INFORM February 2014 Volume 25 (2) 65–128

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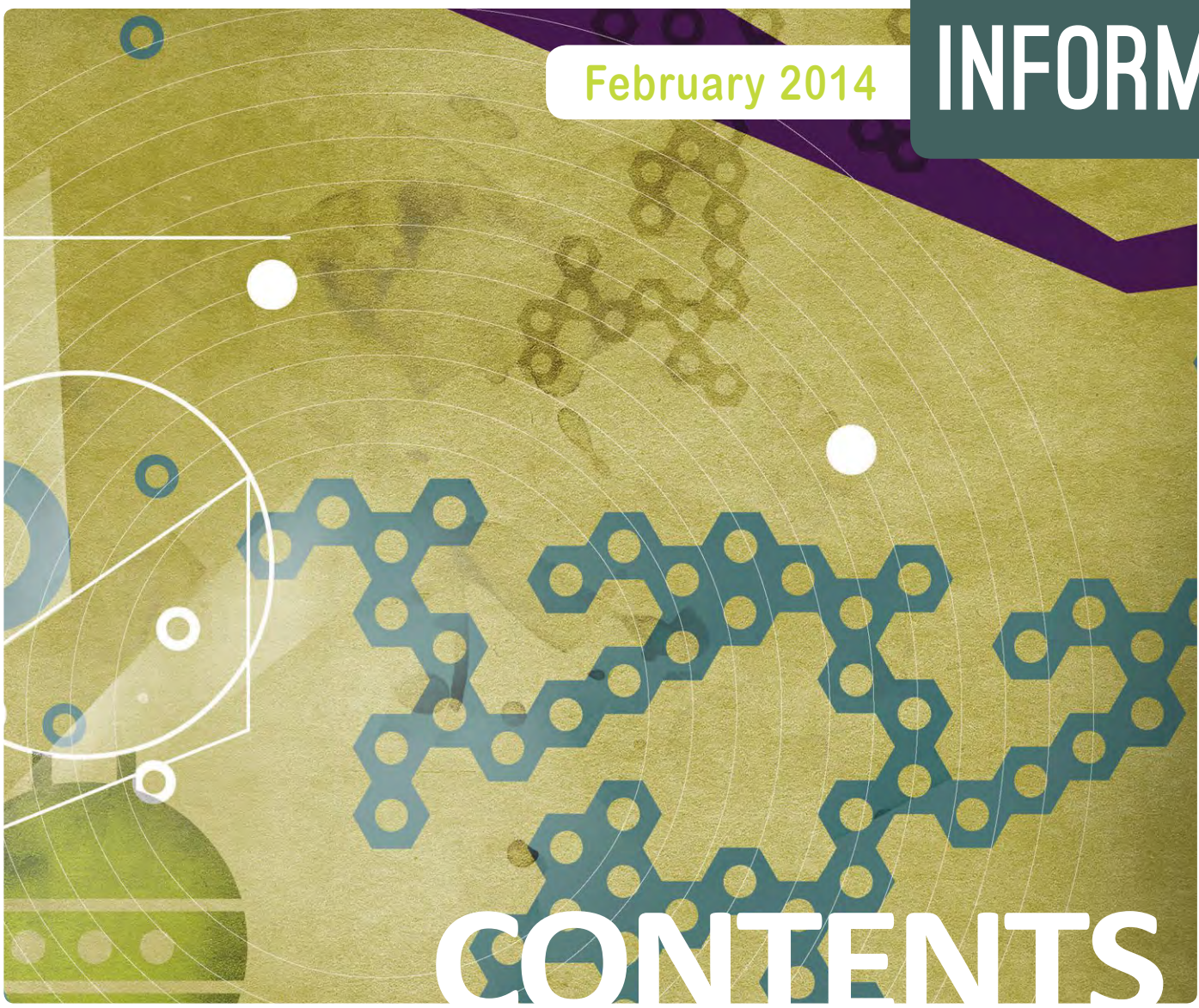

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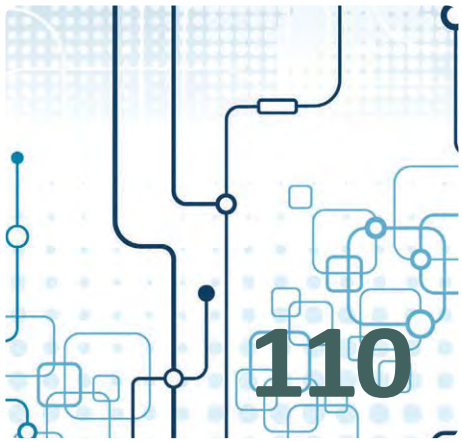

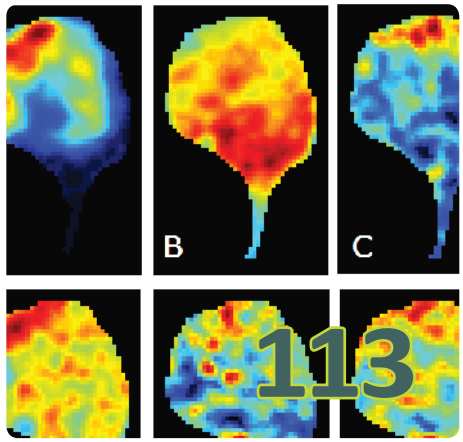

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- A representative list of 17 other companies involved in commercializing pathways to novel biobased chemicals
- A figure showing the complicated nature of fluxolipidomics
- A complete reference list for biodegradable lamellar systems in skin care
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WANT MORE?

A list of alternative pathways and companies not profiled in this article is available in this issue's supplement (digital and mobile editions only).

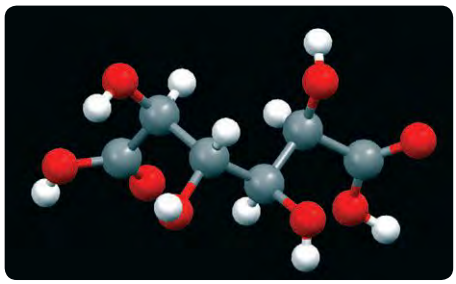
PATHWAYS TO NOVEL CHEMICALS

Many companies in the green chemicals space have responded to the unpredictable price, and sometimes availability, of petroleum by making drop-in replacements (drop-ins). Drop-ins are biobased versions of existing petrochemicals that are chemically equivalent and offer all the functionality and benefits of those products. This equivalence allows them to be substituted with minimal switching costs and without disruptions to delivery. Biobased paraxylene, ethylene, polyethylene, and butanediol are just a few of the drop-ins being pursued by startups and large chemical companies alike. (See example on page 81).

But, as the industry matures, a handful of companies are establishing alternative pathways to biobased chemicals having structures that are entirely new—or that have not yet been applied to commercial markets. Such molecules have enhanced or additional functionality that allows customers to lower their costs, differentiate their products, and/or create entirely new products of their own. Here, we have highlighted six pathways to novel chemicals and one path to high-value drop-ins.

GLUCARIC ACID AND DERIVATIVES

***Company*:** Rivertop Renewables
***Feedstock*:** glucose
***Synthetic process*:** catalytic oxidation
***Potential applications*:** detergent builders, chelating agents, corrosion inhibitors
***Key targeted market segment(s)*:** consumer and industrial and institutional (I&I) detergents; multiple industrial markets
***Contact*:** John Monks, vice president of business development, john.monks@rivertop.com



D-glucaric acid

Key aspects of products/technology:

Products are derived from renewable feedstocks and produced at a low cost enabled by patented innovations on a conventional chemical process. Low cost is achieved through full feedstock conversion, recovery and reuse of reagent, low energy consumption, and use of proven processes. Catalytic oxidation platform can be extended to a range of sugars and organic alcohols (C_3, C_4, C_5, etc.) to produce organic acids, in most cases using the same equipment.

How they compare with traditional (petrochemical or oleochemical) products and surfactants:

In targeted markets, Rivertop's biobased products offer outstanding cost/performance benefits compared with other builders, chelants, and corrosion inhibitors in current use. The multifunctional attributes of the company's novel chemicals include: (i) addressing the ongoing need for safe, sustainable ingredients and (ii) giving customers a new—and sustainable—set of chemical "tools" to use in building better products that will meet the demands of an increasingly health- and safety-conscious customer base.

What differentiates them from other renewable products and technologies?

Rivertop is one of the few biobased companies with a focus on innovation in chemistry vs. biology. The company operates from a versatile and readily scalable oxidation technology platform. Proprietary innovations on traditional oxidation chemistry enable the industrial-scale introduction of novel sugar acids and sugar alcohols, beginning with glucaric acid [HOOC-$(CHOH)_4$-COOH]. Production of large volumes can be scaled up quickly and predictably with moderate capital requirements.

Awards/recognitions:

Cleantech Open finalist. The company was named one of "The 30 Hottest Companies in Biobased Chemicals and Materials" for 2013–2014 by BiofuelsDigest.com.

CONTINUED ON NEXT PAGE

Products and their characteristics:

Rivertop is developing multiple renewable products based on its glucaric acid platform. One example is *Riose™* detergent builder—a novel, cost-effective product to replace phosphates and other less effective and/or nonrenewable builder ingredients in automatic dishwashing detergents. Due to its high binding capacity and multifunctionality as a chelant and corrosion inhibitor, variations on this chemistry are suitable for a range of industry applications in consumer and I&I detergents, water treatment, and related industries. In addition to producing glucaric acid from glucose, Rivertop's platform technology works quite well with a range of natural feedstocks. This enables development of multiple unique novel products. In the longer term, Rivertop's sugar acids will be developed as advanced biodegradable polymers, adhesives, and other functional materials.

Commercialization and scale:

Rivertop plans to be producing commercial quantities of its products at intermediate commercial scale (approximately one million pounds—or 450 thousand kilograms— per month) in the fourth quarter of 2014 with full regulatory approvals in place. Market development is under way in the consumer detergents sector, and the company is directly engaged with key players in the industry.

Business development model and plans:

Rivertop plans to interface directly with leading producers in the consumer detergents market to reach global customers. The company is seeking, and in some instances has begun, commercial development partnerships that will allow it to serve additional markets for its technology platform and products.

Key challenges:

Novel chemicals require a higher burden of proof to qualify for replacement of existing ingredients. Accordingly, Rivertop is focused on markets driven by needs for better cost/performance and sustainability. Investments in new, sustainable, and multifunctional chemicals provide a durable competitive advantage to customers. Co-developing applications with research-driven customers and partners accelerates market adoption of our novel chemicals.

Development of product applications knowledge and performance testing are critical success factors in most of the markets the company plans to serve. Rivertop has invested in its own in-house testing capabilities with respect to consumer auto-dish application, including a laboratory full of dishwashers. In other markets, the company has conducted proof of concept work in-house and has also consulted with potential partners to understand the performance requirements of prospective product applications.

INHERENT™ RENEWABLE BUILDING BLOCKS: DI-FUNCTIONAL ESTERS, RENEWABLE OLEFINS, AND NOVEL MONOMERS

***Company*:** Elevance Renewable Sciences, Inc.

Feedstock: multiple renewable oil feedstocks, including palm, canola, soybean, rapeseed, mustard, to name a few, and, when they become commercially available, new feedstocks such as jatropha and algal oils

***Synthetic process*:** proprietary metathesis technology (based on Nobel Prize-winning chemical catalysts)

***Potential applications*:** surfactants (consumer and industrial cleaning, dispersion, emulsification, rheology, and de-foaming) and personal care (hair care, skin care, film-forming, emollients, and moisturizers)

***Key targeted market segment(s)*:** personal care, home care, agriculture, oil field, paints and coatings, construction materials, biocides, performance waxes

***Contacts*:** Andy Corr, senior vice president, consumer and industrial ingredients, andy.corr@elevance.com; Andy Shafer, executive vice president, sales and market development, andy.shafer@elevance.com

Key aspects of products/technology:

Elevance uses Nobel Prize-winning metathesis catalyst technology to transform renewable oils into novel, high-performing ester, olefin, and triglyceride molecules. The company's unique process has allowed it to scale up easily at high yields and commercially compelling economics. The technology produces a diverse range of building blocks and ingredients through a low-pressure, low-temperature process that consumes significantly less energy and reduces greenhouse gas (GHG) emissions by 50% compared to petrochemical technologies, resulting in low source pollution, production costs, and capital expenditures.

How they compare with traditional (petrochemical or oleochemical) products and surfactants:

Elevance Inherent™ renewable building blocks provide pathways for game-changing performance in surfactants, enabling improved cold-water performance, better degreasing and solvency, improved concentration, and low content of volatile organic compounds (VOC). Elevance Smooth and Soft personal care ingredients provide renewable, silicone-free replacements that provide smooth sensory attributes (silky without sticky feel), unique film-forming characteristics (longer-lasting, moisture barrier), and improved compatibility in formulations.

What differentiates them from other renewable products and technologies?

Elevance technology is one of the first renewable technologies that has reached world-scale commercial production through both the company's 180,000-metric-tons (MT)-per-year biorefinery and toll manufacturing processes. The company is

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focused on providing products that deliver enhanced performance, are derived from renewable feedstocks, and offer sustainable solutions—a new category of materials they call Renewicals™, that meet customer needs that couldn't be met before in personal care, surfactants, and performance waxes. Its materials are available globally as building blocks and ingredients, providing compelling cost performance today.

Awards/recognitions:

Nobel Prize-winning chemical catalysts are at the heart of Elevance's technology. The company was named in *Business Week*'s "Most Successful Start-ups of 2008," won the US Environmental Protection Agency Presidential Green Chemistry Award in 2012, and was named among *Crain*'s "Top 25 Most Innovative Companies" in 2012. It has been included in the Global Cleantech Top 100 list multiple times and it has been listed in the top of *Biofuels Digest*'s "30 Hottest Companies in Renewable Chemicals and Materials" for several years.

Products and their characteristics:

Elevance Smooth personal care ingredients are sold by a global distributor network into leading skin care, hair care, lip balm, and color cosmetic products. They include Elevance Smooth CS110 and Elevance Smooth CG100, both 100% natural renewable emollients that provide unique smooth skin feel, film-forming characteristics, and rheology modification (thickening). In addition, Elevance NatureWax™ products are sold worldwide. The company's partners, including Stepan, are now introducing novel products globally, based on Elevance's Inherent renewable building blocks, into consumer and industrial applications, such as home care, personal care, agriculture, construction, and oilfield, as well as other applications that require high-performance degreasing, cold-water cleaning, and unique foaming characteristics.

Commercialization and scale:

Elevance has been producing and selling its Elevance Smooth and Soft personal care products and Elevance NatureWax line globally for more than five years. It is also now operating its first world-scale biorefinery in Asia based on the company's propriety metathesis technology, has a capacity of 180,000 MT, with the ability to expand up to 360,000 MT of products. Elevance has also announced it is building a second plant to be based in Natchez, Mississippi, USA, that will have a capacity of 280,000 MT. Elevance products have broadly received regulatory approvals in the United States (Toxic Substances Control Act, or TSCA) and Europe (Registration, Evaluation, Authorisation and Restriction of Chemicals, or REACH) and are being offered globally directly, through its global distribution network for personal care. The company's various partners are also now offering products based on Elevance's Inherent™ renewable building blocks.

Business development model and plans:

Elevance's model it is to work with a broad range of partners to bring together complementary capabilities (assets, market expertise, distribution capabilities, R&D, etc.) to develop products that meet customer needs quickly, scale up cost effectively, reach markets rapidly, and support customer application development. Elevance's collaborative partnership model works to create value through the supply chain and facilitate speed to market. Inherent™ renewable building blocks represent a new, versatile tool for companies interested in applying their chemistry to create innovative new derivatives. Elevance also believes there is significant potential for creating new ingredients for companies that need or wish to focus on solutions that can simply be formulated. The company continues to expand its collaborations and partnerships worldwide.

Key challenges:

There are large hurdles to innovation, including regulatory approvals, prolonged reformulation efforts, and qualification processes that exist across the value chain. It takes time for companies and markets within our industry to shift and adjust to innovations, like a large ship trying to turn. Elevance is addressing these hurdles by partnering with all stakeholders—including government agencies, brand owners, R&D labs, and manufacturing partners—up and down the value chain. This enables Elevance to address challenges early, and to work with the appropriate stakeholder to navigate risks rapidly and effectively to bring innovative products to partners and consumers across markets.

TAILORED™ ALGAL OILS

***Company*:** Solazyme, Inc.

***Feedstock*:** feedstock flexible (low-cost agricultural materials such as sugarcane-based sucrose, corn-based dextrose, and sugar from other biomass sources, such as cellulosics)

***Synthetic process*:** Solazyme's process is fermentation-based and involves aspects of molecular biology combined with classical strain improvement.

***Potential applications*:** specialty chemicals (e.g., fatty alcohols, dimer acids), plastics (e.g., polyurethane), functional fluids (e.g., lubricants and greases, plasticizers, solvents, transformer dielectric fluids), and home & personal care (e.g., soaps, surfactants, emollients)

***Key targeted market segment(s)*:** healthful frying oils and food ingredients, high-performance industrial and specialty chemicals, lubricants and greases, solvents, and fatty alcohols and surfactants

***Contact*:** Sales@solazyme.com; SalesBrazil@solazyme.com

Key aspects of products/technology:

Solazyme's breakthrough Tailored™ Algal Oils technology platform uses microalgae to convert non-nutritious, inedible sugars into renewable, Tailored™ Algal Oils for multiple markets—this is the first bridge between what the Earth is good at making (carbohydrates) and what society runs on (oils). Using standard industrial fermentation, Solazyme has efficiently built a platform that produces new, sustainable oils in a matter of days.

How they compare with traditional (petrochemical or oleochemical) products:

Solazyme's Tailored™ Algal Oils provide a unique opportunity to develop and produce next-generation functional fluids and specialty chemicals that boast a number of increased performance benefits compared to those currently available. For example, the company's Tailored™ Algal Oils offer increased fire safety over petroleum-derived oils thanks to a significantly higher flash point. Key performance properties of Solazyme's Tailored™ Algal Oils include biodegradability, which significantly reduces the need for environmental cleanup in the event of a spill; increased thermal and oxidative stability, which potentially improve the performance in a wide range of applications from fiber lubricants to dielectric insulating fluids; and overall viscosity control, as Solazyme's Tailored™ Algal Oils provide an excellent viscosity index that results in consistent fluid properties over a wide temperature range (see case study on page 128). Solazyme's renewable Tailored™ Algal Oils are the first bio-based oils with the quality, purity and consistency of industrially produced petroleum-derived chemicals.

What differentiates them from other renewable products and technologies?

Solazyme can create oils that are uniquely tailored to address specific customer requirements, offering a high-value solution with superior performance at a competitive price and an enhanced value compared to conventional oils. Solazyme's Tailored™ Algal Oil technology means that the company can deliver the highest-value cuts of the barrel of oil to its partners and deliver a completely uniform barrel of oil designed for a specific purpose or product so there is a reduction of waste overall in the process. In lubricant and functional fluid applications, Solazyme renewable Tailored™ Algal Oils represent a paradigm shift. For the first time oils can be designed

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to deliver "best of both" performance with the benefits of bio-based oils (lubricity, fire point, viscosity index, etc.) plus performance of synthetics (thermal /oxidative stability, pour point, hydrolytic stability, etc.)

Awards/recognitions:

Solazyme has received numerous industry recognitions and awards. For example, Solazyme won *Popular Science*'s "Best of What's New" in 2012. Solazyme's CEO, Jonathan Wolfson, was selected as one of *Forbes*' "12 Most Disruptive Names in Business" in 2013. The company was selected as a 2012 Technology Pioneer by the World Economic Forum. Solazyme was named No. 1 in the *Biofuels Digest* "50 Hottest Companies in Bioenergy" for 2010–2011 and for 2012, and it was also awarded the *Biofuels Digest* "Company of the Year" award in 2010. Additionally, the company was named to *Time* magazine's top 20 Green Tech ideas of the year for 2010.

Products and their characteristics:

Solazyme is offering renewable Tailored™ Algal Oils to meet the performance and functional demands of the chemicals industry. By tailoring the composition of the oil, the company is able to develop oils that meet customer needs including improved purity, oxidative stability, fire point, solubility, and flash point. Solazyme's oils also provide low VOC, light color and color stability, and low odor.

Commercialization and scale:

Solazyme has production capability at a range of scale, and the company is rapidly scaling up technology to produce Tailored™ Algal Oils for the chemicals industry. The company's semi-commercial facility in Peoria, Illinois, USA, is fully operational and producing demonstration-scale volumes of oils today. In December 2012, Solazyme announced successful completion of multiple initial fermentation runs at the Archer Daniels Midland (ADM) facility in Clinton, Iowa, USA. Solazyme is initially targeting annual production of 20,000 metric tons (MT) of oil starting in early 2014 at the ADM facility, with targeted expansion to 100,000 MT. The scale achieved at ADM's Clinton facility is comparable to the fermentation equipment currently under construction at the Solazyme Bunge Renewable Oils facility in Orindiúva, Brazil. This 100,000 MT annual nameplate capacity facility in Brazil, expandable to 300,000 MT, is scheduled to come online in the first quarter of 2014.

Key challenges:

Solazyme has already been operating at commercial levels, producing hundreds of thousands of gallons of oil for chemical and industrial applications. The company has continued to demonstrate progress toward building out the key infrastructure, partnerships, and joint venture agreements needed to produce Solazyme oils at scale for multiple markets and industries.

METABOLICALLY ENGINEERING YEAST TO PRODUCE TERPENES

***Company*:** Allylix, Inc.

***Synthetic process*:** fermentation using yeast that has been metabolically engineered to produce terpenes

***Potential applications*:** allows for the commercial development of natural terpenes and their derivatives, as well as the creation of novel compounds

***Key targeted market segment(s)*:** current focus on flavor and fragrance industry; plans to target insect control, cosmetic chemicals, and food ingredients

Contact: Carolyn Fritz, CEO, info@allylix.com

Key aspects of products/technology:

Allylix's multifaceted technology platform includes all of the molecular biology and engineering elements necessary to go from gene isolation to commercial scale production of terpenes, including gene cloning and expression, metabolic engineering, protein engineering, fermentation development, recovery and purification, and organic chemistry. Specifically, Allylix's technology platform consists of: (i) terpene cyclase gene cloning and expression, (ii) protein engineering to improve specificity and/or activity of terpene cyclases, (iii) yeast metabolic engineering to produce terpenes, (iv) fermentation development and recovery processes for the production of sesquiterpene products, and (v) production of commercially inaccessible or novel sesquiterpenes by chemobiosynthesis of combinatorial libraries.

How they compare with traditional (petrochemical or oleochemical) products and surfactants:

Allylix's technology platform allows it to produce terpenes biosynthetically. Because most terpenes have complex structures, historically there has been no cost-effective way to produce them. By producing them biologically, Allylix can now commercially produce this important class of chemicals.

What differentiates them from other renewable products and technologies?

Unlike other companies in the renewable chemicals industry, Allylix can produce terpenes. Because Allylix's scientific founders were pioneers in the field, the company has a strong intellectual property portfolio.

Awards/recognitions:

Allylix won the 2012 FiFi Technological Breakthrough of the Year award in the Fragrance Ingredients, Creation and Testing category; as well as the *San Diego Business Journal*'s Innovation Award in the biotechnology category in 2013.

Products and their characteristics:

Allylix currently offers three flavors and fragrances: valencene, nootkatone, and Epivone™. Valencene is an orange flavor and fragrance that traditionally was extracted from the peel of Valencia oranges, meaning the cost and supply would fluctuate depending on the yearly harvest. Nootkatone is the flavor and fragrance material that gives grapefruit its unique aroma. It is a fresh, citrus, woody note and is naturally present in grapefruit peel oil in minute quantities, although in the past it has primarily been produced by the oxidation of valencene. Epivone is a novel patented and trademarked compound produced by Allylix that is structurally related to β-vetivone, one of the key components of vetiver oil. It is characterized as woody, cassis, rich yet fruity, and containing a grapefruit effect.

Commercialization and scale:

Allylix is currently at full commercial scale and selling product. The company also has a pipeline of products.

