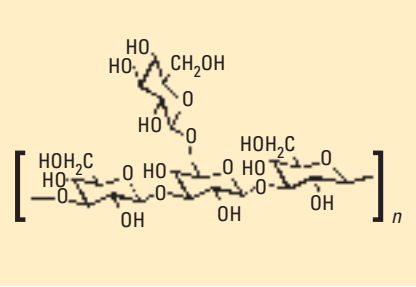
The researchers fed β-1,3-glucan to lymphoma-bearing mice three days before or on the same day they initiated mAb treatment and found that earlier treatment resulted in faster tumor regression. By comparison, treatment with mAb alone elicited no regression. In addition, the effect of β-1,3-glucan appeared to be mediated through complement receptors (CRs), as CR-deficient mice did not respond to glucan treatment.

The researchers subsequently tried to determine the oral uptake mechanism of β-1,3-glucan by fluorescently labeling the carbohydrate. They noted that within three days, lymph node and spleen macrophages contained labeled β-1,3-glucan, and by day four, these molecules had moved to the bone marrow. Over time, the splenic microphages partially degraded the β-1,3-glucans into smaller fragments that concentrated near the cell membranes. The researchers observed that upon moving to bone marrow, the β-1,3-glucan fragments are released and taken up by granulocytes via CRs. It is these cells that then facilitate the tumor cell destruction in combination with complement CRs and mAbs.

"Our research over the past decade has firmly established the efficacy of β-glucan as an immune system enhancer and, more recently, as a highly promising complementary cancer immunotherapy," says Gordon Ross, director of the tumor immunobiology program at the James Brown Cancer Center at the University of Louisville and lead author of the study. As Ross explains, the next step will be to examine the efficacy of β-1,3-glucan with mAbs and cancer vaccines in humans.

The study was partially supported by Biopolymer Engineering, a Minnesota-based biotechnology company that develops natural carbohydrates for drug applications.

—RANDALL C. WILLIS

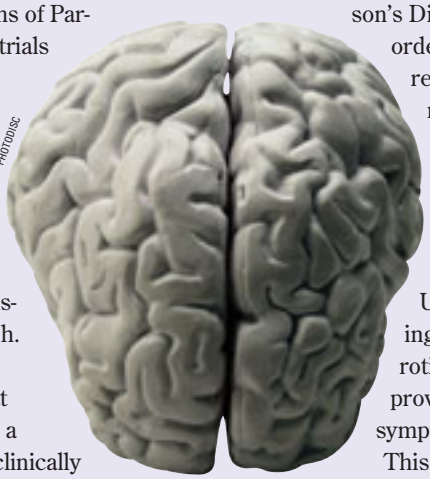


mAB maximizer. The natural carbohydrate β-1,3-glucan might make monoclonal antibody therapies more effective against various cancers.

"Patch" work brain therapy

Schwarz Pharma has reported positive results in separate Phase III and Phase IIb trials with the dopamine receptor agonist rotigotine. The Phase III trial data showed rotigotine significantly reduced the symptoms of Parkinson's disease. Phase IIb trials showed similarly effective results in treating restless legs syndrome (RLS).

In both cases, rotigotine was administered continuously via a silicon-based patch applied once a day that delivered the drug transdermally to the body for 24 h.



Phase IIb study data presented in July shows that a rotigotine patch produced a statistically significant and clinically relevant reduction in RLS symptoms. A neurological disorder that effects up to 9% of the population, RLS is characterized by crawling sensations in the legs and spontaneous leg movement. The chronic condition can be uncomfortable and prevent restful sleep.

"We are encouraged by the magnitude of the reduction of RLS symptoms and the good tolerance that patients experienced during the trial," says Iris Loew-Friedrich, an executive board member of Schwarz Pharma. "A meaningful clinical effect was observed within the first seven days of treatment."

The Phase III clinical development program for rotigotine treatment of RLS is scheduled to start in spring 2005.

At the 8th International Congress of Parkinson's Disease and Movement Disorders in June, pivotal Phase III results were reported for the rotigotine patch in patients with early-stage Parkinson's disease. The study, says Ray Watts, professor and chairmain of the department of neurology at the University of Alabama, Birmingham, "demonstrated that rotigotine can successfully improve [Parkinson's] disease symptoms."

This is third consecutive successful trial for the "Parkinson patch," according to Schwarz Pharma. The company hopes to submit applications for market approval before the end of 2004.

In both the Parkinson's and RLS studies, the therapy produced mild adverse effects, including application-site reaction, drowsiness, nausea, and headache.