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Business development model and plans:

The company is taking products currently in Allylix's pipeline of flavors and fragrances to market directly. For products outside of that pipeline, Allylix is working with partners to bring compounds to market.

Key challenges:

As Allylix completes its flavor and fragrance pipeline, the next challenges the company faces includes expanding into new markets, such as insect control.

LEVULINIC ACID AND DERIVATIVES SUCH AS LEVULINIC KETALS

Company: Segetis, Inc.

***Feedstock*:** any cellulosic or hemicellulosic biomass

***Synthetic process*:** hydrolysis of sugars and esterification

***Potential applications*:** plasticizers for flexible PVC and biopolymers, formulation aids for detergents and cleaners, solubilizers for agrochemical formulations, personal care and cosmetic formulations, intermediates for polyamides and polyesters, and ionic polymers

***Key targeted market segment(s)*:** PVC and biopolymer compounders, household, I&I, agrochemicals, personal care, and polyamide

***Contact*:** Graham Merfield, graham_merfield@segetis.com

Key aspects of products/technology:

Proprietary process for converting sugar to levulinic acid in an efficient, economical way. Proprietary levulinic ketal products. Levulinic acid and levulinic ketals are chemical building blocks that can be used to make a very diverse product portfolio that spans novel, specialty products to drop-in replacements of commodity chemicals.

How they compare with traditional (petrochemical or oleochemical) products and surfactants:

Segetis' levulinic ketal products are new-to-the-world renewably sourced chemicals that offer unique performance profiles. One of Segetis' plasticizers, for instance, is a biobased phthalate-free substitute for the banned butyl benzyl phthalate with improved volatility and superior resistance to nonpolar extraction. The company's formulating aids help hold liquid formulations together and help incorporate difficult-to-use actives. Levulinic ketals are made from levulinic acid, which is a decomposition product of cellulose and hemicellulose. Levulinic acid itself has a unique oxygen-rich chemical functionality—a ketone group connected to an acid group—that enables transformation into a wide array of chemical products that will be impacted by the development of shale gas. Shale gas has been widely heralded as a game changer for the energy and chemicals markets; however, it is increasing availability of only C_1–C_4 petrochemical feedstocks. Industry continues to rely on naphtha and other heavier oil inputs for the C_4–C_8 materials. The molecular composition of levulinic acid provides a pathway to a broad range of C_4–C_8 molecules that are currently sourced from petroleum. Segetis' levulinic acid technology therefore addresses both the upcoming naphtha shortages and the drive toward biomass-based alternatives with advantages in terms of sustainability and a more efficient carbon footprint.

What differentiates them from other renewable products and technologies?

All of Segetis' technologies are based on thermochemical conversion, which provides a biobased chemical platform with the scale-up ease and purity of a traditional chemical product. The technology is suitable for a wide range of feedstocks, giving us the flexibility to source the best available feedstock based on the locale and the economics.

Awards/recognitions:

Early commercial success with levulinic ketal products and clearance of chemical registration hurdles in three of the largest market areas are vital recognition of Segetis' technology.

Products and their characteristics:

Levulinic acid and levulinic ketals are chemical building blocks that are easily derivatized into a range of products. Segetis' levulinic ketal derivatives are currently used as formulation aids and as polymer plasticizers. Levulinic acid is an intermediate for nylon, polylactones, and water-soluble ionic polymers.

Commercialization and scale:

Segetis has several products that are listed on TSCA, Canadian Non-Domestic Substances List (NDSL), and are registered under REACH. The company is currently at the demonstration scale. The next step is to scale up to commercial scale.

Business development model and plans:

The company is working with strategic customers across target market segments to validate its technology and products using material from our demonstration-scale facility. Segetis works directly with these customers to develop firsthand knowledge of the market and customer needs with support from its in-house applications development team.

Key challenges:

Levulinic acid has been identified as a platform chemical for the future (*Top Value Added Chemicals from Biomass Volume I*, US Department of Energy, 2004). For this to become a reality, the market for the downstream derivatives and scaled-up levulinic acid production must be developed so that the economics and capacity to fully realize the market potential can be achieved.

ITACONIX DSP™ POLYMER

***Company*:** Itaconix Corp.

***Feedstock*:** itaconic acid, which is produced by the fermentation of sugar

***Synthetic process*:** polymerization

***Potential applications*:** chelation and dispersion in detergents, cleaners, and personal care

***Key targeted market segment(s)*:** replacement of phosphates and ethylenediaminetetraacetic acid (EDTA) in consumer detergents, I&I detergents, and cleaners

***Contact*:** John Shaw, CEO, +1 603 686-7550, jshaw@itaconix.com

Key aspects of products/technology:

Itaconix DSP polymers are the most cost-effective materials available for chelating calcium and other metal ions to improve the performance of detergents and cleaners. The polymers are made from renewable resources and are readily biodegradable.

How they compare with traditional (petrochemical or oleochemical) products and surfactants:

Itaconix DSP polymers bind more calcium per gram than EDTA or phosphates. With their additional dispersion and detergency properties, these polymers can improve the performance and reduce the cost of standard and green detergent and cleaner formulas.

What differentiates them from other renewable products and technologies?

Itaconix entered the market with its first commercial polymer sales in 2009 and remains the leading

CONTINUED ON NEXT PAGE

commercial producer of polyitaconic acid in the world. Its polymers are in consumer products on store shelves across North America and Europe and are gaining increased adoption and use based on novel performance and cost efficacy.

Awards/recognitions:

Lux Research named Itaconix one of its top 10 emerging technologies of 2012.

Products and their characteristics:

Itaconix DSP polymers have a combination of chelation, dispersion, and detergency properties that are more similar to those of phosphates than other standard or green alternatives. Customers in consumer and I&I detergent markets are able to formulate products with comparable performance to phosphate-based products.

Commercialization and scale:

We sell and provide technical and formulation services directly to companies in North America and Europe. Products are available directly from the company's production facility in the state of New Hampshire and through distribution in the United States and Europe.

Key challenges:

The reformulation process for detergents and cleaners can take up to 18 months to complete. The company is increasing the awareness of its polymers and the advantages of formulations based on its polymers with technical and formulation services.

***Company*:** Virent

***Feedstock*:** Conventional and cellulosic biomass-derived carbohydrates

***Synthetic process*:** BioForming via catalytic chemistry

***Potential applications*:** biobased chemicals for fibers and plastics, as well as biobased gasoline, diesel, and jet fuel

***Key targeted market segment(s)*:** beverage/bottling companies, diesel and jet fuel customers

Contact: Kieran Furlong, Kieran_Furlong@virent.com

Key aspects of products/technology:

Virent's process to make chemicals (and also hydrocarbon fuels) is based on a novel combination of its APR technology with modified conventional catalytic processing technologies. The process, trademarked BioForming®, uses catalytic chemistry to convert aqueous carbohydrate solutions into a mix of hydrocarbons. The BioForming platform expands the utility of the APR process in combination with catalysts and reactor systems similar to those found in standard petroleum oil refineries and petrochemical complexes.

How they compare with traditional (petrochemical or oleochemical) products and surfactants:

Virent's biobased chemicals are molecularly identical to their petrochemical counterparts; therefore, there is no compromise in quality or performance. Unlike petrochemicals, Virent's technology can utilize many different feedstocks, giving companies an opportunity to diversify their supply chain to combat volatile crude oil prices. For any country that is a net importer of crude oil, biobased hydrocarbons can decrease reliance on imported oil and provide development opportunities for agriculture and forestry sectors as well as rural communities.

Compared to petroleum as a feedstock, Virent's process can reduce greenhouse gases (GHG) by up to 55% by using conventional sugars.

What differentiates them from other renewable products and technologies?

- Products are premium substitutes for their conventional counterparts. Unlike newer novel chemicals, Virent's products are molecularly identical to their petrochemical counterparts. They are referred to as "drop-in," meaning they can utilize today's infrastructure investment and be dropped into existing supply chains.
- The molecules on which products are based are well known and understood. While some companies are attempting to make new types of plastics from novel molecules, Virent's strategy is to make a drop-in molecule offering desired performance characteristics that leverage years of previous research and commercialization and, in turn, accelerates time to market.
- The BioForming process has the ability to use conventional and nonconventional sugars for conversion to fuels and chemicals. It can generate products from mixed sugar streams, polysaccharides, and fermentation inhibitors such as furans that are formed during biomass deconstruction. This greatly increases Virent's ability to use cellulosic biomass as a viable and renewable feedstock. Virent's BioFormPX product offers companies an opportunity to diversify their supply chains to combat volatile crude oil prices.
- Products have low carbon footprints. Preliminary greenhouse gas emission calculations have been developed using US corn, European sugar beet, and Brazilian sugarcane as potential feedstocks in an aromatics plant configuration using an economic allocation basis. Virent estimates reduction in GHG emissions of approximately 30%, 35% and 55% using US corn, EU sugar beets, and Brazilian sugarcane, respectively, vs. a petroleum baseline.

Awards/recognitions:

ICIS Chemical Business Innovation Award for Best Innovation by a Small and Medium-Sized Enterprise (2008), Red Herring North America Award (2008), World Economic Forum Award (2008), EPA's Presidential Green Chemistry Challenge Award (2009), Global Cleantech 100 Award (2010), ICIS Chemical Business Innovation Award for Sustainability (2013), second runner-up in Securing America's Future Energy's Emerging Innovation Award (2013) among others.

Products and their characteristics:

The company is currently in the product development phase. It has proven that its technology works and is aggressively seeking partnerships to help de-risk and scale up its technology to bring premium products to market as soon as possible.

NOVELTY VS. EQUIVALENCY

For those familiar with the history of renewables, the recent trend toward novel chemicals is a sign that the industry has come full circle. Initially, some of the first renewables to be developed *were* novel chemicals, but high switching costs, regulatory challenges, and the burden of having to prove to potential customers that such novel chemicals would work exactly the same in an engine or other piece of equipment as the chemical it was designed to replace caused most small startups to adapt their business strategy and focus on drop-ins.

Virent is a prime example of a company that successfully directed its technology toward drop-in chemicals by combining its novel Aqueous Phase Reforming (APR) technology with modified conventional catalytic processing technologies. The resulting premium drop-in BioForming® technology expands the utility of the APR process in combination with catalysts and reactor systems similar to those found in standard petroleum oil refineries and petrochemical complexes.

Commercialization and scale:

Virent intends to move to a 1,000× scale-up of its process in a semi-works plant that will produce tens of thousands of tons of the primary product, *para*-xylene. This will likely be done in conjunction with a strategic manufacturing partner, and timing and ultimate scale will be very dependent on who that partner is and what infrastructure it has. Virent is currently targeting commercial sales by 2016.

Business development model and plans:

Virent aims to commercialize its products through partnerships and is open to joint development and licensing agreements as well as joint ventures. The company believes that focusing on *para*-xylene will allow it to generate positive cash flow in the shortest time, and it is actively engaging in the selection of a strategic chemical manufacturing partner.

Key challenges:

It's challenging to compete against an existing industry with established supply chains and depreciated infrastructure. However, Virent believes in the promise of its technology and is developing innovative concepts to lower costs and offer competitively priced products.

The obstacles Virent faces are not uncommon to other renewable fuel and chemical companies, which are primarily difficulty in accessing capital required to scale up a novel technology. The difference is that Virent has developed needed capabilities and forged key partnerships/relationships to mitigate those risks.

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Governing Board Candidates for 2014–2015

Ballots for the election of the American Oil Chemists' Society Governing Board members are being sent to eligible members by mail or e-mail in December. The deadline for returning completed ballots (either electronically or via mail) is February 15, 2014.

On this ballot, you will have an opportunity to vote in the following categories: AOCS President, Vice President, Secretary, and five (5) Members-at-Large. Photographs and biographical information are presented for each candidate, followed by the opportunity to vote in each category. Please choose one candidate (or write in a candidate) in each category, barring the Member-at-Large category, where you can choose five candidates.

This system will allow you to submit your ballot only a single time.

The new officers will be installed during the 105th AOCS Annual Meeting & Expo in San Antonio, Texas, USA, on Monday, May 5, 2014. Thank you for voting.

PRESIDENT CANDIDATE

Hill

Steven Hill (1987): Vice President, Cheese and Dairy R&D and Quality, Kraft Foods, Glenview, Illinois, USA.

Education: B.S., 1987, Food Science, Iowa State University; M.S., 1989 and Ph.D., 1992, Food Science, University of Illinois.

Previous Employment: Senior Director, Central Research, Kraft Foods, 2012–2014; Director, Cheese R&D, Kraft Foods, 2007–2012; Director, Health and Wellness Policy, Kraft Foods, 2005–2007; Associate Director, R&D Sauces and Dressings, Kraft Foods, 2003–2005; Section Manager, R&D Strategic Research, Kraft Foods, 1999–2003; Group Leader, R&D Ingredient Technology, Kraft Foods, 1997–1999; Senior Scientist, R&D Oil Technology, Kraft Foods, 1995–1997; Scientist, R&D Product Development Oil Products, Kraft Foods, 1992–1995.

AOCS Activities: Vice President, AOCS Governing Board, 2012–present; secretary, AOCS Governing Board, 2007–2009; treasurer, AOCS Governing Board, 2003–2007; member-at-large, AOCS Governing Board, 1999–2003; associate editor, *inform*, 1992–2001; chairperson, *JAOCS* Editor-in-Chief Search Committee, 2000; chairperson, AOCS Governance Committee, 2001–2002; member, Publications Steering Committee (PSC), 2002–2003; member, Financial Steering Committee (FSC), 2001–2002; member, Technical Steering Committee (TSC), 1999–2000; member, Information Technology Management Committee, 1999–2002; member, Campaign AOCS Committee, 1999–2001; local Committee member, 1995 and 1996 Annual Meetings; vice technical chairperson, 1998 Annual Meeting; member, *inform* Editor-in-Chief Search Committee, 1997; member, Edible Applications Technology and Lipid Oxidation & Quality Divisions, 1997–present; president, North Central Section, 1998; vice president, North Central Section, 1997; board member-at-large, North Central Section, 1992–1996; chairperson and member, A.E. Bailey Award Selection Committee, 1998–2001.

Other: Fellow, AOCS, 2012; member, University of Illinois Food Science and Human Nutrition External Advisory Board; adjunct assistant professor, University of Illinois, Department of Food Science and Human Nutrition, 1993–2000; member, Iowa State University Food Science and Human Nutrition Advisory Board, 2002–present; AOCS Honored Student Award, 1991; AOCS Ralph Potts Fellowship Award, 1990; passionate advocate for enhanced AOCS membership services.

Research Interests: Lipid oxidation in food systems; oilseed processing; nutritional lipids related to foods and health.

VICE PRESIDENT CANDIDATE

Trautmann

Manfred Trautmann (2004): Managing Director, Detergents & Intermediates, WeylChem Switzerland, Muttenz, Switzerland, February 2009–present.

Education: B.Sc., 1976, Chemical Engineering, University of Darmstadt, Germany.

Previous Employment: Global Head of Marketing & Sales, RBU Detergents & Intermediates, 2007–2009; Head of New Business Development of the Business Unit Detergents, 2000–2007; Global Marketing & Sales Manager for the Detergents, Personal Protection Additives

Business of the Surfactants/FUN Division during the transition of Hoechst to Clariant, 1996–2000; Product Manager, National Accounts Manager, and Marketing Manager for the Detergents and Personal Care Business of American Hoechst/Hoechst Celanese, North America, Charlotte, North Carolina, 1986–1996; Application Manager for Detergents and Personal Care in R&D, Hoechst AG, 1979–1986; Project Engineer in the Hoechst Engineering Department, 1976–1979.

AOCS Activities: Member-at-Large, AOCS Governing Board, 2011–present; presenter, speaker, and/or co-author, various AOCS Annual Meeting & Expos; member, Executive Committee, 7th & 8th World Conference on Detergents (Montreux Conference), and Singapore 2010 & 2012; member, Organizing Committee, 5th & 6th World Conference on Detergents (Montreux Conference), 2002, 2006.

Other: Member, Organizing Committee, CESIO 2004, Berlin, Germany.

Research Interests: Green chemistry, including chemicals and/or intermediates derived from bioethanol, biodiesel, and glycerol.

VICE PRESIDENT CANDIDATE'S STATEMENT

The very first AOCS Annual Meeting I attended happened to be in New Orleans in 1982, in association with my first business trip to the United States. It had a major impact in my association with the AOCS organization. Not much later, I joined the society and soon thereafter participated in the Annual Meetings with technical presentations. While I was still located in Europe, I made it to every Annual Meeting, which continued when I moved to Charlotte, North Carolina, USA in 1986. During the 10 years I spent in Charlotte, I was eager to learn more about the business, but I was also drawn to AOCS by the breadth and depth of scientific work that was published in *JAOCS*. Other colleagues in our organization at the time were members of AOCS and maintained an archive of *JAOCS*. I spent many hours of my free time learning from that archive.

Later, when I returned to Europe with Hoechst, I continued to attend the AOCS Annual Meetings, as well as the Montreux Conferences held in Europe. AOCS has always been a great source of technical information, as well as a great place for networking for me, which has undoubtedly helped me in my career development within Hoechst/Clariant over so many years. For that, I am eternally grateful to AOCS.

Starting with the Montreux Conference in 2000, I became involved in the preparation of this convention in the Organization Committee, Executive Committee, and as Vice Chair in 2010. We started to organize the Montreux-type convention for the very first time 2012 in Singapore, and I will serve as the General Chair of Montreux 2014, "The World Conference on Fabric and Home Care." As I have become more involved in the management of AOCS at the Board level over the past years, as a Member-at-Large and a Board Member, I have learned much more about the value that AOCS represents to its individual members and to its constituent interested parties.

AOCS represents an interesting intersection of constituent interests—industry, academia, and government—in the area of oilseed processing and its derivative products. My specific interests are in the surfactant chemistry, household, and personal care areas. The role that AOCS plays as an independent, scientifically based entity in publishing, education, standardization of analytical methods, and assistance in the harmonization of global trading rules has never been more relevant than in this new age of instant electronic communication and globalization of industry.

The emergence of natural oil/oilseed products playing an increasingly important role as raw materials in many different industries, thus replacing traditional petrochemical-based raw materials, is one trend that we at AOCS need to monitor closely and to support across our whole organization. It is my 30+ years of experience in the globalization of end-user markets in the detergents and personal care industry, combined or based on natural oil/oilseed processing, that I believe I bring to the role of Vice President, which will help AOCS grow internationally, continuing to strengthen its brand already recognized for independence and excellence.

SECRETARY CANDIDATE

Widlak

Neil R. Widlak (1977): Retired

Education: B.S., 1975 and M.S. 1980, Food Science, University of Illinois, Champaign-Urbana, Illinois; M.B.A., 1985, Lake Forest Graduate School of Management, Lake Forest, Illinois.

Previous Employment: Director, Product Services and Development, ADM Cocoa, Milwaukee, Wisconsin, March 2009–October 2013; Director, Strategic Technology Development, Archer Daniels Midland Co., Decatur, Illinois, 1995–March 2009; manager, Chemistry and Applications, Intermountain Canola, Pennsauken, New Jersey, 1992–1995; manager, research and development, Lou Ana Foods, Opelousas, Louisiana, 1989–1992; section manager, Kraft Foods, Glenview, Illinois, 1976–1989; refinery supervisor, Best Foods, Chicago, Illinois, 1975–1976.

AOCS Activities: Secretary, AOCS Governing Board, 2012–present; second vice chairperson, Membership Steering Committee, 2009–2012; member-at-large, AOCS Governing Board, 2008–2012; member-at-large, Edible Applications Technology Division, 2001–present; member-at-large, Industrial Oil Products Division, 2000–2002; chairperson, Award of Merit Canvassing Committee, 1999–2004; co-author/instructor, short courses on Refining of Fats and Oils (North and Latin America), Physical Properties of Fats and Oils, Applications of Fats and Oils in Baking; session chairperson, Symposia on Fat Crystallization, 1997–present; chairperson, Conference on Physical Properties of Fats, Oils, and Emulsifiers, 1997 and

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2000; chairperson, Edible Applications Technology Division, 1996–2001; technical chairperson, AOCS Annual Meeting & Expo, 1988; member, Education and Meetings Steering Committee (EMSC), 1987–1990; member-at-large, AOCS Governing Board, 1984–1985; chairperson and member-at-large, North Central Section, 1980–1989.

Berbesi Anaya

MEMBER-AT-LARGE CANDIDATES (5)

Roberto Berbesi Anaya (2007): Sales Manager LATAM/ASIA Oil-Dri Corporation of America.

Education: B.S., 1990, Chemical Engineering, Universidad del Valle, Cali, Colombia; M.B.A., 1994, Universidad Javeriana, Cali, Colombia.

Previous Employment: Technical Service Engineer for the Americas, Oil-Dri Corporation of America, 2001–2007; Production Manager, Lloreda S.A., Cali, Colombia 1995–2001; Process Optimization Engineer, Lloreda S.A., Cali, Colombia 1992–1995; Shift Engineer, Lloreda S.A., Cali, Colombia 1990–1992.

AOCS Activities: Co-author of chapter in the Lipid Library, 2013; technical committee member and chairperson over two sessions, LAOCS Congress, Chile, 2013; chairperson, LAOCS Congress, Cartagena, 2011; treasurer, Processing Division, 2012–2014; president, LA Section, 2010–2012; chairperson, LAOCS short course, Cartagena, Colombia, 2009; vice-president, LA Section, 2008–2010; author of IOMSA magazine article on bleaching, 2006; governing board member, LA Section, 2004–present; short course presenter, LAOCS, 2003–2012; short course presenter, AOCS, 2002–2010; member, Processing Division, 2001–present; member, LA Section, 2001–present.

Other: Presenter, Texas A&M Short Courses, 2007–2011; In the process of creating an optional subject on the topic of processing vegetable oils for engineering curriculum in a Colombian university.

Research Interests: New technologies to advance current best practice in oils refining; effects of processing conditions in refining and bleaching on the finished quality of the oil; sustainable ways to manage spent bleaching earth.

Della Porta

Richard (Rick) Della Porta (1991): Senior Principal Scientist, Frito-Lay North America Research and Development Analytical Laboratories, Plano, Texas, 1984–present.

Education: B.S., 1981, Environmental Toxicology, University of New England, Maine; M.S., 1985, Chemistry, University of Texas at Dallas, Texas.

AOCS Activities: Chairperson, Core Value Center Networking Committee, 2012–present; member, Governing Board Finance Tracking Strategic Working Committee, 2011–present; chairperson, Meetings and Education Committee, 2011–2012; member-at-large, AOCS Governing Board, 2007–2009, 2011–present; chairperson, Analytical Division, 2010–2011; vice chairperson, Analytical Division, 2008–2010; chairperson, General Analytical AOCS Annual Meeting Session, 2007–2012; treasurer, Analytical Division, 2006–2008; representative, Membership Development Committee, Analytical Division, 1999–2006; member, Uniform Methods Committee, 2002–2010; Student Mentor Breakfast Host, 2000–2007; member of Lipid Oxidation and Quality Division and Analytical Division, 1992–present.

Other: Editor of Snack Food Association/AOCS Edible Oils Manual 2005; member, AOAC International, 1992–2003; manuscript reviewer, *Journal of Agricultural and Food Chemistry*, 2003–present.

Research Interests: Analytical method development; analysis of lipids and their oxidative reactivity to industrial processes; development and application of rapid, non-destructive analysis techniques to provide real-time feedback for process control of continuous flow fryers; correlation of oil properties to shelf life performance and consumer response; corollary work in lipid-soluble component analysis, with emphasis on nutritional applications and implications; close-working relationship with development groups involved in trans-fat replacement; nutraceutical additives analysis and nutrition labeling; most recently involved in adapting nanotechnology to food applications.

Moreau

Robert A. Moreau (1990): Lead Scientist, Eastern Regional Research Center, Agricultural Research Service, United States Department of Agriculture, Wyndmoor, Pennsylvania, USA

Education: B.A., 1974, Boston University; Ph.D., 1979, University of South Carolina.

Previous Employment: University of California, Davis, 1979–1981.