The rights to rotigotine were acquired by Schwarz Pharma from Aderis Pharmaceuticals in the late 1990s.

—ALLISON L. BYRUM


CHEMICAL & ENGINEERING NEWS

BIO's Feldbaum opposes required stock-option expensing.

Biotech: No to expensing stock options

Legislation passed in the U.S. House of Representatives in July to limit stock-options expensing on a company's balance sheet was strongly supported by the biotechnology industry. The bill directly challenges recommendations made by the Financial Accounting Standards Board (FASB), the private group typically responsible for establishing financial reporting rules, to categorically include options as business expenses.

"Small and midsize biotechnology companies often do not have products yet available on the market and frequently use stock-option plans as a key method for recruiting and retaining highly skilled employees," says Carl Feldbaum, president of the Biotechnology Industry Organization. "The mandatory stock-option expensing proposed by FASB fails to account for the highly volatile nature of biotech stocks and would do little to provide investors with a clear picture of a company's financial health."

Current rules only require companies to report employee stock options, which give holders the right to buy shares in the future at a current price, in the footnotes of financial statements. According to critics of the current rules, such as billionaire investor Warren Buffet and Federal Reserve Board Chairman Alan Greenspan, the FASB proposals, drafted in March, would oblige companies to provide a more accurate picture of profitability.

Hundreds of companies voluntarily expense options, but high-tech industries like biotechnology, which have a large component of start-ups, tend to rely heavily on options as employee incentives and oppose having to report them as expenses.

"Unlike other mature industries, where stock prices are earnings-driven, the biotech industry includes a large number of small, entrepreneurial companies with extremely volatile stock prices hinging on the regulatory product approval process," Feldbaum says.

Feldbaum calls the House's Stock Option Accounting Reform Act, which passed on a bipartisan 312–111 vote, "more reasonable." It would require expensing only the options given to the top five executives of a company. It also calls for the valuation of those options to be fixed instead of being based on long-term estimations of share price fluctuations as the FASB advises.

The biotech industry clearly made substantial efforts to build support for this bill through both direct lobbying and public activism. Genentech, for instance, handed out leaflets at several keynote sessions of the BIO 2004 conference held in June in San Francisco, asking attendees to call their congressional representative on the issue. The biotech powerhouse also was among several high-tech firms that organized a rally of employee stock option holders in Palo Alto later in the month.

The bill, however, is expected to face significant opposition in the Senate, where Republicans and Democrats have introduced a resolution calling on their colleagues not to interfere with the FASB recommendations.

—DAVID FILMORE

"Approvable" no more?

The FDA is proposing changes to the drug review process, including replacing "approvable" and "not approvable" letters and amending provisions on extending the review cycle. The FDA hopes the changes will ensure a single, consistent method of advising drug manufacturers that review of an application is complete.

For new and generic drugs, the current process of issuing approvable and not approvable letters will be replaced with "complete response letters," providing companies with specific information about what needs to be done before their drugs can be approved for marketing. The system has been used for biological drugs, but the proposal formalizes the process for all drugs.

"This new approach will provide a clearer and more consistent method for communicating to new and generic drug applicants about the status of their applications," says Lester M. Crawford, acting FDA commissioner.

The drug industry generally welcomes the proposal, but news reports quote critics who say that the common response letter category will limit the public's insight into the process. An approvable or not approvable letter sent to a company commonly serves as predictor, particularly to investors, of the delay to market for a drug.

For new drugs that are not initially approved, the "complete response" letter will classify the resubmission as either "Class 1" or "Class 2," depending on what needs to be done to obtain marketing approval.

Class 1 resubmissions will start a new two-month review cycle and will require only certain items, such as draft or final printed labeling, safety or stability updates, or other minor clarifying information. Class 2 resubmissions will start a new six-month review cycle and will be reserved for applications requiring additional information beyond the Class 1 requirements, as might need to be presented to a public advisory committee.

Resubmissions of generic drugs will retain the current "major" and "minor" resubmission terminology. A "major" generic drug marketing application resubmission would begin a new six-month review cycle. A "minor" resubmission would begin a shorter cycle, depending on the issues involved.

The proposal aligns with the performance goals outlined in the Prescription Drug User Fee Amendments of 2002 and can be viewed at *www.fda.gov/OHRMS/DOCKETS/98fr/04-16476.htm*. The FDA is accepting comments on the proposal until October 18.

—ALLISON L. BYRUM

Nudging neurons

Recent research at Carnegie Mellon University, the University of Pittsburgh, and Mount Holyoke College may have put scientists one step closer to creating a functional map of the central nervous system (CNS). Using transgenic and fluorescent technologies, Alison Barth, assistant professor of biological science at Carnegie Mellon University, and her team developed mice that express a c-fos-GFP fusion protein in excited neurons (*J. Neurosci.* **2004,** *24,* 6466–5475).