AOCS Activities: Member-at-Large, Governing Board, 2011–present; Editorial Advisory Board of JAOCS, 2008–present; chair of the Publications Steering Committee, 2003–2011; member, Books and Special Publications Committee, 2002–present; vice chair, Publications Activities Coordination Committee, 2000–2003; canvassing chair for the George Schroepfer Medal Award, 2001–present; co-chair, numerous AOCS Symposia including the AOCS Sterol Symposia, 2000–present; associate editor, Lipids Journal, 1997–present; associate editor and contributing editor, *inform*, 1992–present; member, Educational Program Planning Committee, 1990–1992.

Other: Fellow, AOCS, 2009–present; AOCS Herbert Dutton Award, 2006; author and coauthor of 150+ papers for peer-reviewed journals and 30+ book chapters; co-editor of one book; inventor on 4 US Patents; member, Alternate US

Delegate to the Codex Alimentarius Committee on Fats and Oils, 2011–present; Adjunct Associate Professor, University of the Sciences in Philadelphia, 2006–present.

Research Interests: Plant lipid biochemistry, development and application of new methods for the quantitative analysis of health-promoting lipids (including phytosterols, tocopherols, tocotrienols, and carotenoids) in plant oils and foods.

Sidisky

Leonard M. Sidisky (1990): Supelco/Division of Sigma Aldrich-Gas Separation R&D Manager, Bellefonte, Pennsylvania, 2000–present.

Education: B.S., 1980, Biology, and M.S., 1997, Food Science, Pennsylvania State University.

Previous Employment: Supervisor, Capillary Manufacturing, 1997–1999; Senior Research Chemist, Capillary Manufacturing, 1990–1997; R&D Chemist, Capillary Manufacturing, 1982–1989; Capillary Chemist, Applied Science Division of Milton Roy, 1981.

AOCS Activities: Member, AOCS Governing Board, 2010–present; chairperson, AOCS Technical Steering Committee/Technical Services Value Center, 2010–present; chairperson, AOCS Chromatography Committee, 2000–2009; president, AOCS Northeast Section, 2000–2001, vice president, 1998–1999; chairman, Hans Kaunitz Award, 2000–2001; representative, Supelco/Nicholas Pelick AOCS Lipid Award, 1994–present; award winner, Ralph H. Potts Memorial Fellowship Award, 1992; AOCS Outstanding Paper Presentation, 1992; member, AOCS Analytical Division, AOCS Northeast Section 1990–present; presenter at AOCS Annual Meeting, 1984– present.

Other: Member, American Chemical Society (ACS), 1982–present; member, Institute of Food Technologists (IFT), 1992–present; ASTM D16 (Aromatic Hydrocarbons), 1990–present; chairperson, D16.OE.09 Task Group (Capillary Applications), 1995–present; ASTM E13 Committee; presented numerous papers and seminars around the world, and has published over 25 journal articles.

Research Interests: Development of gas chromatographic products for wide range of industrial applications, capillary columns for lipid sample analyses, theory and practical application of capillary gas chromatography and solid phase micro extraction (SPME) product development and applications.

Tsumadori

Masaki Tsumadori (1993): Research Fellow, Global R&D, Kao Corporation, Wakayama-shi, Japan, March 2008–present.

Education: B.S., 1975 and M.S., 1977, Polymer Chemistry, Nagoya Institute of Technology, Nagoya-shi, Aichi-ken, Japan.

Previous Employment: Vice President, Global R&D, Fabric and Home Care, Kao Corporation, 2002–2008; Director of fabric care products, laundry detergents, fabric softeners and bleaches, Kao Corporation, 1997–2002; Manager of laundry detergents/hard surface cleaners, Kao Corporation, 1987–1997; Team Leader of fabric finishers, Kao Corporation, 1982–1987; Research Chemist of laundry detergents/fabric finishers, Kao Corporation, 1977–1982.

AOCS Activities: Member-at-Large, AOCS Governing Board, 2011–present; member, Executive Committee, World Conference on Fabric and Home Care (Singapore 2012 Conference), 2011–present; member, Executive Committee, 7th World Conference on Detergents (Montreux 2010 Conference), 2009–2011; member, Asian Section, 2010–present; member, Surfactants and Detergents Division, 1999–present; chairperson, presenter, speaker, and co-author, Montreux Conferences and AOCS Annual Meeting & Expos; participant, Montreux Conference, 1986–present.

Other: Member, Program Committee for CESIO 2011 (European Committee of Organic Surfactants and Their Intermediates); member, Technical Committee, Japan Soap and Detergent Association; member, International Committee, Japan Oil Chemists' Society; director, Japan Research Association for Textile End-Uses.

Research Interests: Development and application of green surfactants/chemicals and new feedstocks. ■

Notice of Annual Business Meeting

The annual business meeting of the AOCS will be held on Monday, May 5, 2014 at 11:30 am at the Henry B. Gonzalez Convention Center, San Antonio, Texas, USA. Routine business of the Society will be conducted, including reports from the secretary and president, and new officers will be installed.

Held in conjunction with the

105th AOCS Annual Meeting & Expo

May 4–7, 2014 | San Antonio, Texas, USA

NEWS & NOTEWORTHY

FDA continues its *trans*formation of the US food supply

The question on many minds is: "Why November 7, 2013?" Why did the US Food and Drug Administration (FDA) move toward essentially banning partially hydrogenated vegetable oils (PVHO) in foods—because of the *trans* fatty acids (TFA) they contain—when the agency itself says per capita consumption of TFA has dropped from an estimated 4.6 g/d in 2003 to 1.3 g/d in 2012? Why take such a precedent-setting action when the amount of *trans* fatty acids (TFA) in the US food supply has been reduced by almost 75% since mandatory labeling of *trans* fat content in foods began on January 1, 2006?

There is no good answer. After all, the US Food and Drug Administration (FDA) keeps its own counsel. But it is possible that the precipitating factor was a lawsuit brought in August 2013 by Fred Kummerow. Kummerow, professor emeritus of the University of Illinois at Urbana-Champaign (UIUC), has been fighting for more than 50 years to remove TFA from the US food supply. At the age of 99, he is very close to winning the battle.

Long-time AOCS members remember the stirring, standing-room-only debates that took place at AOCS meetings in the mid-1970s between Kummerow and Thomas Applewhite. Applewhite (1924–2012) served as president of AOCS in 1977–1978 and was the founding editor of *Inform*. At the time he was debating Kummerow, he was director of research services for Kraft Inc.

"Although I don't recall specifics, the atmosphere was pretty charged—the AOCS equivalent of Ali vs. Frazier," recalls Frank Flider, a member of the *Inform* Editorial Advisory Committee. "What I recall is that whenever Kummerow was to speak, there was a large crowd and the anticipation that Applewhite was going to debate the issue with him."

Kummerow began researching TFA in 1957; his anti-*trans* lobbying picked up when, in 1968, he urged the American Heart Association to ask members of the Institute of Shortening and Edible Oils to reduce TFA levels in shortenings and margarines.

Between the debates of the '70s and '80s and the FDA ruling in 2013, public and regulatory opinion has gradually shifted on TFA. The research most often credited with beginning the opinion shift is a diet study of 62 university students (27 male and 35 female) conducted by Ronald P. Mensink and Martijn B. Katan at the Wageningen University in the Netherlands. The study, which appeared in 1990, was published in the *New England Journal of Medicine* (*NEJM*; doi:10.1056/NEJM199008163230703). This study reported that substitution of 1 g of dietary TFA for oleic acid increased levels of low-density lipoprotein ("bad") cholesterol by less than 1.3 mg/dL while lowering high-density lipoprotein ("good") cholesterol by less than 0.2 mg/dL.

Epidemiological research that began in the 1990s and often was led by Walter Willett of the Harvard School of Public Health (HSPH) also served to fuel the anti-*trans* fervor. One HSPH finding that has been quoted frequently by the popular press and consumer action groups came out of a meta-analysis led by Harvard's Dariush Mozaffarian and co-authored by Willett and others (*NEJM*; doi:10.1056/NEJMra054035, 2006). This work estimated that dietary industrial TFA was causing more than 50,000 US deaths annually. The fact that epidemiological studies cannot establish cause and effect, however, was seldom if ever mentioned in the popular press.

The Center for Science in the Public Interest (CSPI), a consumer advocacy group based in Washington, DC, USA, entered the fray in 1994 by calling on the FDA to require the labeling

of TFA in foods. In 2004, CSPI petitioned FDA to revoke the GRAS status of partially hydrogenated vegetable oils (PHVO). In 2009, Fred Kummerow formally petitioned FDA, beginning his 3,000-word appeal with, "I request to ban *trans* fats from the American diet." Legally, FDA had 180 days to respond; by day 1,462 (August 9, 2013), Kummerow had had enough and filed suit against the agency.

FDA'S PRELIMINARY DETERMINATION

For anyone who missed the firestorm of press coverage surrounding the November 2013 announcement, FDA said that it had made a tentative determination that PHVO—the largest dietary source of TFA—are not Generally Recognized as Safe (GRAS; see sidebar). The action has set a precedent; it is the first time FDA has acted to withdraw GRAS status from a food ingredient.

Under the preliminary ruling, industry had 60 days (or until January 7, 2014) to provide comment. The comment period later was extended by 60 days to March 8, 2014. If the determination is made final, food manufacturers will have to provide proof that PHVO are safe in order to use them in foods. Although many products have been reformulated to remove all or most TFA, a number still use PHVO for their functional properties. Such products include frozen pizzas and pies, microwave popcorn, cake frosting, some (although not all) baked goods, pancake mixes, and certain fast foods. The most immediate impacts of a ban on PVHO likely would be shorter shelf life for those products still relying on PHVO and an increase in price.

FDA officials, speaking in a news conference on November 7, made it clear that the agency had no idea what time period is feasible for industry to completely remove PHVO from processed foods. "Right now, the critical thing is to get input from industry," said Michael Taylor, the agency's deputy commissioner for foods and veterinary medicine. When asked if there was any possibility that the preliminary determination will not be made final, FDA Commissioner Margaret Hamburg said, "Life has many uncertainties, but we are on a clear track to follow the legal process."

Food manufacturers are concerned that FDA might use the same regulatory approach on other food ingredients. "I would certainly expect FDA to use the approach with respect to other ingredients that are considered safe based on industry self-assessments," Mitchell Cheeseman told *Chemical & Engineering News*. Cheeseman led FDA's Office of Food Additive Safety before joining Steptoe & Johnson, a law firm in Washington, DC.

TFA IN FOODS

TFA in foods come from two sources. Naturally occurring TFA (such as conjugated linoleic acid) are created by microorganisms in the gut of ruminant animals and so are found in the meat and dairy products from cows, sheep, and goats. Dairy products contain between 2 and 8% TFA; meat from ruminant animals contains somewhere between 2 and 11% TFA. Are natural TFA a risk for coronary heart disease (CHD)? Opinion remains divided, although a meta-analysis by scientists at the University of Copenhagen suggests that while industrial-TFA may be positively related to CHD, "ruminant-TFA are not, *but the limited number of available studies prohibits any firm conclusions . . .* " [emphasis added]. (See *European Journal of Clinical Nutrition*: doi:10.1038/ejcn.2011.34, 2011.)

Foods derived from ruminant animals accounted for about 20–25% of total TFA in the US diet based on national survey data collected in 1989–1991 (doi:10.1016/S0002-8223(99)00041-3). That figure may well be lower now because the per capita consumption of red meat has dropped. What has not changed is that the rest of the total intake of TFA comes from vegetable oil—most commonly soybean oil—that has been partially hydrogenated to make them less liquid and increase their oxidative stability, lengthening shelf life. Gary List, a consultant based in Washington, Illinois, USA, estimates the level of TFA at 1–10 wt% in PHVO currently used for salad dressings, cooking

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FDA estimates the cost of reformulation

FDA included an estimate of the cost and potential health effects of removing partially hydrogenated vegetable oils (PHVO) from the US food supply in a memo released in November 2013 as a reference to its tentative determination on PHVO.

"On the average day, at least eight Americans die as a result of the *trans* fats in partially hydrogenated oils," writes Richard Bruns, an economist with the US Food and Drug Administration (FDA), in the memo. He cites the US Centers for Disease Control and Prevention estimate that the removal of PHVO would prevent 3,000–7,000 deaths annually in the United States, as well as 10,000–20,000 nonfatal heart attacks.

Regarding the projected costs to the food industry, Bruns said that FDA estimates that more than two-thirds of dietary *trans* fats from PHVO have already been removed from the US diet. (Another estimate puts that figure at 73%. See doi:10.1080/19440049.2012.664570, 2012.) The estimates of the cost of reformulation differ depending on how much time food manufacturers are given to comply.

- If producers are given only one year to reformulate, FDA estimates a one-time cost to industry of $2.7 billion plus one-time relabeling costs of about $200 million.
- If producers have two years to reformulate, the one-time reformulation costs fall to $2.3 billion and relabeling costs to $60 million.
- If producers have three years to reformulate, the estimated reformulation costs would drop to $1.3 billion and relabeling costs to about $40 million.

For links to all references listed in the FDA's preliminary determination (the Bruns memo is No. 46), see http://tinyurl.com/trans-ref.

oils, and margarines, and up to 20 wt% in oils used for baking or frying applications.

REACTIONS TO THE PRELIMINARY RULING

The Center for Science in the Public Interest (CSPI), which petitioned FDA in 1994 to add *trans* fat to the Nutrition Facts Label, applauded the move. "Getting rid of artificial *trans* fat is one of the most important life-saving measures the FDA could take," said CSPI Executive Director Michael F. Jacobson in a statement. "Thousands of heart attack deaths will be prevented in the years ahead. The FDA deserves credit for letting science, and not politics, shape its new proposed policy on artificial *trans* fat."

Rick Cristol, executive director of the National Association of Margarine Manufacturers, a trade association based in Washington, DC, noted that all US retail branded products he is aware of have eliminated TFA or are below 0.5 g/serving.

Eric Decker, an AOCS member and professor at the University of Massachusetts, is concerned about the unintended consequences of an all-out *trans* ban. Writing for the Institute of Food Technologists, Decker noted that estimates of decreases in cardiac deaths resulting from further reductions in TFA content in foods "assume that the biological effects of TFA follow a dose-dependent linear response," minus any proof of that fact. "Therefore," said Decker, "it does not seem scientifically prudent to make a bold statement of how many deaths a food ingredient is causing without any clinical data." (See http://tinyurl.com/decker-IFT for the complete blog entry.)

Edward Emken, a past AOCS president (1999–2000) who conducted a number of studies on the biochemistry and metabolic fate of stable isotope-labeled fatty acids while at the US Department of Agriculture's National Center for Agricultural Utilization, believes the FDA action is misguided and disagrees with the largely unchallenged conclusion that dietary industrial TFA at the current dietary level of 1.3 g/d are a major public health concern.

"FDA has been bullied by consumer advocate groups that continually bombarded the public with misleading popular press articles based on pseudo-science, flawed research studies, and questionable epidemiology studies that do not prove cause and effect," Emken wrote in an email. "They selectively cite results that support their position, purposely misinterpret results, and take statements by authors out of context."

Based on his research, Emken emphasized that "the metabolism of all *trans* isomers is highly regulated, they do not accumulate in tissue lipids, they are primarily used for energy, and have a modest impact on LDL cholesterol and LDL/HDL ratios. *Trans* isomers are not poison, and *trans* isomers from ruminant sources have been part of the human diet since the Paleolithic era."

For his part, Fred Kummerow said he will continue his research (largely self-funded at this point) on the causes of heart disease. His most recent paper appeared in December 2013 in the *Scandinavian Cardiovascular Journal* ("Effects of *trans* fats on prostacyclin production," doi:10.3109/14017431.2013.856462).

When asked if he will continue with his lawsuit, he said, "As far as I'm concerned, it's a done deal." ■

Understanding the GRAS system

In 1958, the US Congress enacted the Food Additives Amendment to the Food, Drug, and Cosmetic Act of 1938. In it, FDA listed a large number of foods, substances, and additives that had been in use prior to 1958, and stated that as long as these substances are used "for the purposes indicated and in accordance with current good manufacturing practices" they would be considered GRAS (generally recognized as safe).

In FDA's words: "Under sections 201(s) and 409 of the Federal Food, Drug, and Cosmetic Act, any substance that is intentionally added to food is a food additive, [and] is subject to premarket review and approval by FDA, unless the substance is generally recognized, among qualified experts, as having been adequately shown to be safe under the conditions of its intended use, or unless the use of the substance is otherwise excluded from the definition of a food additive."

Thus, partially hydrogenated vegetable oil (PHVO) was grandfathered in as being GRAS, along with such additives as vinegar, baking powder, and many spices. Manufacturers wishing to use ingredients not on the 1958 GRAS list could petition FDA for their use, but were not required to do so. The agency would then review the filing, open a public comment period, review the comments, and issue a final regulation regarding the food additive.

In 1997, FDA—realizing that it could not handle the petition process owing to lack of staff—issued a proposed rule (that has never been made final) amending the GRAS process. Food manufacturers would still voluntarily notify the agency that a preponderance of expert opinion found their proposed new ingredient to be GRAS; FDA was no longer required to issue final rules. Under this self-affirmation process, FDA must respond within 180 days with either (i) notice that it does not object, (ii) questions about the filing, or (iii) notice that the information submitted does not, in FDA's opinion, provide a basis for a GRAS determination.

Two recent reports have criticized the GRAS system. In a February 2010 report to Congress (GAO-10-246), the Government Accountability Office determined that FDA should make the proposed rule final as well as strengthen its oversight of self-determined GRAS ingredients and systematically reconsider the safety of GRAS substances. Likewise, the Pew Health Group noted in October 2011 (see http://tinyurl.com/pew-additives) that because GRAS self-determination process is voluntary, FDA is unaware of many substances that are added to food and can't ensure that self-determinations of GRAS status were properly made.

The tentative determination in November 2013 that PHVO are not GRAS is the first time FDA has revoked the GRAS status of a food additive. Many observers feel it will not be the last.

BRIEFS

Felda Global Ventures Holdings Bhd. (Kuala Lumpur, Malaysia) announced its first-ever biodiesel shipment to Europe in November 2013. The 4,200 metric ton consignment of certified palm methyl ester went to Rotterdam, Netherlands, on its way to Switzerland. Felda is also exploring US and Chinese biodiesel markets.

■■■

A new facility, designed to run at 15 million liters per year, is recycling waste beer and soft drinks into ethanol in Brantford, Ontario, Canada. The technology employed by Energentium Inc. will create by-products such as electricity, animal feed, omega-3 oils, and CO_2. The system can also be adapted to recycle other food wastes including fruit and vegetables, jams, syrups, sugars, and candies. The process incorporates reverse osmosis. The Brantford location was chosen for its proximity to several Ontario breweries and wineries that will provide expired and bad batches of beverages to the Energentium plant.

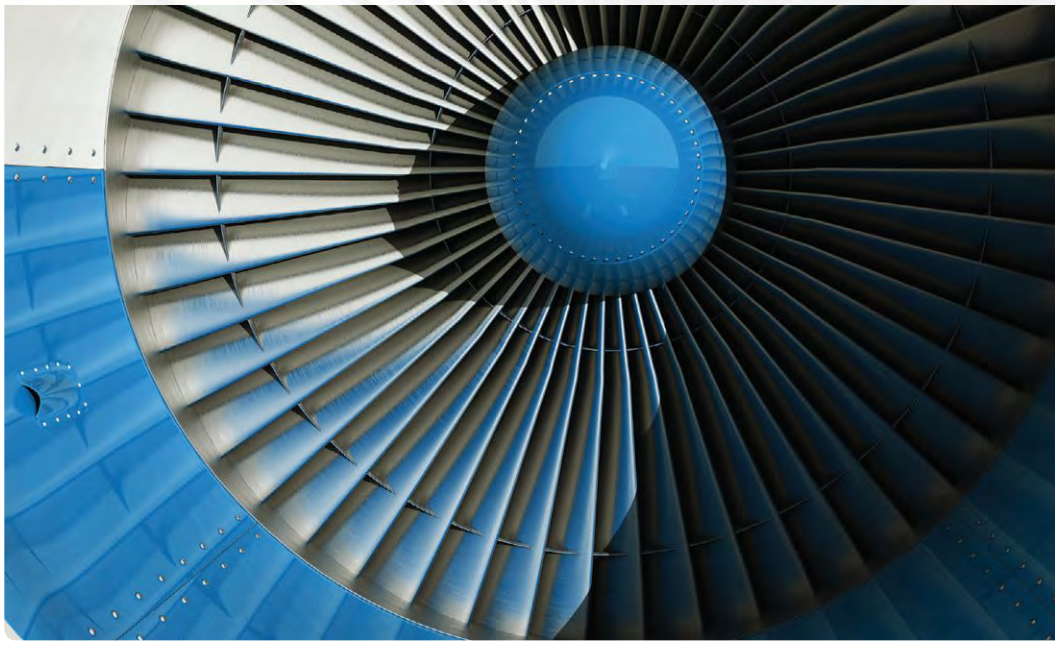
■■■

At *Inform* press time, Brazil was expected to raise its required biodiesel blend from 5% to 7%. According to Reuters.com, this action would relieve pressure on Brazil's state-run oil company, Petróleo Brasileiro SA, which had been importing diesel fuel because domestic refining was not meeting demand, thus leading to an economic loss. This 2% increase would require crushing an additional 8 million to 9 million metric tons of soybeans annually.

■■■

Algasol Renewables announced its first commercial algae project. Biodiesel Misr, the leader of the project, will grow algae in Algasol-patented photobioreactors (PBR) on up to 5 hectares of land at its facility at the First Industrial Zone in Giza, Egypt. The harvested algae will then be converted to biodiesel to meet local demand. Once the project has reached sufficient size, algal biomass for processing may also be brought in from Algasol's South African facilities (http://tinyurl.com/Algasol-Misr-algae). ■

BIOFUELS+



GE Aviation to increase biofuel purchases

Airlines have been investigating the possibility of switching at least in part to biofuels for a number of years now. But first, it must be demonstrated that the planes can safely fly on these fuels.

Starting in 2016, GE Aviation plans to buy at least 500,000 gallons (2 million liters) of biofuel every year for the next decade for use in engine testing. The company has an option to scale the order up to 10 million gallons per year as testing volumes start to peak due to its current order backlog worth $229 billion (http://www.gereports.com/the-selling-point). The fuel will be used at the test facility of GE Aviation, which has headquarters in Evendale (suburban Cincinnati), Ohio, USA.

The fuel will be made from a gasified mixture of greasewood (a perennial shrub that grows in the western half of the United States), corn cobs, and algae through a Fischer-Tropsch process. The D'Arcinoff Group Energy Program will carry out the process at a facility to be located in Hudspeth County, Texas, USA. According to D'Arcinoff's Chief Executive Officer Michael C. Darcy, "The bottom line of our process is that it very efficiently captures and converts all carbon into fuel product. The result is an ASTM-approved fuel that can be produced in large quantity with good environmental life cycle properties."

Mike Epstein, chief technologist leading the alternative fuels efforts at GE Aviation, said, "Aircraft, engines, airport fuel hydrants, even interstate pipelines have been designed and built around jet fuel." The use of biofuels could result in major reductions in greenhouse gas emissions, once prices for biofuels come down. Epstein added, "We've started the transition. We've taken the first steps.

GE Aviation has already tested biofuels made from camelina oil and from jatropha oil.

Can castor oil ever be a feedstock for commodity biodiesel?

The bean-shaped seeds of the castor plant (*Ricinus communis*) are known for their content of oil, which can be used as a fuel, a lubricant, a soap component, and in textile finishing. But the beans are also known for their content of the poisonous protein ricin and the allergen 2S albumin.

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Researchers from the University of Southern Denmark (USD; Odense) and from four institutions in Brazil have recently succeeded in using proteomics technologies to map proteins of castor beans. Their intention was that, with this information, it would be possible to alter the bean's genetic makeup, increase the beans' production of oil, and eliminate the production of ricin and 2S albumin. If this could be accomplished, the pulp remaining after oil extraction could be used as animal feed.