The transcription factor *c-fos* has long been used as a marker of neural activity because its expression is induced under a wide variety of conditions, including dehydration and pain. Earlier studies of *c-fos* expression were hampered by the need to study expression after tissue fixation, whereas the fusion protein offers researchers the ability to see expression in vivo, in direct response to stimulation.

Barth's team examined the fusion protein's expression under three sets of stimuli—behavioral, physiological, and pharmacological. When they triggered dehydration in the mice, the researchers noted strong fluorescence induction in the same parts of the brain known to express *c-fos* under identical conditions. Similarly, when they administered the antipsychotic drug clozapine, they detected fluorescence in regions of the CNS characteristic of *c-fos* expression.

They also triggered expression in the cells responsible for sensory input of a single whisker. This result showed they could actually identify stimulatory responses in individual cells. Furthermore, Barth's group noted that the electrical properties of neurons in stimulated areas were different from those in areas deprived of sensation.

"These changes are hypothesized to be part of a dynamic interplay between forces that maintain neural firing within an optimal range and those that strengthen particular connections between cells—thought to underlie learning," Barth says.

Although this experiment was largely proof of principle, the researchers are excited about its potential applications. "Our transgenic mouse is a novel tool that can be used to visualize, in living brain tissue, a single neuron that has been activated in response to an animal's experience," Barth explains. She adds that the fluorescent mouse should allow scientists to identify specific neurons involved in different neuropathologies or to rationally develop new drugs for these diseases.

—RANDALL C. WILLIS

PHOTODISC

Launching a safer bet

The formation of a new company called Accentia BioPharmaceuticals was announced in July. Also announced at that time were the company's "solid existing revenue stream" and a pipeline including more than 15 products. Is there a catch?

Actually, yes. Accentia isn't really a completely new company. It is an amalgam of acquisitions made by St. Louis-based venture capitalist firm Hopkins Capital Group over the past two years. But its head-start entrance into the biopharmaceutical fray reflects the model the company will continue to follow: purchasing late-stage investigational drugs with high profit potential and leaving discovery and early development to others.

HOPKINS CAPITAL GROUP

O'Donnell stresses Accentia's hybrid business model.

"The management of Accentia recognizes that drug discovery can be a tedious, inefficient process that often results in wasted dollars spent on research that doesn't pan out," says Steven Arikian, president of product development and market services. The model of purchasing compounds with established safety profiles and emerging efficacy profiles, he says, is "a much safer bet."

According to Hopkins Capital Managing Partner and Accentia Chairman Frank O'Donnell, the company sees itself as a combination of the best of the biotech business model with its focus on highly innovative potential blockbusters and the specialty pharmaceutical strategy of acquiring outside products. At the same time, he says, it is avoiding the typical downsides of each of these approaches, including starting out with a single candidate or seeking only niche products with limited revenue.

The pipeline is headlined by Biovaxid, a "personalized" non-Hodgkin's lymphoma vaccine currently in Phase III trials that originated from a cooperative research and development agreement between the National Cancer Institute and BioVest International, a biologics company in which Hopkins Capital became a majority partner in mid-2003.

Accentia's products on the market are Histex and Respi-TANN from TEAMM Pharmaceuticals, acquired by Hopkins in early 2003. It also has several copromotional deals for other companies' drugs and markets cell culture services obtained from BioVest.

The final piece of the company comes from Hopkins's acquisition of Analytica International, which offers research, commercialization, and communications services.

Accentia is billing itself as "poised to change the paradigm of business development in the health care industry." Whether or not this occurs, its model does point to a possible alternative for diminishing the uncertainty inherent in starting from scratch.

—DAVID FILMORE

Microfluidics and mutations

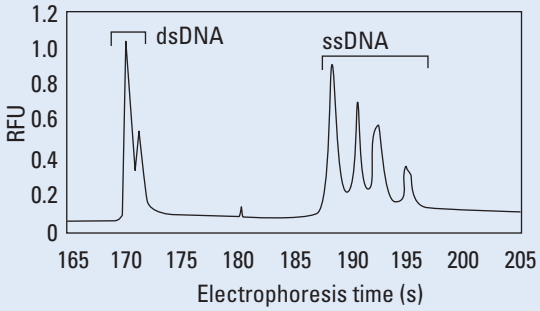
With the sequencing of the human genome, genetic characterization has become the focus in detecting and treating human disease. However, as scientists try to process more samples, assay cost and sensitivity become increasingly important factors. To improve process efficiency, University of Alberta and University of Calgary researchers have developed a microfluidic system that combines on-chip sample preparation with mutation analysis (*Electrophoresis* **2004,** *25,* 2346–2356).