Their results, though, indicate that accomplishing this improvement may not be an easy task. The researchers reported mapping 1,875 castor bean proteins (*J. Proteome Res. 12*:5012–5024, 2013). Biochemist Peter Roepstorff of USD, a member of the research team, said, "Now we know where the proteins are, and we know when during bean development they are produced. Especially the protein ricin and the allergen 2S albumin are interesting in this context. Unfortunately, our research shows that it does not seem to be easy to get rid of them" (http://tinyurl.com/castor-proteome-USD).

In the early stages of their growth and development, the beans contain only low amounts of both ricin and 2S albumin. But the content of both increases as the beans mature, and the oil content increases with time as well.

Roepstorff added, "Unfortunately, the mature beans also have the highest oil content and therefore an oil producer will not harvest the beans before they are fully mature. The price of optimizing the oil production is that you also get a toxic and allergenic pulp that cannot be used for animal feed." Without a profitable use for the by-product pulp, the prospects of making castor oil a commodity product for the manufacture of fuel are limited.

At present several countries, including Brazil and India, use oil extracted from the seeds of the castor plant as feedstock for biodiesel production. The cake remaining is used for fertilizer or biofuel. Methods to extract or neutralize ricin and 2S albumin to make the cake safe as an animal feed are not presently economically feasible on a large scale.

Promising new, highly efficient biobutanol technology

Advocates for the production of biobutanol point out its potential as an additive to gasoline that could reduce its cost and provide more energy, molecule per molecule, than the more economically developed ethanol. Research coming out of Taiwan's Industrial Technology Research Institute (ITRI) has identified a new biobutanol technology that can increase carbon utilization of a typical sugar fermentation from 67% to 94%.

Under classic conditions, fermenting sugar to alcohol releases one-third of the underlying biomass in the form of CO_2. Carbon dioxide is not a particularly high-value product, however, creating an impediment on the profit that can be achieved.

ITRI claims its ButyFix technology allows yields approaching the theoretical maximum of 100% at a cost of $2 per gallon ($0.53 per liter) for its biobutanol.

ITRI's efforts were recognized by its sixth consecutive R&D 100 award (http://tinyurl.com/R-D-ITRI-BuOH), sponsored by *R&D* magazine. According to *R&D*, ButyFix produces butanol through two chemical processes: (i) hydrolytic conversion of cellulose and hemicellulose into sugar and (ii) sugar fermentation into butyrate. To convert butyrate to butanol, ITRI uses "proprietary processes to regulate certain genes in specially adapted microorganisms and redirect carbon dioxide generated during fermentation to the desired pathway where the carbon dioxide can be reutilized, leading to a high yield of butyrate" (http://tinyurl.com/RDmagazine-ITRI).

ITRI claims its ButyFix technology allows yields approaching the theoretical maximum of 100% at a cost of $2 per gallon ($0.53 per liter) for its biobutanol.

BiofuelsDigest.com (http://tinyurl.com/BFDigest-BuOH) points out cellulose is much less expensive than corn as a feedstock for alcohol production. The ButyFix technology, according to ITRI, can be "bolted on" to existing ethanol production facilities, meaning expenses to build new plants to produce butanol could be avoided or reduced. And biobutanol can be blended with gasoline at a higher rate (≥16%) than the 10–15% for ethanol in non-flex-fuel vehicles.

Furthermore, the properties of biobutanol are such that changes are not necessary in piping, storage tanks, and vehicles, unlike the characteristics of ethanol that do require these changes.

Iowa farmers harvest both corn and cobs

In the US state of Iowa, farmers, as usual, harvested corn in 2013—lots of it. The corn went to its usual destinations (food, feed, fermentation to ethanol). In northwestern Iowa some of the farmers also harvested corn stover—the cobs, leaves, husks, and stalks—to be used for production of cellulosic ethanol.

The purchaser of the stover in this region is POET-DSM Advanced Biofuels LLC, which plans to open its Project

LIBERTY facility in Emmetsburg, Iowa, in 2014. The plant has been coming up to scale gradually as the company has constructed its facility and designed its logistics. POET-DSM has already organized four previous commercial-scale harvests that have brought in nearly 200,000 tons (180,000 metric tons) of stover that were used to test operations at the facility.

In October 2013, General Manager Daron Wilson said in a company statement, "Things are going smoothly. Our advance work over the last few years on feedstock logistics is paying off." POET-DSM announced its intention in 2013 to purchase about 100,000 tons of baled stover as feedstock for startup and continuing operations through the 2014 harvest. This should allow the production of 20 million gallons (76 million liters) of ethanol annually.

Crop residue represents a new market for farmers, providing additional revenue with minimal input costs. It does not require any additional planting, and crop residue can be harvested with a standard baler. Nutrient replacement at POET-DSM's suggested rate of stover removal, approximately 1 ton per acre (2.3 metric tons per hectare), or 25% of the above-ground biomass, is minimal.

Getting rid of diesel bugs

At present, diesel fuel sold in England is B5 (5% biodiesel + 95% petrodiesel) at a minimum. Allen Blake of Garforth, near Leeds, West Yorkshire, England launched a new business this past year, which he calls Diesel Polishing (www.dieselpolishing.co.uk), to rid engines fueled with biodiesel of "diesel bugs." These are the materials that result from the growth of bacteria in water-contaminated biodiesel. Over time, the bacteria die, fall to the bottom of the tank, and accumulate to form a black "goo" that clogs fuel filters.

Blake described his service in this way: "We suck the fuel from a third of the way down the tank and take it into a filtration process. This removes anything from the fuel and then puts it back into the tank at a fast rate. When we put it back in the tank it is put to the bottom and that stirs up any contaminants." He added, "We ensure this is done at least three times and by the time we have finished all the contaminants have come out of the tank and we have 'polished' the fuel creating a much clearer, cleaner diesel tank at the same time" (http://tinyurl.com/DieselBugs).

Blake says diesel bugs are especially prevalent in boats fueled by biodiesel because they are often allowed to stand over the winter without having drained the fuel from the engine. The possibility of water condensation in the fuel is greatly increased by such practices. The availability of water allows the bacteria to grow, and the water may also contribute to formation of rust and particulates. " So when you start up your boat after a long winter of doing nothing," Blake says, "you might find that the first thing that stirs up is the bacteria, which then clog up your filter and you could be faced with quite a bill to put it right."

Cars, trucks, and farm machinery are also susceptible to diesel bugs, but because these engines are used more regularly the fuel is used more quickly, allowing less opportunity for bacteria to thrive. ■

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BRIEFS

A word to ecologically minded readers who use cloth shopping bags in place of plastic: Hugh Pennington of the University of Aberdeen (Scotland) recently told the *Daily Mail* newspaper neither meat nor vegetables covered in soil should be carried in cloth bags because bacteria can be transferred from the meat onto food that is subsequently eaten raw. He based his comments on a 2012 research paper by Jonathan Klick of the University of Pennsylvania and Joshua D. Wright of George Mason University titled "Grocery Bag Bans and Foodborne Illness" (see http://tinyurl.com/Klick-Wright). The article notes that hospitalizations and deaths from foodborne illnesses have nearly doubled in San Francisco since the city banned the use of disposable plastic shopping bags in 2007. Further, Klick and Wright found that 8% of reusable shopping bags are contaminated with *E. coli* and that 97% of people admit to never washing their reusable bags.

■■■

The use of milk-based "growing-up" formula does not bring additional value to a balanced diet in meeting the nutritional requirements of young children in the European Union, according to the European Food Safety Authority (EFSA; see http://tinyurl.com/EFSA-formula). There is "no unique role" for young-child formula (commonly called "growing-up formula") in the diet of young children (those aged 1–3), the report said.

■■■

Tufts Medical Center in Boston, Massachusetts, USA, received a grant of more than $40 million over five years from the US National Institutes of Health to investigate whether taking vitamin D3 supplements can help prevent type 2 diabetes in those at increased risk. The study will involve 2,400 patients over the age of 30 with pre-diabetes, who will randomly receive vitamin D3 (4,000 international units per day) or a placebo. See www.d2dstudy.org. ■

FOOD, HEALTH & NUTRITION

University of Granada researchers sample the subject of their research: (left to right) Jonatan R. Ruiz, Magdalena Cuenca-García, Manuel J. Castillo, and Francisco B. Ortega. Photo courtesy of the University of Granada.

Too good to be true?

Talk about a news release topic summary guaranteed to excite health news editors: "Scientists at Spain's University of Granada have disproved the old idea that chocolate is fattening." But wait, there's more: "Higher chocolate consumption was associated with lower levels of total fat—fat deposits all over the body—and central—abdominal—fat, independently of whether or not subjects were physically active, and of their diet."

The researchers report that they were intrigued by previous work suggesting that a higher frequency of chocolate intake is linked to lower body mass index (BMI) in adults (*American Journal of Clinical Nutrition*—doi:10.3945/ajcn.2008.26058, 2008). So the team decided to investigate whether higher chocolate consumption also is associated with lower BMI, as well as other markers of total and central body fat, in adolescents.

Their cross-sectional study—published in *Nutrition* (doi:10.1016/j.nut.2013.07.011, 2013)—included 1,458 European adolescents between the ages of 12 and 17 who were participants in the HELENA (Healthy Lifestyle in Europe by Nutrition in Adolescence) project. This research, financed by the European Union, studies eating habits and lifestyle in young people in nine European countries.

Results showed that a higher level of chocolate consumption was associated with lower levels of total and central fat when these were estimated through BMI, body fat percentage—measured by both skinfolds and bioelectrical impedance analysis—and waist circumference. These results were independent of the participants' sex; age; sexual maturation; total energy intake; intake of saturated fats, fruit, and vegetables; consumption of tea and coffee; and physical activity.

As lead author Magdalena Cuenca-García explains, although chocolate is considered to be a high-energy-content food—it is rich in sugars and saturated fats—"recent studies in adults suggest chocolate consumption is associated with a lower risk of cardiometabolic disorders."

In fact, chocolate is rich in flavonoids—especially catechins—that have many healthful properties: "They have important antioxidant, antithrombotic, anti-inflammatory, and antihypertensive effects and can help prevent ischemic heart disease," Cuenca-García notes.

CONTINUED ON NEXT PAGE

Another recent cross-sectional study, this one in adults, conducted by University of California researchers found that more frequent chocolate consumption also is associated with a lower BMI, Cuenca-García says (doi:10.1001/archinternmed.2011.2100, 2012). "What's more, these results were confirmed in a longitudinal study in women who followed a catechin-rich diet."

The effect, she suggests, could be partly due to the influence of catechins on cortisol production and on insulin sensitivity, both of which are related to overweight and obesity.

The University of Granada researchers sought to go further and analyze the effect of chocolate consumption in adolescence by also controlling for other factors that could influence the accumulation of fat. These factors include a large number of body measures, objective measurement of physical activity, and detailed dietary recall with two nonconsecutive 24-hour registers using image-based software.

The authors stress that the biological impact of foods should not be evaluated solely in terms of calories. "The most recent epidemiologic research focuses on studying the relation between specific foods—both for their calorie content and for their components—and the risk factors for developing chronic illnesses, including overweight and obesity," they write.

Despite their results, the authors suggest that chocolate consumption should always be deliberate. "In moderate quantities, chocolate can be good for you, as our study has shown. But, undoubtedly, excessive consumption is prejudicial. As they say: you can have too much of a good thing."

Tomato therapy: engineered variety targets intestinal lipids

University of California, Los Angeles (UCLA) researchers report that tiny amounts of a specific type of lipid in the small intestine may play a greater role than previously thought in generating the high cholesterol levels and inflammation that are thought to lead to clogged arteries.

The team also found they could reduce the negative effects of these lipids in mice by feeding the animals a new genetically engineered tomato being developed at UCLA that is designed to produce a peptide that mimics high-density lipoprotein (HDL, or "good") cholesterol.

The study, which appeared in the *Journal of Lipid Research* (doi:10.1194/jlr.M033555, 2013), focused on a group of small-intestine lipids called unsaturated lysophosphatidic acids (LPA).

"These lipids may be a new culprit that we can target in the small intestine in fighting atherosclerosis," said senior author Alan Fogelman, executive chair of the department of medicine and director of the atherosclerosis research unit at the David Geffen School of Medicine at UCLA.

Previously, it was believed that the role of the small intestine in response to a high-fat, high-cholesterol diet was simply to package the fat and cholesterol for transport to the liver. Once delivered to the liver, the large load of fat was thought to cause increased blood levels of LDL (low density lipoprotein, or "bad")-cholesterol, decreased levels of HDL (high density lipoprotein)-cholesterol, and the rise of systemic inflammation.

But that may not be the complete story. The UCLA researchers revealed that LPA, previously considered minor components because they are found in far smaller amounts in the small intestine than other lipids, may play a more direct role in contributing to the factors that cause atherosclerosis.

The scientists found that mice fed a high-cholesterol, high-fat (21%) diet showed a twofold increase in the amount of LPA in the small intestine over mice fed normal low-fat (4%) mouse chow.

When researchers added LPA at only one part per million (by weight) to the normal low-fat, low-cholesterol mouse chow, they observed the same increase in LPA in the small intestine as when the mice were fed the high-cholesterol, high-fat diet.

Surprisingly, with the addition of LPA to the low-fat diet, the UCLA team also found alterations in the patterns of gene expression in the small intestine, changes in cholesterol levels (increases in LDL and decreases in HDL), and increases in blood markers of inflammation typically seen when the mice consumed a high-fat, high-cholesterol diet.

The findings suggest that some of the factors leading to atherosclerosis occur in the small intestine and not just the liver. Targeting LPA in the small intestine may be a way to help stop changes in blood cholesterol and inflammation before the load of packaged fat even reaches the liver, the researchers said.

"Recognizing the importance of these minor lipids in the small intestine may lead to ways to reduce their levels and prevent abnormalities in blood levels of 'good' and 'bad' cholesterol . . . ," Fogelman said.

The next step was to test the impact of the genetically engineered tomatoes on reducing the effects of these lipids in the small intestine. The tomatoes, created at UCLA, produce a small peptide called 6F that mimics the action of apoA-1, the chief protein in HDL.

Researchers added 2.2% (by weight) of freeze-dried tomato powder from the peptide-enhanced tomatoes to low-fat, low-cholesterol mouse chow that was supplemented with LPA. They also added the same dose of the peptide-enhanced tomatoes to the high-fat high- cholesterol diet.

They found that this addition to both diets prevented an increase in the level of LPA in the small intestine and also stopped increases in LDL-cholesterol, decreases in HDL-cholesterol, and systemic inflammation. Tomatoes that did not contain the peptide had no effect.

According to Fogelman, the peptide-enhanced tomatoes may work in large part by reducing the amount of the LPA in the small intestine. ■

BRIEFS

In the fourth quarter of 2013, biotech seed company Monsanto (St. Louis, Missouri, USA) and plant genomics company Evogene Ltd. (Rehovot, Israel) extended their existing research and development collaboration agreement to August 2016. The goal of the collaboration, which began in 2008 and was extended once before in 2011, is to find key genes in soybean, cotton, canola, and corn related to yield, environmental stress, and fertilizer utilization. An added five-year goal will be to identify genes providing resistance to the *Fusarium*-caused stalk rot disease in corn.

■■■

DuPont announced in October 2013 that it was spinning off its Performance Chemicals segment. The separation should be complete in about 18 months, at which time 100% of the new public entity will be owned by DuPont shareholders. The spinoff occurred, according to a company statement, so that the original company can advance "its unique integrated capabilities in biology, chemistry, and materials science to further strengthen its industry-leading positions in agriculture and nutrition, bio-based industrials, and advanced materials" (http://tinyurl.com/DuPont-spinoff).

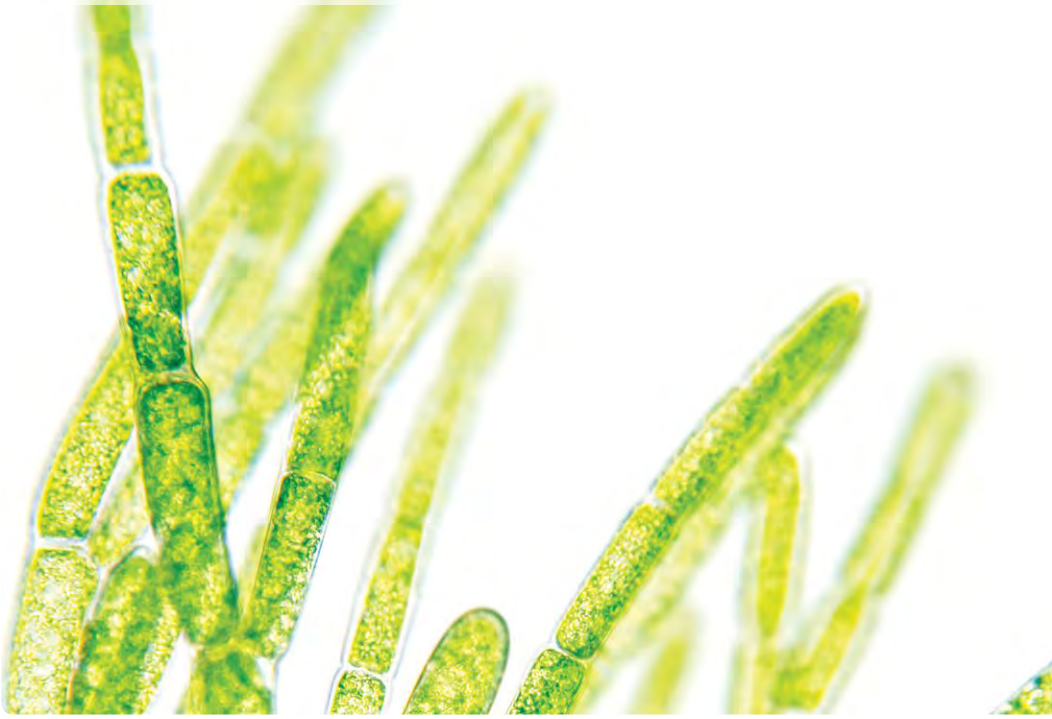
■■■

According to the *Hindustan Times* newspaper (November 3, 2013), Bangladesh has approved planting and cultivation of four varieties of brinjal (also called eggplant) genetically modified to produce *Bacillus thuringiensis* (Bt) protein (http://tinyurl.com/brinjal-India-Bangladesh). India is concerned that seeds of these Bangladeshi plants could be carried across the India-Bangladesh border into West Bengal, India's largest brinjal-growing state.

Although the Genetic Engineering Appraisal Committee, India's biotech regulator, previously approved Bt brinjal, the environment minister in 2009 said there was no "overriding urgency to approve" GM brinjal, and it was put under an "indefinite moratorium" that year.

The concern is that Bt brinjal could displace several traditional varieties in India, the center of origin of the vegetable. ■

BIOTECHNOLOGY



Stopping the clock on algae

Research out of Vanderbilt University (Nashville, Tennessee, USA), the Craig Venter Institute (Rockville, Maryland, USA), and Waseda University (Tokyo, Japan) has found that tricking the biological clock of cyanobacteria—also known as blue-green algae—to remain in the daytime setting can considerably raise the amount of valuable compounds these plants can produce when they are grown in constant light.

Earlier work had established that the clock mechanism in cyanobacteria is based on three proteins, KaiA, KaiB, and KaiC. In the current study, the researchers discovered that KaiA and KaiC act as switches that turn the cell's daytime and nighttime genes on and off. When KaiA is upregulated and KaiC is downregulated, then the 95% of the cell's genes that are active during daylight are turned on, and the 5% of the cells genes that operate during the night are turned off. And when KaiC is upregulated and KaiA is downregulated, then the day genes are turned off and the night genes are turned on.

According to corresponding author Carl H. Johnson (Vanderbilt), "All we have to do to lock the biological clock into its daylight configuration is to genetically upregulate the expression of KaiA, which is a simple manipulation in the genetically malleable cyanobacteria" (http://tinyurl.com/algae-clock).

To test the effects this capability has on the algae's ability to produce commercially important compounds, the researchers inserted a gene for human insulin in some of the cyanobacteria cells, a gene for the fluorescent protein luciferase in a second set of cells, and a gene for hydrogenase in yet others. They found that cells with locked clocks produced 200% more hydrogenase, 500% more insulin, and 700% more luciferase when grown in constant light than they did when the genes were inserted in cells with normally functioning clocks.

If bioclock stopping lives up to its promise, it could have economic benefits. Algae are already used to produce commercial quantities of anticancer drugs, cosmetics, bioplastics, biofuels, and nutraceutricals.

For further information, see Y. Xu, P.D. Weyman, M. Umetani, J. Xiong, X. Qin, Q.

CONTINUED ON NEXT PAGE

[C]ells with locked clocks produced 200% more hydrogenase, 500% more insulin, and 700% more luciferase when grown in constant light . . .

Hu, H. Iwasaki, and C.H. Johnson, Circadian yin-yang regulation and its manipulation to globally reprogram gene expression, *Current Biology,* DOI: 10.1016/j.cub.2013.10.011.

Do GMOs lead to superweeds?

In the continuing discussion about the pros and cons of planting crops genetically modified to be resistant to the application of herbicides, there is debate regarding whether the use of genetically modified organisms (GMOs) creates new "superweeds" that are resistant to herbicides. A case in point in the United States is the growth and expansion of a type of pigweed called Palmer amaranth (*Amaranthus palmeri*).

In 2004, Palmer amaranth that was resistant to the herbicide glyphosate (Roundup) used on cotton crops was found in a single county in the state of Georgia (http://tinyurl.com/NC-pigweed). By 2011, it had spread to 76 counties, or nearly every agronomic county in the state. (http://tinyurl.com/GA-pigweed).

Palmer amaranth is especially noxious because of its growth rate. Under optimal conditions, it can add several inches a day, and a single female plant can produce up to one million seeds. The small seeds easily move within and between fields in a number of ways, including harvesting and tillage equipment.

In comparison with soybeans, Palmer amaranth can produce more and longer roots, and amaranth roots are better at penetrating compacted soils. According to Aaron Hager, professor of weed sciences with the University of Illinois (Urbana-Champaign, USA), in fields infested with Palmer amaranth soybean yield losses approaching 80% and corn yield losses exceeding 90% have been reported in peer-reviewed scientific literature (http://tinyurl.com/pigweed-IL).

It is fair to ask whether the introduction of GMOs caused the explosion of weeds, as typified by Palmer amaranth. Keith Edmisten, professor of crop science at North Carolina State University (Raleigh, USA), points out that the lack of chemical rotation is the culprit, not the GMOs themselves. That is, farmers have depended too much on one herbicide to control weeds, without rotating from one herbicide to another over the course of several years.

"The resistance was in the Palmer amaranth population," Edmisten told the NCSU student newspaper *Technician Online*. "We selected for [the gene] by using Roundup. . . . Roundup was used on multiple crops, often with multiple applications, so we selected for the resistant individuals" (http://tinyurl.com/NC-pigweed).

Edmisten added that if any crop is continually exposed to a single herbicide, the crop will eventually become resistant to that herbicide

Voters in Washington state reject GM labeling

Citizens of the state of Washington (USA) voted on November 6, 2013, against Proposition 522, which would have required labeling of foods containing genetically modified (GM) ingredients. The final tally was 55% opposed, 45% in favor.

Biotech seed companies Monsanto, DuPont Pioneer, Dow AgroSciences, and Bayer CropScience contributed $11 million to the campaign against the proposition, and the food industry lobbying group Grocery Manufacturers Association (GMA) contributed another $11 million. Less than $1,000 was raised for the No campaign within the state of Washington.

The largest contributors to the campaign for the bill were the Center for Food Safety (Washington, DC), and California-based organic soap maker Dr. Bronner's Magic Soaps. A total of almost $7.9 million, was raised, almost a third of that coming from Washington residents.

Proponents for labeling argued that consumers should know what is in their food. GMA Spokesman Brian Kennedy told *USA Today* newspaper that passage of the proposition would lead to increased food prices, and labels would lead consumers to think that GM ingredients are "somehow different, unsafe, or unhealthy."

Proposition 37, which was similar to Washington's Proposition 522, was voted down in California in 2012. The states of Maine and Connecticut passed labeling laws in 2013, but these will not come into effect unless and until neighboring states pass similar labeling laws.

According to 2013 figures from the US Department of Agriculture, 97% of US sugar beets, 93% of soybeans, 90% of cotton, and 90% of feed corn for animals are GM. About 60% of the US papaya crop, which is entirely grown in Hawaii, is also GM. The state's main island recently passed a law banning the production or cultivation of GM crops, excluding papaya (see *Inform* 25:36, 2013).

The US Food and Drug Administration does not require GM labeling because it considers these ingredients to be functionally equivalent to conventionally grown crops.

P&G recognizes Novozymes

On October 28, 2013, for the sixth year in a row, Procter & Gamble (P&G) named Novozymes as one of its 15 External Business Partners of the Year. P&G has more than 82,000 suppliers and agencies. Novozymes' main collaboration with P&G is in developing and selling enzymes for use in detergents.

For the first nine months of 2013, Novozymes sales to the household care industry increased by 5%, as compared to the first nine months of 2012. Sales growth was identified as being related to customer demand to improve wash performance, enable low-temperature washing, and replace chemicals with enzymes.

GM around the world

Mexico. A Mexican judge suspended further approval of permits to plant or sell genetically modified (GM) corn within the country's borders effective October 10, 2013. The action was intended to give various lawsuits brought by citizens, farmers, scientists, and other concerned parties the needed time to work their way through the judicial system. Judge Jaime Eduardo Verdugo J. (http://tinyurl.com/Mexico-bans), who promulgated the decision, wrote that GM corn posed "the risk of imminent harm to the environment."

The ruling was the result of a class action lawsuit filed on July 5, 2013, by activist groups such as the Slow Food Youth Network and Sin Maíz No Hay País (No Corn No Country).

According to Bloomberg Businessweek (http://tinyurl.com/GM-Brazil-Mexico), growing GM corn for consumption has been illegal in Mexico since 1998. The decision means no one can grow GM corn in the country for any reason, not even for a strictly export market.

Businessweek also contends that corn, having originated in Mexico, is part of the nation's sense of identity. Mexicans feel it is part of who they are to protect their landraces of corn (i.e., the many distinctive local varietals). Thus, the question of genetic "contamination" of corn, which is integral to genetic engineering, is considered a serious matter. There is less emotion attached to genetic modification of crops lacking corn's special position in Mexican society. For instance, Mexico allows commercial cultivation of GM soybeans and cotton.

European Union (EU). DuPont Pioneer (Johnston, Iowa, USA) applied to grow its maize 1507 variety in Europe back in 2001. The variety, genetically altered to produce its own insecticides against the European corn borer, has received six positive safety opinions from the European Food Safety Authority since 2005, but divisions within the EU have kept the crop in limbo.

In response to a September ruling by the Court of Justice of the European Union to act on the issue, the European Commission (EC) proposed on November 6, 2013, to have the Council of Ministers, a collection of representatives from each of the 28 EU member states, decide whether to issue the approval. If the 28 representatives in the Council cannot collect a weighted majority to make a decision, the EC has the right to approve the crop and says it will do so.

Previously, only one GM crop had been commercially grown in Europe: Monsanto's MON810, which has also been modified for resistance to the European corn borer pest. BASF's GM Amflora potato variety, which produces amylopectin starch in the tuber but not amylose, has been approved in the EU since 2010, but was withdrawn in 2012 due to lack of demand.

Although acceptance of Pioneer's maize 1507 for planting in Europe has been slow coming, it already can be sold as food and animal fodder, according to the publication Global Milling (http://tinyurl.com/GlobalMilling-1507).

Brazil. EMPRAPA, the national agricultural research and development institute of Brazil, has spent R$3.5 billion ($1.6 billion) over the past decade to develop a GM bean (*Phaseolus vulgaris*) that is resistant to the golden mosaic virus, one of the most damaging diseases to the crop in South America (http://tinyurl.com/bean-GM-Brazil). The disease is spread by the whitefly, and leads to losses of this staple in the Brazilian diet of up to 280,000 metric tons of beans a year, or 8% of the average annual production.

EMPRAPA used RNA interference to modify the bean so that it can no longer produce a protein that the virus needs to replicate. The new bean will be available to the public by 2015.

Brazil has adopted other GM crops as well. Since regulatory approval in 2003, GM soybeans have grown to constitute about 85% of Brazil's crop. The country is now the second-largest exporter of soybeans, behind the United States.

The Centro de Tecnologia Canavieira (CTC; Piracicaba, São Paulo, Brazil), the world's largest sugarcane technology center, has been working since 1994 to genetically engineer cane to increase sugar production. CTC expects to win approval for its first GM variety within about four years (http://tinyurl.com/GM-sugarcane). Development of these new varieties has been slow owing to the complexity of sugarcane's DNA. The first GM varieties to be introduced will likely provide higher resistance to a particular disease. Although conventional cross-breeding can achieve 10–15% more sugar, CTC's goal with genetic engineering is to develop plants producing 30% sugar or even more above current yields. ■

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AOCS Code of Ethics • Chemistry and its application by scientists, engineers, and technologists have for their prime objective the advancement of science and benefit of mankind. Accordingly, the Society expects each member: 1) to be familiar with the purpose and objectives of the Society as expressed in its articles of incorporation; to promote its aim actively; and to strive for self-improvement in said member's profession; 2) to present conduct that at all times reflects dignity upon the profession of chemistry and engineering; 3) to use every honorable means to elevate the standards of the profession and extend its sphere of usefulness; 4) to keep inviolate any confidence that may be entrusted to said member in such member's professional capacity; 5) to refuse participation in questionable enterprises and to refuse to engage in any occupation that is contrary to law or the public welfare; 6) to guard against unwarranted insinuations that reflect upon the character or integrity of other chemists and engineers.

Tata Strategic Management Group (Mumbai and Delhi, India) recently released a report on the market for oleochemicals (see http://tinyurl.com/Tata-report). According to the report, the global oleochemicals market reached sales of 14 million metric tons (MMT) in 2013 and is expected to grow at 6% per year over the next five years to reach 18 MMT. The Asia-Pacific region accounts for 68% of the market, which is expected to grow at 8% per year over the next five years. Surfactants, home care, and personal care are the largest end-use segments.

■■■

Authors of a new market forensics analysis using purchased market sales data in *Environmental Science: Processes & Impacts* (doi:10.1039/C3EM00418J, 2013) note that fatty alcohol-based surfactants are widely used in detergents and personal care products; such products are typically disposed of down the drain and their active ingredients are degraded or removed during wastewater treatment. The authors found that the per capita usage rate for fatty alcohols in 4000+ top-selling products was 4.9 g per day, with 88% coming from liquid laundry detergents and hand dish detergents. This extrapolates to a national usage of 185,000 metric tons of fatty alcohols per year in the United States.

■■■

Australia's National Industrial Chemicals Notification and Assessment Scheme has proposed to ban the use of dibutyl phthalate (DBP) in cosmetics, according to the *Chemical Watch* (CW) news service. The proposal comes after the agency's final assessment of the chemical as part of its Priority Existing Chemical reports on phthalates. DBP is widely used in cosmetics as a solvent, fixative, suspension agent, lubricant, antifoamer, skin emollient, and also as plasticizer in nail polish and fingernail elongator, CW said. The report on DBP proposes it be placed in Appendix C of the Standard for the Uniform Scheduling of Poisons, a list of substances considered sufficiently dangerous to health that their sale, supply, and use are prohibited. ■

HOME & PERSONAL CARE

Disruptive technology? Walmart's "green" product line may signal a big change

Wal-Mart Stores Inc. (Walmart; Bentonville, Arkansas, USA) has introduced a line of four "all-natural" household cleaning products under its Great Value private-label brand in 2,000 US stores. That brand, which debuted in 1993, includes hundreds of household consumer products. Is the company simply "greenwashing" or has it recognized that the technology behind the new formulation promises a big change in the green cleaning products arena?

On the face of it, the November 2013 product introduction of "100% chemical-free" (in Walmart's words) Great Value Naturals cleaning products seems straightforward: The company, which is by far the world's largest retailer, has since 2009 been assessing companies and products along its supply chain using a 15-question Sustainability Index. In addition, Walmart is a founding member of the Sustainability Consortium (www.sustainabilityconsortium.org)—a worldwide coalition of participants from industry, government, and academia "that work collaboratively to build a scientific foundation that drives innovation to improve consumer product sustainability."

Sustainability aside, there is profit to be had in the green category. Global spending on green cleaning products is expected to reach $9.3 billion in 2017 from an estimated $2.7 billion in 2012, according to Global Industry Analysts Inc. (GIA; San Jose, California, USA). That growth trajectory is still only a fraction of the relatively steady $150 billion market for all household cleaning products, GIA noted, but it is still an attractive niche market for manufacturers and retailers.

WHO BUYS GREEN PRODUCTS AND WHY?

The Shelton Group is a marketing communications agency based in Knoxville, Tennessee, USA. The company focuses entirely on energy efficiency and sustainability and conducts an annual Eco Pulse™ survey of US public opinion about green products and preferences. The 2013 report turned up a number of interesting facts that go a long way toward explaining the relatively slow growth of green consumer product sales.

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TABLE 1. Percentage of those "seriously searching" for green products by category and year[a]

	% searching		
Category	2011	2012	2013
Light bulbs	N/A	70.8	67.9
Home cleaning products	66.6	60.7	65.1
Paper products	65.7	58.0	62.7
Food and beverages (net)	54.6	46.5	58.9
Laundry and dishwashing products	64.8	59.2	55.1
Food	N/A	42.0	52.9
Personal care products	53.9	46.0	46.6

[a]Shelton Group, Inc., Eco Pulse 2013.

The percentage of respondents searching for greener products climbed (from 59.9% in 2009 to 70.2% in 2012) and then leveled off (65.5% in 2013), Shelton says (see Table 1). "It is likely that we have simply reached the maximum potential market for green products," the report notes. Demographically, the Shelton data show that minorities were more likely to be searching for greener products than were nonminorities (73% to 61%). Householders with children were also more likely to be searching than were householders without children (72% vs. 62%). Respondents 65 years old or older were the least likely age group to be searching (55% vs. 68% of those 64 and younger).

Shelton has also tracked increasing concern by consumers about the chemical content in a variety of nonfood products. This concern "varies by category and is generally vague and uninformed," the report says. For example, although more than 50% of those polled claim to be searching for more natural alternatives within the personal care category, "only 18% say they are regularly buying them—and this number hasn't changed in three years." Even respondents who are worried about this issue are mostly unable to identify chemicals (from a list) that they feel they should be concerned about.

The Eco Pulse study found that the only product category for which there is a broad, mainstream awareness and concern about chemical content is household cleaners. "We think it is not just coincidence that this is also the category with the highest green product market penetration among consumer packaged goods," the report's authors write.

FOUR PRODUCTS EQUALLY AS EFFECTIVE AS PETROLEUM-BASED PRODUCTS

"Great Value Naturals cleaning products are made from plant-based, all-natural Evolve® cleaning technology and are sold exclusively at Walmart," the company news release stated. "The products are 100% chemical-free [by which the company means "petrochemical-free"] and toxin-free, biodegradable, nonallergenic, and packaged in recycled materials."

The products are priced below well-established brands such as Clorox Co.'s Green Works, the Method cleaning product line, or Ecover (which purchased Method in 2013).

The Walmart line includes:

- Great Value Naturals liquid laundry detergent: $8.97 for a 100-fluid-ounce (3-liter) bottle (64 loads)
- Great Value Naturals multi-surface cleaner: $2.47 for a 32-fluid-ounce sprayer
- Great Value Naturals glass and window cleaner: $1.98 for a 32-fluid-ounce sprayer
- Great Value Naturals automatic dishwasher gel: $3.97 for a 75-fluid-ounce bottle

Evolve, according to Walmart, "is the first all-natural cleaning technology that is lab-tested to achieve results at levels of efficacy previously seen only through the use of petroleum-based cleaners, detergents, degreasers, and solvents."

TRUTH OR HYPE?

Benjamin Shell, CEO of Agaia, the Fort Lauderdale, Florida, USA-based company that developed the Evolve technology and has only been in business since 2011, remembers his approach to Walmart with both clarity and pleasure.

"We sent samples on a Friday in early 2012 and heard back from them the following Monday," recalls Shell. Walmart is not alone in exhibiting enthusiasm about the performance of the Evolve technology. Shell reports that Agaia is in the process of picking up a number of major industrial cleaning clients that he is not at liberty to name and is licensing Evolve for private branding to a "growing list of Fortune 100 and 500 companies." Agaia distributes its products in North, Central, and South America; Asia, and Europe; those products include a full range of industrial and institutional (I&I) cleaning and laundry solutions and will soon include consumer products.

On its website, Agaia suggests that its I&I products can reduce operating costs up to 30% through a reduction in labor costs as well as usage of water and electricity. The laundry detergent is effective at 110–120°F (43–49°C), Shell says; further, Evolve reduces rinsing by 60% or more, saving both water and electricity.

AOCS member Jeffrey Harwell, a professor at the University of Oklahoma, worked as a consultant to Benjamin Shell's brother, Christopher Shell, a chemical and petroleum engineer by training, on the development of the Evolve technology. The

The Evolve technology "likely exhibits a significantly different cleaning mechanism than traditional cleaners and laundry detergents."

brothers were introduced to Harwell through Candida West of the US Environmental Protection Agency, who had co-authored an article on surfactants and subsurface remediation with Harwell.

Although Harwell is bound by the terms of a nondisclosure agreement, he says that the Evolve technology "likely exhibits a significantly different cleaning mechanism than traditional cleaners and laundry detergents." Benjamin Shell suggests that the mechanism involves hydrogen bonding rather than covalent chemistry.

As part of the development process, Harwell says he "threw out a crazy suggestion" concerning "a surprising formulation component" and also tweaked the pH of the formulation. "We had used the component as part of our work on surfactants in environmental remediation," he added. "Ironically, we never actually employed it in that arena."

Shell says that efficacy testing conducted by Phillips & Associates (Minneapolis, Minnesota, USA) has shown that the Evolve technology cleans better than petroleum-based formulations. In a commercial laundry setting, the base Evolve technology (Evolve 203L) remediated 100% of 14 of 16 different standard stains and 40–50% of the final two (lipstick and black shoe polish). Phillips has called it "the most powerful technology" the company had ever tested, according to Agaia.

FORMULATION DETAILS

The Material Safety Data Sheet (MSDS) on the Agaia website (www.agaia.com) and the patent filing (US8455426, granted June 4, 2013) give some tantalizing clues about the new formulation, which is 100% renewable, including the use of essential oil-based fragrance rather than synthetic perfumes.

The patent describes a liquid cleaning composition containing alkyl polyglycoside (APG) surfactants, an alkylated vegetable oil, water, and a sodium-containing base. APG are nonionic surfactants derived from sugars and fatty alcohols; the MSDS for the undiluted formulation describes the product as being amphiphilic in its action. The MSDS also asserts that the formulation "contains no chemical, petroleum or synthetic ingredients or catalysts." It is "manufactured from edible food stock oils where possible." The composition is identified as being 72% "proprietary plant-based surfactants"; 8% palm, soy, coconut oils; 10% glucose/dextrose; 4% natural fatty acids; and 6% GRAS-exempt ingredients.

Industry observers have questioned the "all-natural" claim made by both Walmart and Agaia (having deemed the "chemical-free" claim as patently absurd). Agaia, for its part, points to the use of plant-based feedstock and catalysts in the production of the APG as the basis of its "all-natural" claim. Shell also noted that Agaia has relied on "a certification by Oklahoma University, who reviewed all ingredients and their MSDS, as well as manufacturing methods, to determine our all-natural claims."

"The notion that you can perform a chemical reaction with so-called 'natural' ingredients and say that the resulting product is neither chemical nor synthetic is ridiculous," said Warren Schmidt, a surfactant consultant based in Cincinnati, Ohio, USA, and a member of the *Inform* Editorial Advisory Committee. "Further, there is no such thing as a 'natural fatty alcohol,' as the alcohol is made by the catalytic hydrogenation of fatty methyl esters."

This difference of opinion points to a fact of product marketing, whether of food or home/textile care: The word "natural" is, essentially, meaningless. In the United States, the Federal Trade Commission (FTC) is charged with administering consumer protection legislation in pursuit of free and fair competition in the marketplace. The FTC abandoned the effort to regulate "natural" claims as early as 1978, deciding instead to scrutinize claims "on a case-by-case basis" (*Federal Register 48*:23270, May 24, 1983). In 2010, the agency declined to define "natural" in its Proposed Revision to its *Guides for the Use of Environmental Marketing Claims*. FTC noted that no commenters had provided data on how consumers understand the use of the term and that "natural" conveys different meanings depending on the context. Likewise, the US Food and Drug Administration (FDA) has declined to define "natural" with respect to food, food ingredients, drugs, and cosmetics.

A number of class-action lawsuits have been filed challenging "natural" claims, where products contain synthetic ingredients such as dipropylene glycol, propylene glycol, triclosan, and tetrasodium ethylenediamine tetraacetic acid (in a "natural" deodorant—see Trewin v. Church & Dwight, Inc., filed in March 2012). The lawsuits have led to a renewed effort within industry to set third-party natural standards such as the Natural Products Association Home Care Standard, which was introduced in February 2010 (see http://tinyurl.com/NPA-HC).

FUTURE PLANS

Benjamin Shell says Agaia plans to bring the Evolve brand to the retail consumer market eventually. Toward that end, Doug Meyer—former president/CEO of Burt's Bees—has been hired to head the Agaia Home Division. "Lots of financial partners" are in the works for 2014, Shell says, adding that he plans to "continue to demonstrate to the world that there are all-natural solutions for removing toxic chemicals from the home." ■

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trans *Fats Alternatives*

Edited by Dharma R. Kodali and Gary R. List. Product code: 213.

Scientists around the world are working to create solutions or alternatives to *trans* fats. *Trans Fats Alternatives* is an indispensable guide for everyone who is interested in *trans* fats—from food product manufacturers who provide the *trans* fat solutions to the researchers who would like to create innovative solutions.

trans *Fats in Foods*

Edited by Gary R. List, Nimal Ratnayake, and David Kritchevsky. Product code: 217.

Now more than ever *trans* fatty acids are an issue for the edible oil industry, food processors, health professionals, and the general public. This book brings together relevant information on the effect of *trans* fatty acids on health and nutrition, labeling, analysis, and food products; and is a great resource for food technologists and dietitians.

Official Methods for the Determination of trans *Fat*, 2nd Edition

Edited by Magdi M. Mossoba and John K.G. Kramer. Product code: 247.

This monograph describes the most common gas chromatographic and infrared spectroscopic official methods required for the determination of *trans* fatty acids for food labeling purposes.

Edible Oleogels: Structure and Health Implications

Edited by Alejandro Marangoni and Nissim Garti. Product code: 258.

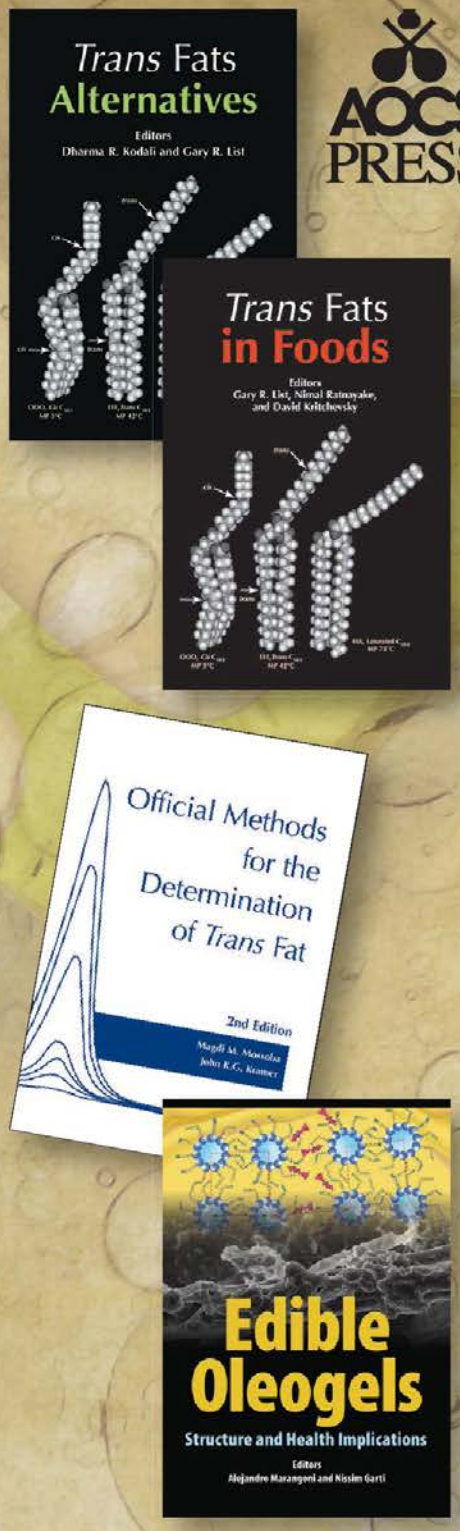
In an effort to provide alternatives to *trans* and saturated fats, scientists have been busy modifying the physical properties of oils to resemble those of fats. These new oil-based materials are referred to as oil gels, or "oleogels," and this emerging technology is the focus of many scientific investigations geared toward helping decrease the incidence of obesity and cardiovascular disease.

Available early 2014

trans *Fats Replacement Solutions*

Edited by Dharma Kodali. Product code: 271.

This book offers a comprehensive understanding of the *trans* fats chemistry, labeling regulations, and *trans* fat replacement technologies. It is an extensive resource on worldwide trends in *trans* fat replacement solutions and regulations.



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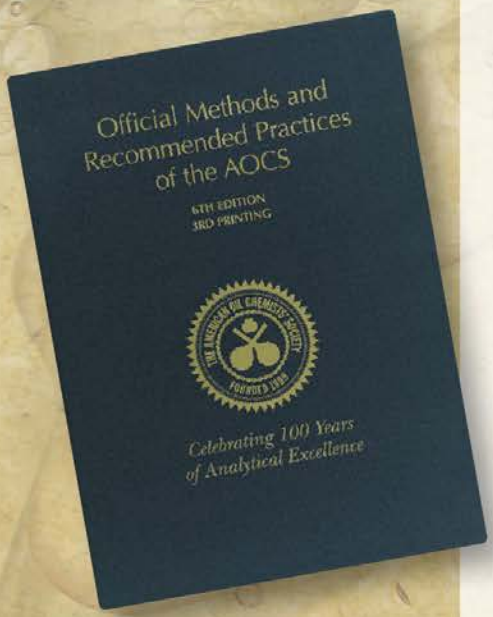
  Determination of *cis-*, *trans-*, Saturated, Monounsaturated and Polyunsaturated Fatty Acids in Extracted Fats by Capillary GLC. The FAME of extracted fat samples are separated according to chain length, degree of unsaturation, and geometry and position of double bonds by capillary GLC with a highly polar stationary phase.
- **AOCS Official Method Ce 2b-11**
  Direct Methylation of Lipids in Foods by Alkali Hydrolysis. FAME directly from food matrices are prepared with simultaneous alkali hydrolysis and methylation without prior digestion. Incorporation of triacylglycerol (TAG) standards allows the quantification of total fat and fatty acids using Theoretical Correction Factors (TCFs) and Empirical Correction Factors (ECFs).
- **AOCS Official Method Ce 2c-11**
  Direct Methylation of Lipids in Foods by Acid-Alkali Hydrolysis. FAME directly from food matrices are prepared by *in situ* acid digestion followed by alkali hydrolysis and methylation.



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From AOCS Meetings

105th AOCS Annual Meeting & Expo | May 4–7, 2014 | San Antonio, Texas, USA

Emerging Topics Symposia | Monday, May 5, 2014 | 8:30–11:00 am

This year's Emerging Topics Symposia will feature a special session on *trans* fats and will expand beyond the science to address how this current critical issue is impacting the business of fats and oils.

To view full descriptions, visit AnnualMeeting.aocs.org/EmergingTopics.

PEOPLE/INSIDE AOCS

Sköld promoted

Sköld

The new director of Oilseeds and Grains Worldwide at Solex Thermal Science Inc. (Calgary, Alberta, Canada) is **Farah Sköld**, P.Eng. This is a promotion within the company. Previously, Sköld's title was vice president, Product Development. She has been with Solex for five and a half years.