Gene sequencing is perhaps the most sensitive method for mutation detection and identification, but it is also expensive, whereas array-based methods are typically less expensive but have significant false positive rates. University of Alberta engineering professor Chris Backhouse and his colleagues determined that if they combined double-stranded (ds) DNA- (heteroduplex analysis, HA) and single-stranded (ss)DNA-mutation detection methods (single-stranded conformation polymorphism, SSCP) on a microfluidic chip, they could greatly enhance sample throughput while minimizing sample size and costs.

Using cross-shaped microchips, the researchers loaded PCR samples into the left well and positively charged dye into the right well such that, upon electrophoresis, the dye would mix with and label the DNA. When they switched the current longitudinally, the labeled DNA molecules moved into the separation channel and were detected using laser-induced fluorescence.

Likewise, because each run takes about 4 min and uses only 250 pL of sample, researchers could perform multiple runs on a single sample to ensure separation reproducibility.

The researchers also added formamide to the DNA samples to facilitate denaturation. As the molecules entered the separation channel, the neutral formamide remained behind, allowing the DNA molecules to self-anneal as looped ssDNA structures (detected via SSCP) or form homo- and heteroduplexed dsDNA molecules (detected via HA).

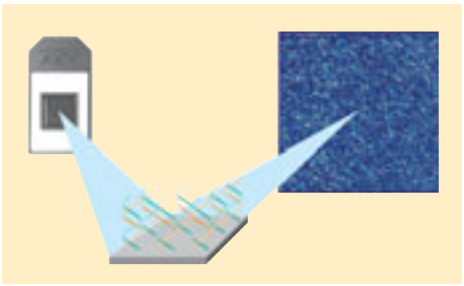


Separation anxiety. Using a microfluidic system, researchers can perform simultaneous heteroduplex analysis (dsDNA) and single-stranded conformation polymorphism studies (ssDNA) to identify DNA mutations. (Adapted with permission from Vahedi, G.; et al. *Electrophoresis* **2004,** *25,* 2346–2356.)

Even though the differences between DNA molecules can be very subtle—for single nucleotide polymorphisms, potentially tens of daltons—Backhouse's group is confident that a combination of HA and SSCP will allow researchers to not only detect mutations but also identify specific mutations on the basis of migration patterns. Furthermore, although they used PCR-amplified DNA for these experiments, the researchers are confident that their method will work with DNA from any source.

—RANDALL C. WILLIS

AFFYMETRIX



Diabetes collaboration

Affymetrix, ParAllele BioScience, and Cambridge University will collaborate on a large-scale Type 1 diabetes study. Headed by Professor John Todd of the university's Juvenile Diabetes Research Foundation/Wellcome Trust Diabetes and Inflammation Laboratory, the study uses two new products from a recently formed partnership between Affymetrix and ParAllele.

Affymetrix's GeneChip Tag Arrays and ParAllele's MegAllele genotyping reagents are being used together to compare the genotypes of 1000 control samples and 1000 diabetic samples. The research will be the first stage in analyzing over 20,000 DNA samples.

"This research project is the most exciting and important genetics experiment I've ever been involved in," Todd says. "Using this new solution for a genomewide gene association study provides us with the best opportunity we have ever had to discover new disease-associated genes and polymorphisms."

The new products from Affymetrix and ParAllele's partnership let researchers perform large-scale genotyping in their own labs with their own panels of single nucleotide polymorphisms (SNPs). The Cambridge study will be the first to use a ParAllele-developed standard panel of 10,000 nonsynonymous SNPs. With the combined MegAllele and GeneChip products, scientists can select SNPs from custom panels of 3000 to 5000 SNPs and standard panels of up to 10,000 preselected SNPs.

The MegAllele products include reagent kits and software specifically designed for use with the Affymetrix GeneChip Tag Arrays and instrument systems.

"We're delighted to offer a full range of tools to analyze the genome in different ways," says Greg Yap, senior marketing director of DNA analysis at Affymetrix. "Our partnership with ParAllele enables us to provide researchers with another solution for genetic association studies and complements our existing solutions for genomewide genotyping and resequencing."

Affymetrix and ParAllele BioScience announced a nonexclusive distribution and marketing agreement in May to combine the MegAllele and GeneChip technologies. The Cambridge University study is the formal launch of the pairing.

—ALLISON L. BYRUM ■