Solex is a global supplier of heater, cooler, and dryer technologies designed specifically for use with free-flowing bulk solids, such as sugar, fertilizers, oilseeds, chemicals, plastics, biosolids, minerals, and many other types of grains, crystals, and powders.

AGROW presents awards

AGROW, publisher of AGROW World Crop Protection News, presented 15 awards on October 28, 2013, in Amsterdam (Netherlands) to companies involved in crop protection. **EPL Bio Analytical Services** (Niantic, Illinois, USA), a corporate Silver member of AOCS, was recognized for best supporting role. EPL had to meet criteria for client support, service provision, and a willingness to "go the extra mile" for its customers.

EPL is a contract analytical laboratory that provides services exclusively customized for the agricultural industry. It provides data that have been used to support hundreds of crop protection products, GM crops, organic crop farming, and biopesticides.

White named AACCI fellow, Singh recognized

White

AOCS Past President **Pamela J. White**, who is a professor in the Department of Food Science and Human Nutrition at Iowa State University (Ames, USA), was named a Fellow of AACC International at its 2013 Annual Meeting held September 29–October 2 in Albuquerque, New Mexico (USA). Her selection was based on distinguished contributions to the field of cereal science and technology in research, industrial achievement, leadership, education, administration, communication, or regulatory affairs.

At the same meeting, **Vijay Singh** was presented the Excellence in Teaching Award. Singh is in the Department of Agricultural and Biological Engineering and is associate director of the Center for Advanced BioEnergy Research at the University of Illinois (Urbana-Champaign, USA). Singh is an associate editor for the *Journal of the American Oil Chemists' Society*.

New position for palm oil scientist

T. Tiger Tangavelu, formerly with California Oils Corp., has joined Global Agri-trade Corporation (GATC) as its technical service director. GATC is a major supplier of palm-based animal nutrition ingredients and bakery shortenings and margarines in North America. Tiger is based in Long Beach, California, USA.

He may be contacted by telephone at +1 562-320-8550 or by email at ttangavelu@globalagritrade.com. ■

AOCS MEETING WATCH

May 4–7, 2014. 105th AOCS Annual Meeting & Expo, Henry B. Gonzalez Convention Center, San Antonio, Texas, USA. http://annualmeeting.aocs.org

October 6–9, 2014. World Conference on Fabric and Home Care—Montreux 2014, Montreux Music & Convention Centre, Montreux, Switzerland. http://montreux.aocs.org

May 3–6, 2015. 106th AOCS Annual Meeting & Industry Showcase. Rosen Shingle Creek, Orlando, Florida, USA

For in-depth details on these and other upcoming meetings, visit http://aocs.org/meetings or contact the AOCS Meetings Department (email: meetings@aocs.org; phone: +1 217-693-4821; fax: +1 217-693-4865).

Also, be sure to visit AOCS' online listing of industry events and meetings at http://tinyurl.com/industry-calendar. Sponsoring organizations can submit information about their events to the web-based calendar by clicking a link and completing a web form. Submission is free. No third-party submissions, please. If you have any questions or comments, please contact Valorie Deichman at valoried@aocs.org.

PATENTS

Enzymatic oil recuperation process

Kellens, M., and W. De Greyt, N.V. Desmet Ballestra Engineering SA, US8435766, May 7, 2013

A process for the recuperation of acylglycerols or acylglycerols containing free fatty acids from gums, present as aqueous emulsions, said gums being obtained by subjecting triglyceride oil to one or more degumming processes, said recuperation process comprising the steps of: subjecting said gums to enzymatic hydrolysis catalyzed by one or more enzymes with phospholipase activity; allowing said gums to separate into two or more phases, said two or more phases including at least an oily phase and an aqueous phase; and recuperating said oily phase comprising acylglycerols or acylglycerols containing free fatty acids; wherein said enzymatic hydrolysis is accelerated by adding at least part of the one or more enzymes with phospholipase activity to at least part of the water used in the recuperation and/or at least one degumming process; and/or at least part of said triglyceride oils treated in said degumming process; and/or by facilitating mixing at least part of said one or more enzymes with phospholipase activity into said gums by increasing the triglyceride oil content of said gums; and an oil obtained by this process.

Reduced-odor thiol compositions

Medri, M.W., Consumer Products Corp., US8440219, May 14, 2013

The present invention provides compositions for and methods of delivering a therapeutically effective dose of a malodorous sulfide or disulfide group-containing compound, for example glutathione (reduced) and/or glutathione disulfide, in a vehicle that is effective to reduce the unpleasant odor and/or taste of the compound. The invention further provides methods for reducing the amount of oxidation occurring when sulfide group-containing compounds, such as glutathione, are incorporated into sugar and or sugar-free hard candies without subjecting the glutathione to thermal and/or moisture degradation where degradation is expressed as oxidation. The invention further provides vehicle compositions including the protected sulfide group-containing compounds and their use as medicaments. The sulfide group-containing compounds are protected from degradation by their dispersion into fats, oil, and/or fractionated or partially hydrogenated oils prior to their blending into the vehicle.

Fat crystallization accelerator

Nakano, M., *et al.,* Fuji Oil Co., Ltd., US8440250, May 14, 2013

Provided is a crystallization accelerator capable of accelerating fat crystallization a short time during a production step of a product of margarine, shortening, chocolate, or hard butter to be used for confectionary production or bread production. A phenomenon that sorbitan fatty acid ester having an esterification ratio of from 28% to 60% and a sorbitol-type content of from 20% to 40% exhibits a remarkable effect on acceleration of fat crystallization in a short time was found, and thus, the invention was completed.

Method for reducing 1, 2-diglyceride content of an edible oil

Wassell, P., *et al*, DuPont Nutrition Biosciences ApS, US8440435, May 14, 2013

The present application provides a method of reducing and/or removing diglyceride from an edible oil, comprising admixing an edible oil with an acyl acceptor substrate and a diglyceride:glycerol acyltransferase, wherein the diglyceride:glycerol acyltransferase is characterized as an enzyme which in an edible oil is capable of transferring an acyl group from a diglyceride to glycerol. The diglyceride:glycerol acyltransferase can comprise the amino acid sequence motif GDSX. The present invention also relates to the use of a diglyceride:glycerol acyltransferase in the manufacture of an edible oil, for reducing and/or removing diglyceride from said edible oil and to the use of said enzyme in the manufacture of a foodstuff comprising an edible oil for improving the crystallization properties of said foodstuff.

Molecular aggregate capable of undergoing phase transition by dehydrating condensation and method of phase transition thereof

Kunishima, M., Japan Science and Technology Agency, US8449979, May 28, 2013

Provided is a bilayer membrane vesicle capable of undergoing a phase transition. The bilayer membrane vesicle includes (i) a fatty acid salt having 6–20 carbon atoms; (ii) an alcohol or an amine compound having an aliphatic chain of 6–20 carbon atoms; and (iii) an artificial synthetic lipid or a phospholipid capable of forming a bilayer membrane. Preferably, this bilayer membrane vesicle further contains (iv) a tertiary amine as a component of the membrane. Also provided is a method of inducing a phase transition of a bilayer membrane vesicle, the method including the step of adding a dehydrating condensing agent or a dehydrating condensing agent precursor having the property of accumulating at an interface to the bilayer membrane vesicle. By causing the lipids that form a molecular aggregate to chemically change, it is possible to change the physical property and the morphology of the molecular aggregate and control the timing

CONTINUED ON PAGE 119

EXTRACTS & DISTILLATES

Biotechnology for the functional improvement of cereal-based materials enriched with PUFA and pigments

Čertík, M., *et al.*, *Eur. J. Lipid Sci. Technol. 115*:1247–1256, 2013, http://dx.doi.org/10.1002/ejlt.201300092.

Cereals as the major food supply are deficient in essential nutrients, such as PUFA [polyunsaturated fatty acids] and pigments. Biotechnological techniques based on solid state fermentation (SSF) and genetic engineering have been developed to naturally prepare functional cereals enriched with PUFAs and carotenoids. SSF represents a promising approach where the selected fungi (Zygomycetes) effectively utilize and transform raw cereal substrates to cereal-based bioproducts containing high amounts of valuable PUFA and carotenoids. Depending on the strain, types of cereal substrates, and cultivation conditions, a range of cereal-based bioproducts enriched with PUFA (up to 2.4% γ-linolenic acid, 4.2% arachidonic acid, 2.1% dihomo-γ-linolenic acid, 2.3% eicosapentaenoic acid) and pigments (8.5 mg β-carotene/kg prefermented cereals) have been prepared. In addition, cereals (barley, wheat) consisting of γ-linolenic and stearidonic acids have been prepared by genetic transformation of the fungal fatty acid Δ6-desaturase gene. Such functional prefermented cereal-based bioproducts are characterized by the acceptable nutritive, functional, and flavor values, the improved antioxidant, radical-scavenging, and thermal oxidation , as well as the enhanced safety; therefore, they may find applications in the food/feed fields.

Optimization of subcritical fluid extraction of carotenoids and chlorophyll a from *Laminaria japonica Aresch* by response surface methodology

Lu, J., *et al.*, *J. Sci. Food Agric. 94*:139–145, 2014, http://dx.doi.org/10.1002/jsfa.6224.

The carotenoids and chlorophyll a of *Laminaria japonica* Aresch were extracted using ethanol-modified subcritical 1,1,1,2-tetrafluoroethane (R134a). In the present study, the effects of pressure (5–17 MPa), temperature (303–333 K) and the amount of cosolvent (2–6% R134a, wt/wt) were investigated. Response surface methodology (RSM) combined with a Box–Behnken design was applied to evaluate the significance of the three independent variables on each response. A desirability function was conducted to simultaneously optimize the multiple responses. The optimum extraction conditions were as follows: extraction temperature 324.13 K, extraction pressure 17 MPa, and a cosolvent amount of 4.73%. Under these conditions, the yields of carotenoids and chlorophyll a were predicted to be 0.239 and 2.326 g kg^{-1}, respectively. It has been proved that subcritical R134a is a potential solvent, which can be an alternative to supercritical CO_2 for extraction of natural ingredients under mild conditions.

Suppression of visceral adipose tissue by palm kernel and soy-canola diacylglycerol in C57BL/6N mice

Tang, T.-K., *et al.*, *Eur. J. Lipid Sci. Technol. 115*:1266–1273, 2013, http://dx.doi.org/10.1002/ejlt.201300111.

The present study investigated the health effect of two types of diacylglycerol (DG) produced from (i) palm kernel (PK) oil of medium-chain saturated fatty acids and (ii) soy-canola oil (SC) blend of long-chain unsaturated fatty acids in C57BL/6N mice. As compared to diet containing 30% PK triacylglycerol (TG), 16 wk feeding trial on C57BL/6N mice with a diet consisting of 30% PKDG and 30% SCDG significantly ($p < 0.05$) reduced the fat accumulation in epididymal and retroperitoneal regions. Serum glucose, cholesterol, leptin, and insulin levels were significantly ($p < 0.05$) suppressed in PKDG- and SCDG-fed mice. In terms of gene expression, PKDG diet induced expression of acyl-CoA synthase long-chain (ACSL) and acyl-CoA synthase medium-chain mRNA in the small intestine of the mice while SCDG-fed mice upregulated ACSL in liver and small intestine. This suggests that the difference in fatty acid composition of DG may potentially induce β-oxidation in different organs in mice. Besides, expression of apolipoprotein B mRNA was reduced in mice fed with PKDG and SCDG, indicating the ability of PKDG and SCDG to reduce low density lipoprotein levels. In conclusion, structural differences between DG and TG markedly influenced the metabolism of lipids in the body while fatty acid composition has only showed a minor effect.

The dependence of lipid asymmetry upon polar headgroup structure

Son, M., and E. London, *J. Lipid Res. 54*:3385–3393, 2013, http://dx.doi.org/10.1194/jlr.M041749.

The effect of lipid headgroup structure upon the stability of lipid asymmetry was investigated. Using methyl-β-cyclodextrin-

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JAOCS
JOURNAL OF THE AMERICAN OIL CHEMISTS' SOCIETY

Journal of the American Oil Chemists' Society (December)

- Crystallization behavior of high-oleic high-stearic sunflower oil stearins under dynamic and static conditions, Martini, S., J.A.R. Cardona, Y. Ye, C.Y. Tan, R.J. Candal, and M.L. Herrera
- A spectroscopic method for hydroxyl value determination of polyols, Chalasani, S.R.K., S. Dewasthale, E. Hablot, X. Shi, D. Graiver, and R. Narayan
- Using SPME-GC/MS to evaluate acrolein production in cassava and pork sausage fried in different vegetable oils, Osório, V.M., and Z. de Lourdes Cardeal
- Physicochemical properties of dry-heated peanut protein isolate conjugated with dextran or gum arabic, Li, C., B. Zhu, H. Xue, Z. Chen, Q. Ding, and X. Wang
- Structures and binary mixing characteristics of enantiomers of 1-oleoyl-2,3-dipalmitoyl-*sn*-glycerol (S-OPP) and 1,2-dipalmitoyl-3-oleoyl-*sn*-glycerol (R-PPO), Mizobe, H., T. Tanaka, N. Hatakeyama, T. Nagai, K. Ichioka, H. Hondoh, S. Ueno, and K. Sato
- Identification of TAG and DAG and their FA constituents in lesquerella (*Physaria fendleri*) oil by HPLC and MS, Lin, J.-T., and G.Q. Chen
- Identification of tetraacylglycerols in lesquerella oil by electrospray ionization mass spectrometry of the lithium adducts, Lin, J.-T., and G.Q. Chen
- Isolation and characterization of a docosahexaenoic acid-phospholipids producing microorganism *Crypthecodinium* sp. D31, Okuda, T., A. Ando, E. Sakuradani, and J. Ogawa
- Testing the antioxidant effect of essential oils and BHT on corn oil at frying temperatures: a response surface methodology, İnanç, T., and M. Maskan
- Interaction of light and temperature on tocopherols during oxidation of sunflower oil, Choe, E.
- Quality changes and antioxidant properties of microencapsulated kenaf (*Hibiscus cannabinus* L.) seed oil during accelerated storage, Razmkhah, S., C.-P. Tan, K. Long, and K.-L. Nyam
- Changes in fatty acid, tocopherol and xanthophyll contents during the development of Tunisian-grown pecan nuts, Bouali, I., H. Trabelsi, I.B. Abdallah, A. Albouchi, L. Martine, S. Grégoire, G. Bouzaien, M. Gandour, S. Boukhchina, and O. Berdeaux
- Maleinization of soybean oil glycerides obtained from biodiesel-derived crude glycerol, Echeverri, D.A., W.A. Perez, and L.A. Rios
- Effects of monoacylglycerols on kinematic viscosity and cold filter plugging point of biodiesel, Dunn, R.O.
- Physical properties of low viscosity estolide 2-ethylhexyl esters, Cermak, S.C., J.W. Bredsguard, B.L. John, K. Kirk, T. Thompson, K.N. Isbell, K.A. Feken, T.A. Isbell, and R. Murray
- A study of process optimization of extraction of oil from fish waste for use as a low-grade fuel, Jayasinghe, P., I. Adeoti, and K. Hawboldt
- Adhesion and physicochemical properties of soy protein modified by sodium bisulfite, Qi, G., N. Li, D. Wang, and X.S. Sun
- Localisation of storage reserves in developing seeds of *Pongamia pinnata* (L.) Pierre, a potential agroforestry tree, Pavithra, H.R., B.K.C. Sagar, K.T. Prasanna, M.B. Shivanna, and B. Gowda
- Determination of organophosphorus pesticides in edible oils by dispersive microextraction based on acetonitrile/water-coated Fe_3O_4, Liu, T., Y. Ren, J. Xie, G. Song, and Y. Hu

Lipids

Lipids (December)

- Background diet and fat type alters plasma lipoprotein response but not aortic cholesterol accumulation in F1B golden Syrian hamsters, Dillard, A., N.R. Matthan, N.L. Spartano, A.E. Butkowski, and A.H. Lichtenstein
- Xanthophylls, phytosterols and pre-β1-HDL are differentially affected by fenofibrate and niacin HDL-raising in a cross-over study, Niesor, E.J., K. Gauthamadasa, R.A.G.D. Silva, G. Suchankova, D. Kallend, H. Gylling, B, Asztalos, E. Damonte, S. Rossomanno, M. Abt, W.S. Davidson, and R. Benghozi
- The effect of an energy restricted low glycemic index diet on blood lipids, apolipoproteins, and lipoprotein (a) among adolescent girls with excess weight: a randomized clinical trial, Rouhani, M.H., R. Kelishadi, M. Hashemipour, A. Esmaillzadeh, and L. Azadbakht
- Genetic polymorphisms in the *APOA1* gene and their relationship with serum HDL cholesterol levels, Bandarian, F., M. Hedayati, M.S. Daneshpour, M. Naseri, and F. Azizi
- Lipoprotein subfraction profile and HDL-associated enzymes in sickle cell disease patients, Ozturk, O.H., Y. Can, Z. Yonden, S. Motor, G. Oktay, H. Kaya, and M. Aslan
- Postpartum weight retention is associated with elevated ratio of oxidized LDL lipids to HDL-cholesterol, Puhkala, J., R. Luoto, M. Ahotupa, J. Raitanen, and T. Vasankari
- Identification of plasmalogen cardiolipins from *Pectinatus* by liquid chromatography–high resolution electrospray ionization tandem mass spectrometry, Řezanka, T., D. Matoulková, L. Kyselová, and K. Sigler
- Molecular species of phospholipids with very long chain fatty acids in skin fibroblasts of Zellweger syndrome, Hama, K., T. Nagai, C. Nishizawa, K. Ikeda, M. Morita, N. Satoh, H. Nakanishi, T. Imanaka, N. Shimozawa, R. Taguchi, K. Inoue, and K. Yokoyama
- Confirmation of the presence of squalene in human eyelid lipid by heteronuclear single quantum correlation spectroscopy, Borchman, D., M.C. Yappert, S.E. Milliner, R.J. Smith, and R. Bhola
- Separation of the fatty acids in menhaden oil as methyl esters with a highly polar ionic liquid gas chromatographic column and identification by time of flight mass spectrometry, Fardin-Kia, A.R., P. Delmonte, J.K.G. Kramer, G. Jahreis, K. Kuhnt, V. Santercole, and J.I. Rader

induced lipid exchange, sphingomyelin (SM) was introduced into the outer leaflets of lipid vesicles composed of phosphatidylglycerol, phosphatidylserine (PS), phosphatidylinositol, or cardiolipin, in mixtures of all of these lipids with phosphatidylethanolamine (PE), and in a phosphatidylcholine/phosphatidic acid mixture. Efficient SM exchange (>85% of that expected for complete replacement of the outer leaflet) was obtained for every lipid composition studied. Vesicles containing PE mixed with anionic lipids showed nearly complete asymmetry, which did not decay after 1 day of incubation. However, vesicles containing anionic lipids without PE generally only exhibited partial asymmetry, which further decayed after 1 day of incubation. Vesicles containing the anionic lipid PS were an exception, showing nearly complete and stable asymmetry. It is likely that the combination of multiple charged groups on PE and PS inhibits transverse diffusion of these lipids across membranes relative to those lipids that only have one anionic group. Possible explanations of this behavior are discussed. The asymmetry properties of PE and PS may explain some of their functions in plasma membranes.

Analytical approaches for the characterization and identification of olive (*Olea europaea*) oil proteins

Esteve, C., *et al., J. Agric. Food Chem. 61*:10384–10391, 2013, http://dx.doi.org/10.1021/jf4028359.

Proteins in olive oil have been scarcely investigated probably due to the difficulty of working with such a lipidic matrix and the dramatically low abundance of proteins in this biological material. Additionally, this scarce information has generated contradictory results, thus requiring further investigations. This work treats this subject from a comprehensive point of view and proposes the use of different analytical approaches to delve into the characterization and identification of proteins in olive oil. Different extraction methodologies, including capture via combinational hexapeptide ligand libraries, were tried. A sequence of methodologies, starting with off-gel isoelectric focusing followed by sodium dodecyl sulfate–polyacrylamide gel electrophoresis or high-performance liquid chromatography using an ultraperformance liquid chromatography column, was applied to profile proteins from olive seed, pulp, and oil. Besides this, and for the first time, a tentative identification of oil proteins by mass spectrometry has been attempted.

Mechanism of formation of *trans* fatty acids under heating conditions in triolein

Li, C., *et al., J. Agric. Food Chem. 43*:10392–10397, 2013, http://dx.doi.org/10.1021/jf402854b.

To elucidate the relationship between heat-induced *cis*/*trans* isomerization and reaction temperature and energy in unsaturated lipids, we investigated the molecular mechanism of the heat-induced *cis*/*trans* isomerization of 18:1 isomers. Triolein (18:1,9*c*) was heated at two range temperatures (130, 160, 190, 220°C and 135, 140, 145, 150, 155°C) and analyzed by the gas chromatography method. When the heating temperature increased to 150°C, the amount of *trans* 18:1n-9 changed from 0.0897 mg/g oil (1 h) to 0.1700 mg/g oil (3 h). This study shows that the *cis* to *trans* isomerization may occur at 150°C. The formation of fatty acid isomers followed a proton transfer route. All key geometries, transition states, intermediates, and bond dissociation energies (BDE) were optimized at the B3LYP/6-31G* level for the density functional theory. The zero-point energy corrections of the isomers were carried out using calculations at the B3LYP/6-311++G** level. The calculated energy difference between the *cis* and *trans* oleic acid was equal to 7.6 kJ/mol, and the energy barriers of the transition from *cis* 18:1n-9 to *trans* 18:1n-9 were 294.5 kJ/mol. The intrinsic reaction coordinates were obtained to be used as an expression of the reaction route and to analyze the transition states and intermediates. The study results suggest that the heating temperature should be kept under 150°C, to avoid the risk of *trans* fatty acid intake in daily food.

Determination of α-tocopherol in vegetable oils using a molecularly imprinted polymers–surface-enhanced Raman spectroscopic biosensor

Feng, S., *et al., J. Agric. Food Chem. 61*:10467–10475, 2013, http://dx.doi.org/10.1021/jf4038858.

We report the development of a novel hybrid "capture-detection" molecularly imprinted polymers–surface-enhanced Raman spectroscopic (MIPs-SERS) biosensor for the detection and quantification of α-tocopherol (α-Toc) in vegetable oils. α-Toc served as the template for MIPs synthesis. Methacrylic acid formed as the functional monomer. Ethylene glycol dimethacrylate was the cross-linking agent, and 2,2′-azobisisobutyronitrile was used as the initiator. The synthesized MIPs functioned to rapidly and selectively adsorb and separate α-Toc from oil components. We validated a dendritic silver nanostructure synthesized by a displacement reaction to be a suitable SERS substrate for the enhancement of Raman signals. Second-derivative transformations and chemometric models based upon SERS spectral features confirmed the possibility of a rapid and precise detection and quantification of different spiking levels of α-Toc in four different sources of vegetable oils (Mahalanobis distance from 15.93 to 34.01 for PCA model; $R > 0.92$, RMSE < 0.41 for PLSR model). The MIPs-SERS biosensor had a high sensitivity as well as a good recovery for α-Toc analysis in vegetable oils. The entire analysis required 15 min or less to complete with limited sample preparation.

More Extracts & Distillates can be found in this issue's supplement (digital and mobile editions only).

An important note to AOCS Members

AOCS is excited to announce the launch of our new Association Management System (AMS) is complete. We all know new software means many new changes and we want to let you know how this may have affected you.

Why upgrade?
Converting to the new system provides us with new technology and improved processes to help us better serve you. Not only are we now able to offer you the best in products and services, you'll also be able to choose your username and password. (Something we have heard our members prefer.)

What was upgraded?
In December 2013, AOCS began the process to switch to the new Association Management System. This new software system handles everything we do, from processing transactions to generating invoices, to managing your membership benefits. We've been preparing for this switch since February 2013 and are excited to bring this upgrade to our members.

How does this affect me?
The updates have been made with you in mind and will be most noticeable through the AOCS website:

- We have a new online store with an easier check-out system.
- Your member profile page has been upgraded. Visit your profile to access past orders, manage how AOCS communicates with you, and keep up-to-date with AOCS member benefits and dues renewals.

We encourage you to take time to login to your account and explore your new profile page.

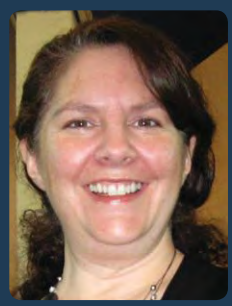
I still have questions – who do I contact?
If you still have questions, please contact us by phone +1 217-693-4813 or by email: membership@aocs.org. Although our technology has changed, the same friendly faces remain ready to serve you every day! Please know that we are doing everything we can to make this process as easy as possible for you, our member.

When it comes to your professional society, we know that you have a choice. Thank you for choosing AOCS.

Sincerely,
Barbara Semeraro, Membership Manager

Professional Pathways

Professional Pathways is a regular Inform *column in which AOCS members discuss their professional experiences and share advice with young professionals who are establishing their own careers in oils and fats-related fields.*



Terri Germain is a consultant for the surfactant industry. She has more than 26 years of experience in the industry, starting with lab research and product development at Stepan Co. (Northfield, Illinois, USA) before later switching to business development at Oxiteno (Pasadena, Texas, USA).

Why did you join AOCS?

I was introduced to AOCS by other members and colleagues I worked with. I especially was drawn to the technical info at the annual meetings, which gives me a great opportunity to learn things that are related to my career but that I would not normally run across as part of my job.

Describe your career path.

I started as an entry-level chemist at Stepan in their Household, Industrial and Institutional application department. I worked my way up to senior manager before leaving to take a job as the research and development (R&D) manager at McIntyre Group (subsidiary of Solvay-Rhodia as of 2009; University Park, Illinois, USA). After more than 20 years in R&D, I decided to learn more about the commercial side of business and have worked as a new business development manager and market development manager.

I have needed to make several key career decisions. One early on in my career was deciding whether I wanted to manage people or continue only as a technical expert. I chose management and learned a great deal about people as well as myself. I feel having a balance of technical, management, and leadership skills has really been an asset for me.

What is the biggest challenge you have encountered in your career and how did you address it?

I am currently facing the biggest challenge of my career. Nine months ago, I was laid off due to a location closure. I have been self-employed as a consultant as a means to pay the household bills while I consider my next career move and look for a full-time position. I have been doing very well as a consultant and at the same time learning more about my personal tolerances of the unknown. Will I have work next

month? Do I have the knowledge and confidence to write about specific topics? Can I take on a mundane project and stay focused for days on end? Do I really want to learn how to pay quarterly income taxes? What about health insurance? I am addressing these challenges by staying in contact with colleagues, friends, and mentors. It is nice to know that I can succeed as a consultant (so far) and this gives me breathing room to be choosy about job opportunities.

What do you love about your job?

Almost everything! I'm not too crazy about filling out long product questionnaires, but the rest is mostly fun. I love the opportunity to travel and present. Teaching and training are very rewarding when others "get it" and feel some of the excitement and passion I have about surfactants.

How do you see the industry changing in the next five years?

I see a continued emphasis on care, whether it is care for clothes, surfaces, people, or the environment. One thing that must start changing is our connection to the public. Our industry, and the chemical industry in general, is not trusted. There is a lot of misleading and just plain wrong information out there. We need to embrace our lack of connection thus far and do everything in our power to earn the trust of the public we serve. We are public servants and provide a valuable commodity to our communities; we help keep the world clean and healthy. I'm proud of our industry and our integrity—I think the world should know how great we are too!

> "Learn some biochemistry; it is going to be a big part of the future of our industry. Learn what product development is about even if you are going into another field."

How has your industry changed since you entered the field?

The biggest change has been trying to do more at a faster pace with less people but more tools. When I started my career, we did not have the internet. I can't imagine working now without it. I remember making search requests to our internal library, waiting a few days for the results, reviewing and then requesting more documents, which I then needed to wait for as well. Now I get frustrated if I can't find a data sheet or a patent online in less than 20 seconds!

Regulatory work is much more prominent in the development process due to market claims we did not have early in my career. So much needs to be understood and known about the origin of a product and its raw materials. Another big change is the reduction in R&D personnel over the past two decades and the retirement of so many knowledgeable people. The remaining people spend a large majority of their time formulating and reformulating and addressing the ever-changing needs of regulations and consumers. There is so much to know and so much problem solving that is needed that I feel basic research has taken a backseat.

Do you have any advice for those looking to enter your field?

Do it! It's a great field. Learn some biochemistry; it is going to be a big part of the future of our industry. Learn what product development is about even if you are going into another field. It is extremely important to understand how your job function relates to other job functions and the overall business and success of your company. Product development is where innovation developments touch the commercial aspects of a company. Also, join AOCS! Be part of your industry beyond just being a part of your company.

Describe memorable job experiences.

I remember so many . . . Here's a bittersweet one. Our company had been bought and we all needed to brush up our resumes and re-interview with the new company. Many of us were not going to keep our jobs. One chemist who worked for me remarked how surprised she was at all the things she knew how to do after only a few years. She could synthesize products, run analytical procedures, handle technical service requests, formulate, and provide training. I smiled and remarked that of course she knew all of these things; she was good at her job. Internally, I smiled as well because I felt I had succeeded in my goal of cross training the chemists that worked for me to broaden their experience and knowledge. I did my job; I helped advance the career of another.

If you were starting your career again, what would you do differently?

I'm not sure; it's been a great career so far.

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Do you have any advice for young professionals who are trying to develop an effective network of other professionals?

Get involved with trade organizations, volunteer, and present at your local schools, ask questions, use LinkedIn, post on blogs, develop relationships with the sales people of your company.

Please describe a course, seminar, book, men-tor, or speaker that has inspired you in ways that have helped you advance your career.

I have to say that Arno Cahn was a significant inspiration to me. Cahn, who was AOCS president in 1996–1997, was a highly-regarded expert in surfactants and detergents. I met him early in my career and late in his. He was a regular presenter for a Detergents Fundamentals seminar. He made it seem so easy to do and easy to understand. He talked to his audience, he understood them, and he taught them. I try to present like him, with a love of the subject and a deep understanding that can match the needs of the audience wherever they are in their knowledge base. I want to make it fun to learn and less intimidating. I want to build the confidence of the audience as he did. I enjoy presenting and I continue to aspire to make surfactants seem easy.

What are the opportunities for advancement in your career/field and how can someone qualify for such advancements?

There is a need for technical people that can market and sell. Beyond just a technical service role, we need more people that truly understand the chemistry and can connect the vision and missions of companies to their products and how they are positioned. Customers need people that understand their needs and requirements and can have conversations that connect with them. Read and continue taking seminars and classes. Also work on listening and learn about personality types. Take professional development seminars on negotiation, selling strategies, supervising and managing people, statistics, and experimental design. Be curious and enthusiastic.

How would you describe the culture in your field, and how has it developed?

The culture is diverse because my field touches so many markets. I don't know if the culture has improved much lately because of the disconnect between our industry and the general public. We constantly seem to be in defensive mode as industry representatives. I feel we are the "good guys in the white hats" if only we could get that message out better.

In your area/field and considering today's market, is it more important to be well rounded or a specialist?

Of course the answer is both! A specialist needs to be well-rounded in order to be a specialist. A depth of understanding is only possible if you can understand the many direct and indirect relationships between your specialty and how it interacts with other things and situations.

What is your opinion toward the value of obtaining or possessing a graduate degree during a challenging economy?

I have only a bachelor's degree, but getting a college degree changed my life and opened up opportunities I would never have had any other way. In my field, a higher degree is very beneficial but not a necessity unless you aspire to a director level or higher. It is hard to go back to school once you start working, but the learning process is so much more meaningful with some work experience first. Get the degrees you want and need to meet your goals. Don't let the economy dictate your education opportunities if you can—your career will see several economic swings over the years. ■

[FAST FACT]

TURNING BACK THE CLOCK ON MIDDLE AGE SPREAD

Babies are born with lots of calorie-burning brown fat. But, as people age, they lose their stores of metabolically active brown fat and accumulate energy-storing white fat, which can lead to weight gain. The recent identification of brown fat stem cells in adult humans offers new hope that it may be possible to turn back the clock.

"The unique identification of human brown fat stem cells in the chest of patients aged from 28 to 84 years is profound. We were able to isolate the human stem cells, culture and grow them, and implant them into a pre-human model which has demonstrated positive effects on glucose levels," said Amit N. Patel, who led the team that reported the findings in the journal *Stem Cells* in November 2013. The discovery could help researchers identify potential drugs that increase the body's own ability to make brown fat or find novel ways to directly implant brown fat stem cells into patients.

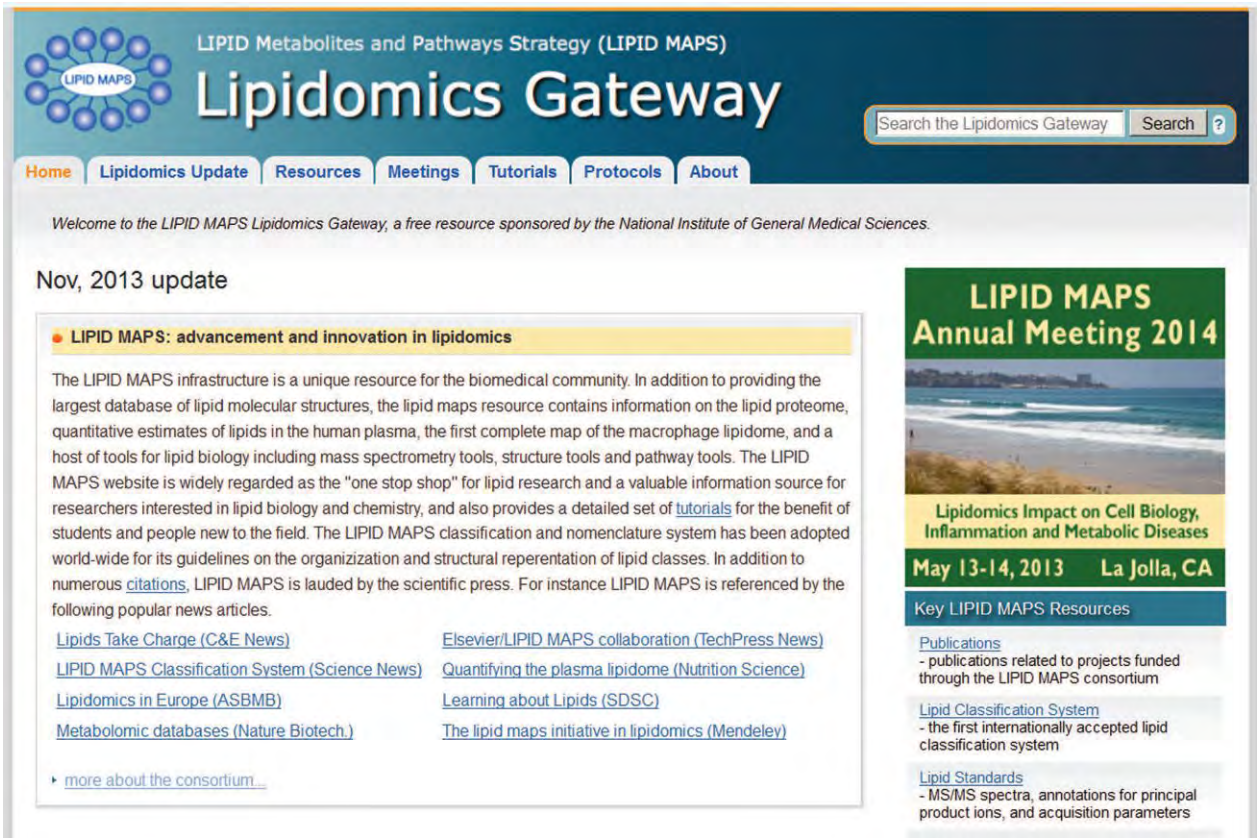

FIG. 1. Home page of the Lipidomics Gateway, in November 2013. See www.lipidmaps.org.

Lipidomics comes of age

Laura Cassiday

A decade ago, many cell and molecular biologists viewed lipids as mere structural components of cell membranes. DNA and proteins were where the action was, giving rise to their own –omics fields, while cellular lipids quietly toiled away, performing vital yet underappreciated roles in energy metabolism, cell and organelle membrane structure, intracellular trafficking, and cell signaling.

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- **A growing appreciation of the roles lipids play in biology and disease has inspired the development of tools that allow researchers to probe the complexity of lipids in cells.**
- **Researchers are now applying such tools to identify markers that can be used to diagnose diseases, monitor therapies, and identify promising new drug targets.**
- **Meanwhile, the vast size of the human lipidome and the fleeting nature of cellular lipid metabolites make the measurement of many lipids and their spatial distribution in individual cells extremely difficult.**

Today, a growing recognition of lipids' many roles, combined with a new suite of tools to probe the overwhelming complexity of lipids in cells, is fueling the burgeoning field of lipidomics—defined as the identification, classification, and quantification of all lipids in biological systems. And researchers are discovering that lipids, which dwarf both genes and proteins in chemical diversity, are every bit as interesting and relevant in terms of biology and disease.

The rapid maturation of the lipidomics field owes much to a consortium of lipids researchers known as the Lipid Metabolites and Pathways Strategy (LIPID MAPS). "In 2002, when I proposed this consortium to the National Institutes of Health in a grant, the word 'lipidomics' did not appear in any publications," says Edward Dennis, professor of chemistry, biochemistry, and pharmacology at the University of California, San Diego, and director of the LIPID MAPS initiative.

At that time, researchers were just beginning to appreciate the enormous complexity of lipids in organisms—current estimates of unique lipids number in the hundreds of thousands—but they lacked tools to identify and quantify the molecules. Moreover, a single lipid might be known by multiple names, and researchers weren't sure how to group structurally similar lipids into categories. This lack of a standardized nomenclature and classification system thwarted collaborations and stymied the development of a universal lipids database.

Then, in 2003 Dennis and 11 co-principal investigators and other researchers from universities, medical centers, and companies across the United States were awarded a five-year, $35 million "Glue Grant" from the National Institute of General Medical Sciences, and LIPID MAPS was born. The grant, which was renewed for an additional five years in 2008, totaled $73 million and ended on July 31, 2013. "The consortium started at ground zero, and now, a decade later, there are over 1,000 publications in lipidomics," says Dennis. "The LIPID MAPS consortium itself has produced over 370 publications, with many more coming."

A COMMON CLASSIFICATION SYSTEM

The overriding goal of LIPID MAPS was to inventory all of the major, and many of the minor, lipids in mammalian cells. In addition, the researchers wanted to investigate how diseases and other perturbations could alter the cellular lipid complement. But first, the consortium needed to build the infrastructure to support the nascent field of lipidomics.

They began by developing a classification system with eight categories of lipids, grouped together according to their chemical backbones: fatty acyls, glycerolipids, glycerophospholipids, sphingolipids, sterol lipids, prenol lipids, saccharolipids, and polyketides. Although the first six classes are found in all organisms, the final two occur only in plants and bacteria.

Along with the new classification system, LIPID MAPS researchers adopted nomenclature rules to simplify naming of complex lipids and to systematically name recently discovered lipids. In addition, each lipid was given a unique 12-digit identifier so that it could be unambiguously identified in databases. Finally, the consortium devised a structural drawing representation for lipids, with head groups drawn on the right and hydrocarbon chains extending to the left. According to Dennis, the new classification, naming, and drawing systems have been "very widely and internationally accepted."

With these bookkeeping details resolved, the LIPID MAPS consortium was eager to start collecting data. Fortunately, mass spectrometry (MS)—the workhorse of the proteomics field—is also ideal for analyzing lipids. MS works by ionizing chemical compounds and then separating them according to their mass-to-charge (m/z) ratios. Researchers identify molecules from characteristic peaks on a mass spectrum.

Because many compounds in cells share the same or similar m/z, complex samples such as cell extracts are often subjected to liquid chromatography (LC) or gas chromatography (GC) prior to MS. LC and GC separate molecules on the basis of their chemical interactions with a chromatography column. To help identify tricky compounds, investigators can use tandem MS (MS/MS), in which a specialized mass spectrometer, such as a triple quadrupole instrument, selects a specific precursor ion and fragments it, producing a characteristic pattern of MS peaks.

USING MS

MS is well suited for lipidomics for several reasons. Most lipids are below 1,200 Da, well within the range of mass spectrometers. Also, during ionization for MS, most lipids become singly charged, simplifying their identification from mass-to-charge ratios. Moreover, MS is sensitive enough to detect small amounts of minor lipid species in cells.

However, the tools and tricks of the proteomics trade had to be adapted to lipids. For example, to identify and quantify specific molecules by MS, researchers need standards. MS standards are compounds, usually labeled with stable isotopes, that have known mass-to-charge ratios and fragmentation patterns. Researchers can identify unknown peaks in mass spectra by comparison to standards, and in some cases, quantify the unknown compounds.

A decade ago, researchers had very few lipid MS standards in their toolkit. Now, they can choose from more than 500 lipid standards, which are available through companies such as Avanti Polar Lipids, Inc. (http://avantilipids.com), a LIPID MAPS core facility and supplier of MS lipid standards developed by the consortium. Investigators can use one standard for several structurally related lipids. "Investigators all over the world are now using the same MS lipid standards, and they were developed by LIPID MAPS," says Dennis.

BEST IN CLASS

The LIPID MAPS consortium also optimized methods for analyzing each of the six major lipid categories found in mammals. The consortium established six core laboratories, each of which optimized the sample extraction, MS standards, chromatography columns and solvents, and MS techniques for a particular category of lipid (Table 1). The consortium named this series of protocols CLASS, for comprehensive lipidomics analysis by separation simplification.

CLASS enables the sensitive detection of lipids within each of the six lipid categories. An alternative to CLASS is shotgun lipidomics, in which investigators inject all lipids in a sample

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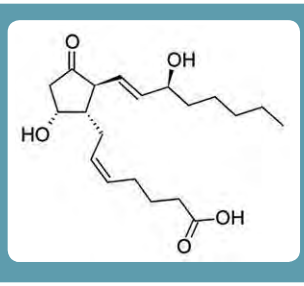
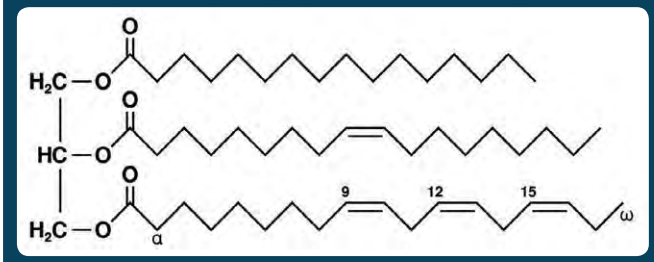
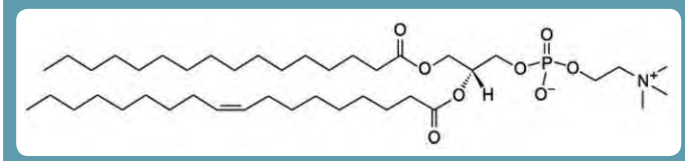
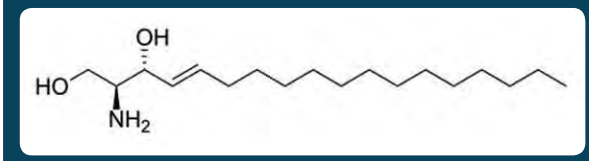
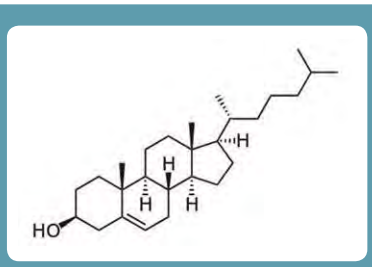
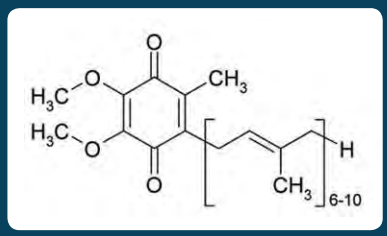
TABLE 1. CLASS protocols for the six categories of mammalian lipids

Lipid category	Example	Extraction	Chromatographic separation
Fatty acyls	Prostaglandin D2 	Methanolic HCl/ isooctane (fatty acids) C_{18} SPE cartridge (eicosanoids)	Gas chromatography (fatty acids) HPLC/RP C_{18} column (eicosanoids)
Glycerolipids	Triacylglycerols 	Ethyl acetate/ isooctane	HPLC/NP silica column
Glycerophospholipids	Phosphatidylcholines 	Methanolic HCl/ chloroform	HPLC/NP silica column
Sphingolipids	Sphingosine 	Incubation overnight in methanol/ chloroform at 48°C followed by methanolic KOH	HPLC/NP NH_2 column
Sterol lipids	Cholesterol 	Methanol/ chloroform (free form) Ethyl acetate/ isooctane (fatty acyl esters)	HPLC/RP C_{18} column (free form) HPLC/NP silica column (fatty acyl esters)
Prenol lipids	Coenzyme Q10 	Methanol/ chloroform	HPLC/RP C_8 column

Abbreviations: CLASS, comprehensive lipidomics analysis by separation simplification; SPE, solid phase extraction; HPLC/RP, reversed-phase high-performance liquid chromatography; HPLC/NP, normal-phase HPLC.

INFORMATION

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directly into the mass spectrometer without prior separation. Although faster and less laborious than CLASS, shotgun lipidomics can miss minor species of lipids that are overwhelmed by more abundant ones. However, the technique can be good for obtaining a quick overview of the major lipids that differ between two states, for example, between cancerous and normal tissue.

Dennis and his colleagues put their new methods to the test by analyzing the lipidome of pooled human blood plasma from 100 healthy people who were representative of the United States ethnic population (Quehenberger, *et al.*, 2010). The study revealed a remarkable diversity of lipids in human plasma, with over 500 distinct species detected and quantified using CLASS and LC- or GC-MS/MS. This reference plasma lipidome is an important first step toward identifying lipids that change in disease states or in response to therapies, the researchers say.

In a separate study, LIPID MAPS researchers examined how lipids in mammalian immune cells change in response to a simulated bacterial infection (Dennis, *et al.*, 2010). The investigators analyzed the lipidome of a mouse macrophage cell line treated with an inflammatory bacterial molecule that induces macrophage activation. The team detected changes in more than 400 lipids over a 24-hour period. The immediate response of the macrophages to the inflammatory molecule was characterized by increased eicosanoid synthesis, while a delayed response involved sphingolipid and sterol biosynthesis.

HELPFUL RESOURCES

Another contribution of LIPID MAPS is the creation of a free, comprehensive website for lipids researchers, known as the Lipidomics Gateway at www.lipidmaps.org (Fig. 1). "You can now type in the name of any lipid—traditional name, shorthand name, or full chemical name—and the website will immediately give you all the information on that lipid, the mass spec breakdown, and the stereochemically correct structure," says Dennis. There are currently more than 40,000 different lipids in the database, with new lipids added regularly. In addition, the Lipidomics Gateway includes lipidomics tutorials, protocols, publications, and other resources.

The success of the LIPID MAPS consortium has inspired similar initiatives in Japan, Europe, the United Kingdom, and Singapore, according to Dennis. Meanwhile, several universities worldwide have established academic centers specializing in lipidomics, including the Kansas Lipidomics Research Center at Kansas State University; the Institute for Multidisciplinary Biochemistry of Lipids at the Université de Lyon, France; and the Lipidomics Research Center Graz, in Austria.

The Medical University of South Carolina (MUSC; Columbia, USA) has established a Lipidomics Shared Resource (Fig. 2). According to Operations Director Alicja Bielawska, the Lipidomics Shared Resource uses tools and methods that she and her colleagues developed to study the role of sphingolipids in cancer. Sphingolipids function in cell signaling pathways that control vital processes such as apoptosis, proliferation, and inflammation. Such "bioactive" lipids are of particular interest to cancer researchers.

The MUSC Lipidomics Shared Resource (http://tinyurl.com/MUSC-lipidomics), which specializes in sphingolipids, is composed of two units: a synthetic unit and an analytical unit. The synthetic unit makes sphingolipids, molecular probes, and various inhibitors of sphingolipid-metabolizing enzymes. The analytical unit uses LC-MS/MS to profile lipid components of biological samples such as cells, blood, and tumor tissue (Fig. 2). "We now offer 14 analyses covering more than 300 different lipid species," says Bielawska. Researchers around the world can access a form on the Lipidomics Shared Resource website to place their synthetic or analytical orders.

LIPIDS IN DISEASE

Many researchers hope that lipidomics will help them identify robust new biomarkers to diagnose diseases and monitor therapies. Indeed, abnormal lipid levels have already been linked to ailments such as cancer, cardiovascular disease, neurological disorders, and diabetes.

A recent paper on the lipidomics of influenza infection demonstrates the feasibility of such an approach (Tam *et al.*, 2013). The investigators, who were members of the LIPID MAPS consortium, obtained nasopharyngeal lavage samples from patients during the 2009–2011 influenza seasons and analyzed their lipidomes. Interestingly, the ratio between two closely

related lipids, 9- and 13-hydroxyoctadecadienoic acid, differentiated individuals with worse clinical symptoms and heightened immune responses. Although the study needs to be repeated in a larger cohort to confirm the results, these preliminary findings suggest that the ratio between the two lipids could help doctors monitor the severity of and recovery from influenza infection.

Lipidomics is also likely to identify promising new drug targets. Lipid-lowering drugs called statins are already widely used for the treatment and prevention of cardiovascular disease. Statins lower cholesterol levels by inhibiting an enzyme, HMG-CoA reductase, that produces cholesterol in the liver. In the future, however, researchers may use drugs that directly target disease-promoting lipids, instead of the proteins that synthesize them.

Roger Sabbadini, founder and vice president of Lpath, Inc., has taken this approach by developing monoclonal antibodies against bioactive lipids. One lipid that Lpath has targeted is sphingosine-1-phosphate (S1P). Tumors secrete S1P to stimulate the growth of new blood vessels, which feed the tumor—a process called tumor angiogenesis. S1P also has anti-apoptotic properties that protect tumors from cell suicide. S1P levels are elevated in the blood plasma from patients with several forms of cancer.

FIG. 2. Jason Pierce, operator of the liquid chromatograph–tandem mass spectrometer in the Analytical Unit of the Medical University of South Carolina (MUSC; Columbia, USA), is shown loading lipid extracts of human plasma samples of cancer patients and normal controls for comprehensive sphingolipid analyses. Credit: MUSC Shared Lipidomics Resource.

In animal studies, a monoclonal antibody against S1P blocked tumor cell proliferation, metastasis, and angiogenesis (Sabbadini, R. A., 2011). The LPath antibody is now being tested in a phase II clinical trial for the treatment of renal cell carcinoma. A different formulation of the antibody is in a phase II clinical trial for age-related macular degeneration, a condition characterized by the spontaneous overproduction of blood vessels in the back of the eye. Both trials will be complete in 2014.

"So far, we're the only ones that have been able to develop therapeutic-grade antibodies against bioactive lipids," says Sabbadini. Making antibodies against lipids is challenging because lipids are small and not very immunogenic. Lpath's ImmuneY2™ platform prepares lipids in a way that makes them recognizable to the immune system of a mouse or rabbit, enabling antibody production. Lpath scientists make thiolated lipid analogs that can be conjugated to another small molecule known as a hapten: Together, the lipid and hapten elicit antibody production. The ImmuneY2 platform also includes assays to measure the strength of the lipid-antibody interaction.

In the future, Sabbadini expects more lipidomics companies like Lpath to emerge. "Pharmaceutical companies are going to say, 'we've got a gold mine here that nobody's ever mined, and we're going to put the energy into mapping the lipidome and differentiating the important lipids from the unimportant ones,'" he says.

However, the vast size of the lipidome, numbering in the hundreds of thousands of molecules, makes such a task daunting. In addition, accumulating evidence suggests that a single snapshot of the lipidome in time and location is not enough to gain a full understanding of how lipids modulate disease. A new branch of lipidomics called fluxolipidomics seeks to follow lipids and their metabolites as a function of time and compartment.

FLUXOLIPIDOMICS

"Fluxolipidomics is the only way to approach phenotypes," says Michel Lagarde, a professor at the Institute for Multidisciplinary Biochemistry of Lipids at the Université de Lyon, France. This is because in biological systems, lipids are in a constant state of flux, being broken down into various stable and unstable metabolites and trafficking between compartments.

Some of the most interesting lipids are fleeting, making their measurement extremely difficult or impossible. For example, thromboxane A2 is a lipid produced by activated blood platelets that aids blood clotting. However, thromboxane A2

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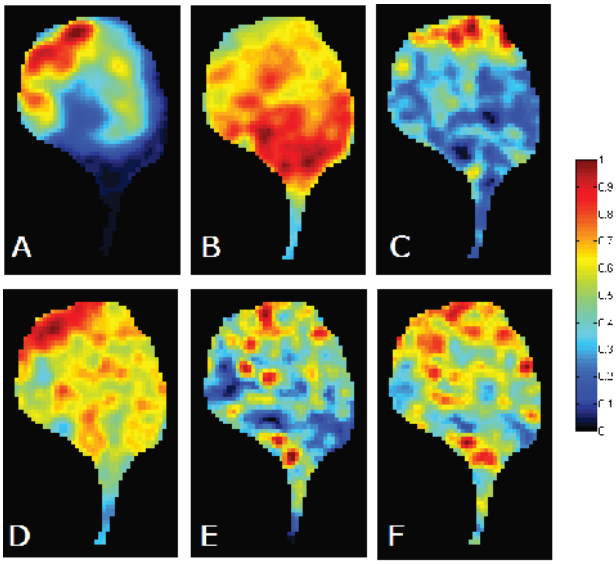

FIG. 3. Time-of-flight secondary ion mass spectrometry (TOF-SIMS) images obtained from a single Aplysia neuron: (A) vitamin E, α-tocopherol, $C_{29}H_{50}O_2$, M•, m/z 430.3, (B) hydrocarbon (m/z 128.1), (C) cholesterol, $C_{27}H_{45}$, $[M + H - OH]^+$ (m/z 369.3), (D) phosphocholine head group (m/z 184), $C_5H_{15}NPO_4$, $[M + H]^+$, (E) 1-hexadecyl-2-octadecenoyl-sn-glycero-3-phosphocholine (PC; 16:0e/18:1) high-mass fragment (m/z 709.6), $C_{39}H_{75}O_7PNa$, [M + Na – TMA]+, and (F) sum of PC(16:0e/18:1)-related peaks [m/z 709 ($C_{39}H_{75}O_7PNa$, [M + Na – TMA]+), m/z 725 ($C_{39}H_{75}O_7PK$, $[M + K - TMA]^+$), m/z 746 ($C_{42}H_{85}NO_7P$, $[M + H]^+$), m/z 768 ($C_{42}H_{84}NO_7PNa$, $[M + Na]^+$), and m/z 784 ($C_{42}H_{84}NO_7PK$, $[M + K]^+$)]. (Scale bar = 100 µm; TMA = trimethylamine) Reprinted with permission from *Anal.Chem. 85*:2231–2238, Copyright © 2013. The American Chemical Society.

is very unstable, with a half life of only 30 seconds. As a result, researchers must measure its stable metabolite thromboxane B2 as a surrogate for A2. But thromboxane B2 itself is converted into at least three metabolites. "Depending on the moment you measure, you can get a different answer," says Lagarde.

Lagarde's laboratory is currently contemplating fluxolipidomics experiments on the essential fatty acids linoleic and linolenic acids. A figure showing the complexity of this undertaking can be viewed in this issue's supplement (digital and mobile editions only). "If we gave people a mixture of linoleic and linolenic acids labeled with ^{13}C, a stable isotope of carbon, to ingest, we could follow as a function of time the labeled fatty acids and their various metabolites, and their incorporation into phospholipids in different compartments," he says. For example, the researchers could collect blood samples at various time points over a 48-hour period, separating the samples into blood platelets, red cells, white cells, and plasma. In each cell type and at each time point, they could measure by MS the different molecules labeled with ^{13}C.

SINGLE-CELL LIPIDOMICS

Another challenge remaining for lipidomics is determining the spatial distribution of lipids on individual cells—so-called single-cell lipidomics. According to Nicholas Winograd, professor of chemistry at Penn State University, unique characteristics of individual cells can be lost in conventional lipidomics studies.

So Winograd has pioneered the imaging of lipids on single cells with a technique called secondary ion MS (SIMS). To generate an image with SIMS, researchers scan across the cell surface to acquire hundreds of mass spectra at different locations. A specialized mass spectrometer bombards the cell with a beam of C_{60} cluster ions, or buckyballs, that dislodge secondary ions for detection by MS/MS. Researchers identify lipids by their characteristic fragmentation patterns and color-code them to produce an image.

Winograd says that SIMS is ideal for lipid imaging because the technique is unusually sensitive to lipids—more so than proteins—and it has submicron spatial resolution, which is important because mammalian cells are, on average, around 20 microns in diameter. In comparison, matrix-assisted laser desorption/ionization (MALDI) MS, another type of MS used for imaging tissues, has a spatial resolution of 20–200 microns. In addition, cluster ion beams can erode material in a more controlled manner than a laser. "You can erode a few nanometers of the cell surface a minute, and then stack images up as you erode to get a 3D representation of the cell," says Winograd.

As part of the LIPID MAPS consortium, Winograd's lab imaged lipid distributions on a lawn of activated mouse macrophages and compared the data to the lipidome obtained by conventional LC/MS (Passarelli *et al.*, 2013a). Although improvements in sensitivity are needed, the researchers were able to identify more than 50 glycerophospholipids on the macrophage surface. In general, the lipids that changed upon macrophage activation were consistent with those identified by LC/MS.

In another recent study, Winograd and his colleagues used C_{60} SIMS to image lipids on the surface of individual neurons from the California sea slug, *Aplysia californica* (Passerelli *et al.*, 2013b). These neurons are significantly larger than mammalian cells, simplifying single-cell imaging. The investigators discovered that the dominant lipid on the surface of *A. californica* neurons was 1-hexadecyl-2-octadecenoyl-*sn*-glycero-3-phosphocholine [PC(16:0e/18:1)], which partially colocalized with vitamin E (Fig. 3). This overlap is consistent with evidence suggesting a role for vitamin E in the synthesis of PC. Studying the distributions of lipids on single cells may help elucidate their roles in cellular functions, the researchers say.

Although the imaging of individual *A. californica* neurons is a major step forward in single-cell lipidomics, technological improvements are needed to eventually obtain high-resolution images of individual mammalian cells. "It's challenging because a single mammalian cell is only 20 microns in diameter, and the lipid concentration may be only a few percent, so there just aren't that many molecules to detect," says Winograd.

In addition, the probability of ionizing a lipid molecule by MS is very low, somewhere between 1 in 10,000 and 1 in 1,000,000, Winograd estimates. "We and others are working on clever ways to enhance the ionization probability, which

is needed to get meaningful lipid distributions on such small objects," he says.

As the LIPID MAPS initiative draws to a close, the final studies being completed and papers written, Dennis concludes that the consortium has accomplished what it set out to do: nurture the growing field of lipidomics. Over the past decade, the new field has overcome most of its major growing pains, yet challenges remain. For example, determining the stereochemistry of lipids can be difficult because stereoisomers have the same mass and are thus indistinguishable by MS. "In the future it will be absolutely mandatory to distinguish between stereoisomers because we know more and more that stereochemistry is very important for function," says Lagarde.

Perhaps the biggest accomplishment of the lipidomics field is the growing recognition that lipids are a large and essential component of the "interactome" of genes, transcripts, proteins, and metabolites that characterize biological systems. Only by adopting such a systems biology approach can we hope to understand the complex interplay between the various types of molecules that make life possible.

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Patents (cont. from page 105)

of phase transitions such as membrane fusion. In the membrane fusion, for example, fusion can occur without leakage of the contents of the bilayer membrane vesicle.

Low viscosity mono-unsaturated acid containing oil-based dielectric fluids

Amanullah, M., *et al.,* Biolectric Pty. Ltd., US8440116, May 14, 2013

Disclosed herein are a dielectric fluid composition and a method thereof. Also disclosed are viscosity modifiers and a method of lowering the viscosity of an oil-based dielectric fluid. The composition includes an oil with a high mono-unsaturated fatty acid content and one or more fatty acid alkyl esters, each having a fatty acid and an alkyl moiety, wherein the alkyl moiety of the fatty acid alkyl esters has 1–4 carbon atoms, and wherein both the oil and the fatty acid alkyl ester are in the range of 40–60% v/v of the dielectric fluid composition. The viscosity modifier includes one or more fatty acid alkyl esters with an alkyl moiety and a fatty acid moiety, wherein the alkyl moiety has 1–4 carbon atoms. The method of lowering the viscosity includes blending the viscosity modifier and a vegetable oil-based dielectric fluid in a ratio of 40:60–60:40.

Solubilizing composition

Ohkawa, Y., NOF Corp., US8440726, May 14, 2013

A solubilizing composition containing (i) at least one oily component selected from esters of oleic acid with alcohols having 1–3 carbon atoms and triglycerides of fatty acids having 6–12 carbon atoms, (ii) a polyoxyethylenesorbitan fatty acid ester, (iii) a polyhydric alcohol which is liquid at 15–25°C, and (iv) one or more acidic phospholipids where the fatty acid constituting an acyl group thereof is selected from saturated fatty acids having 6–14 carbon atoms and unsaturated fatty acids have 16–18 carbon atoms, which contains 20–40% by weight of the component (i), 30–45% by weight of component (ii), 15–40% by weight of component (iii), and 0.5–4% by weight of the component (iv), based on 100% by weight of a total amount of the components (i) to (iv).

Lipid-treated particles and polymers containing the particles

El-Shoubary, M., Cristal USA Inc., US8436077, May 7, 2013

A coated particulate solid composition includes a particulate inorganic solid having deposited on its surface a treatment oil comprising lipids. The particulate inorganic solid can include a base particle of titanium dioxide, zinc sulfide, zinc oxide, iron oxide, lead oxide, aluminum oxide, silicon dioxide, zirconium oxide, and/or chromium oxide. The treatment oil is optionally a vegetable oil. The treatment oil containing lipids optionally contains glycerides such as triglycerides and diglycerides, and can contain phospholipids. In one embodiment, the coated particulate solid composition includes particulate titanium dioxide, wherein the treatment oil provides increased bulk density and enhanced dispersibility in plastic as well as improved lacing resistance.

Lubricating composition containing borated phospholipid

Baker, M., and G.B. Rhoads, The Lubrizol Corp., US8445417, May 21, 2013

The invention relates to a lubricating composition containing (i) a borated phospholipid, (ii) an amine salt of a phosphoric acid ester, and (iii) an oil of lubricating viscosity. The invention further provides for the use of the lubricating composition for lubricating a limited slip differential.

Patent information is compiled by Scott Bloomer, a registered US patent agent with Archer Daniels Midland Co., Decatur, Illinois, USA. Contact him at scott.bloomer@adm.com.

Oil of clove (*Syzygium aromaticum*)

Mohamed Fawzy Ramadan Hassanien

The cold-pressing technique for extracting vegetable oil is an alternative to conventional practices and speaks to consumers' desire for natural products. Cold pressing involves no thermal or chemical treatments and no refining. Cold-pressed oils may contain lipophilic phytochemicals as well as natural antioxidants.

Cloves are the dried flower buds of an aromatic evergreen tree (*Syzygium aromaticum*) that is cultivated in many tropical countries. Cloves are used in the food industry for their special aroma and their health benefits. Cloves contain a variety of potentially bioactive compounds such as sesquiterpenes, tannins, and triterpenoids. The main aroma constituent of clove buds, eugenol (4-allyl-2-methoxyphenol, Fig. 1), is present in clove essential oil. Reportedly, eugenol has biological and antimicrobial activity (Gulcin *et al.*, 2012; Shukri *et al.*, 2012).

The US Food and Drug Administration lists eugenol as Generally Regarded As Safe when administered at levels not exceeding 1,500 mg/kg in food. In addition, the World Health Organization Expert Committee on Food Additives has established the acceptable daily human intake of clove oil as 2.5 mg/kg body weight.

- **Nine different fatty acids were identified in cold-pressed clove oil (CPCO).**
- **High levels of unsaponifiables, particulary tocopherols, were also detected in CPCO.**
- **The antiradical power of CPCO was found to be higher than that of extra virgin olive oil (EVOO).**

FATTY ACIDS AND TOCOLS OF COLD-PRESSED CLOVE OIL (CPCO)

The fatty acid profile of CPCO is presented in Table 1. Nine fatty acids were identified. Linoleic (18:2) and oleic (18:1) acids were the main fatty acids. Together, they comprised about 80% of the total fatty acid. CPCO contained significant levels of monounsaturated fatty acids (MUFA; 39.7 g/100 g total fatty acids), which are comparable to the values for cold-pressed hemp, cranberry, blueberry, onion, and milk thistle seed oils but much lower than that of 81% and 82% in parsley and carrot cold-pressed seed oils, respectively. CPCO had a polyunsaturated fatty acids (PUFA) content of 42.1 g/100 g of total fatty acids. This PUFA content was lower than that in onion (64–65 g/100 g) and milk thistle (61 g/100 g) cold-pressed seed oils.

From a health point of view, MUFA have been shown to lower "bad" LDL (low density lipoprotein) cholesterol and retain "good" HDL (high density lipoprotein) cholesterol. This is in fact the major benefit of olive oil over other highly polyunsaturated oils, where PUFA reduce both the "bad" and the "good" serum cholesterol levels in our blood. A rapidly growing literature illustrates the benefits of PUFA in alleviating cardiovascular, inflammatory, and heart diseases; atherosclerosis; autoimmune disorders; diabetes; and other diseases.

The saturated fatty acids (SFA) in CPCO represented about 18 g per 100 g of total fatty acids (Table 1), which is much lower than the 30.8 g/100 g of total fatty acids in cold-pressed cardamom seed oil and comparable to the 13.8 and 15.9 g/100 g of total fatty acids found in cold-pressed milk thistle and roasted pumpkin seed oils, respectively. The SFA levels of 18% in CPCO were higher than those of 7.4–9.7 g/100 g of total fatty acids in cold-pressed parsley, onion, and hemp seed oils.

CPCO was characterized by high levels of unsaponifiables (25.3 g/kg). Levels of α-, β-, γ-, and d- tocopherols in CPCO were 148, 5.6, 418, and 186 µg/kg oil, respectively. In addition, amounts of α-, β-, γ-, and d- tocotrienols were 1.11, 55, 85, and 9.49 µg/kg oil, respectively. a-Tocopherol is the most

TABLE 1. Comparison of fatty acid content of cold-pressed clove oil (CPCO) with some seed oils

Fatty acid	CPCO	Parsley	Cardamom	Caraway	Hemp	Cranberry	Carrot	Onion	Roasted pumpkin	Milk thistle
10:0	2.8	ND	ND	ND	ND	ND	ND	ND	ND	ND
12:0	0.1	ND	ND	ND	ND	ND	ND	ND	ND	ND
14:0	0.1	ND	1.5	0.2	ND	ND	ND	ND	0.1	0.1
16:0	8.6	3.1	26.4	12.1	6.3	7.8	3.8	6.4	8.9	8.9
16:1	0.4	0.1	1.6	1.0	ND	ND	ND	0.2	0.1	0.1
18:0	6.6	4.2	2.3	3.4	2.7	1.9	0.5	2.4	6.4	4.8
18:1	39.4	80.9	49.2	24.0	11.7	22.7	82.0	24.8	36.3	23.8
18:2	40.2	11.0	15.2	55.8	59.3	44.3	13.2	65.2	47.2	60.8
18:3	2.0	0.5	2.7	0.2	20.0	22.3	0.5	0.1	0.2	0.2

[a]ND = not detected. Sources: Parry *et al.* (2006); Ramadan (2013).

efficient antioxidant of the tocopherol isomers, b-tocopherol has 25–50% of the anti-oxidative activity of a-tocopherol, and the g-isomer has 10–35% (Ramadan, 2012). The levels of tocopherols detected in CPCO may contribute to the stability of the oil toward oxidation.

ANTIRADICAL POWER (AP) OF CPCO COMPARED WITH EXTRA VIRGIN OLIVE OIL (EVOO)

The oxidative stability of oils and fats depends on the fatty acid composition, the presence of minor fat-soluble bioactives, and the initial amount of hydroperoxides. The AP of antioxidants may be influenced by the radical system and other testing conditions. Two or more radical systems are needed to better study a selected antioxidant for its AP (Ramadan, 2013; Ramadan *et al.*, 2012).

Antiradical characteristics of CPCO and EVOO (as a standard crude oil with respect to high levels of nutritive antioxidants and bioactives) were compared using stable DPPH• (diphenylpicrylhydrazyl radical) and galvinoxyl radicals. Table 2 (page 122) shows that CPCO had higher AP than olive oil. After 60 min of incubation, 70% of DPPH• were quenched by CPCO, while olive oil was able to quench only 45%. Electron spin resonance measurements showed also the same pattern, wherein CPCO quenched 57% of galvinoxyl radical and olive oil deactivated about 38% after 60 min of reaction.

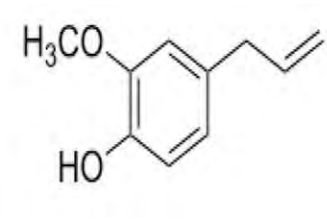

FIG. 1. Eugenol (4-allyl-2-methoxyphenol).

CPCO and olive oil have different compositions of fatty acid and lipid-soluble bioactives. CPCO has higher levels of phenolics than EVOO (4.6 vs.3.4 mg/g, respectively). Thus, CPCO could be used in foods to provide nutrition and health benefits.

Phenolic compounds in vegetable oils are very important for the oxidative stability of PUFA they contain. Also, edible oils rich in natural antioxidants may play a role in reducing the risk of chronic diseases.

The antioxidant effect of phenolic compounds is mainly due to their redox properties and is the result of their free-radical scavenging activity, transition metal-chelating activity, and/or singlet-oxygen-quenching capacity. The total phenolic values of CPCO were higher than that of 1.8–3.4 mg GAE [gallic acid equivalent]/g oil for cold-pressed parsley, onion, cardamom, and milk thistle seed oils. On the other hand, tocopherol levels

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TABLE 2. Scavenging effect of CPCO and EVOO at different incubation times

Time (min)	DPPH•[a] (% remaining)		Galvinoxyl radical[b] (% remaining)	
	CPCO	EVOO	CPCO	EVOO
30	40	60	54	73
60	30	55	43	62

[a]As measured by changes in absorbance values at 515 nm; DPPH•, diphenylpicrylhydrazyl radical; CPCO, cold-pressed clove oil; EVOO, extra virgin olive oil.
[b]As measured by electron spin resonance.

in oils may have a great impact on their AP. Increasing ring methyl substitution led to an increase of scavenging activity against the DPPH• radical, and also to a decrease in oxygen radical absorbance capacity.

The stronger AP of CPCO compared to EVOO may be due to (i) the differences in content and composition of unsaponifiable materials, (ii) the diversity in structural characteristics of phenolic antioxidants present, (iii) a synergism of phenolic antioxidants with other active components, and (iv) different kinetic behaviors of potential antioxidants. From our results we can suggest that CPCO may serve as a dietary source of phenolic substances, which may act as antioxidants for disease prevention and/or general health promotion through improved nutrition (Ramadan, 2012).

CONCLUDING REMARKS

CPCO is a good source of essential fatty acids and lipid-soluble bioactives. The high linoleic and oleic acid contents make CPCO nutritionally valuable. Tocols and phenolics present in CPCO may be of nutritional importance as natural antioxidants, reacting directly with and quenching free radicals and preventing lipid peroxidation. CPCO could be a non-conventional supply for pharmaceutical industries and edible purposes and may provide health benefits to consumers.

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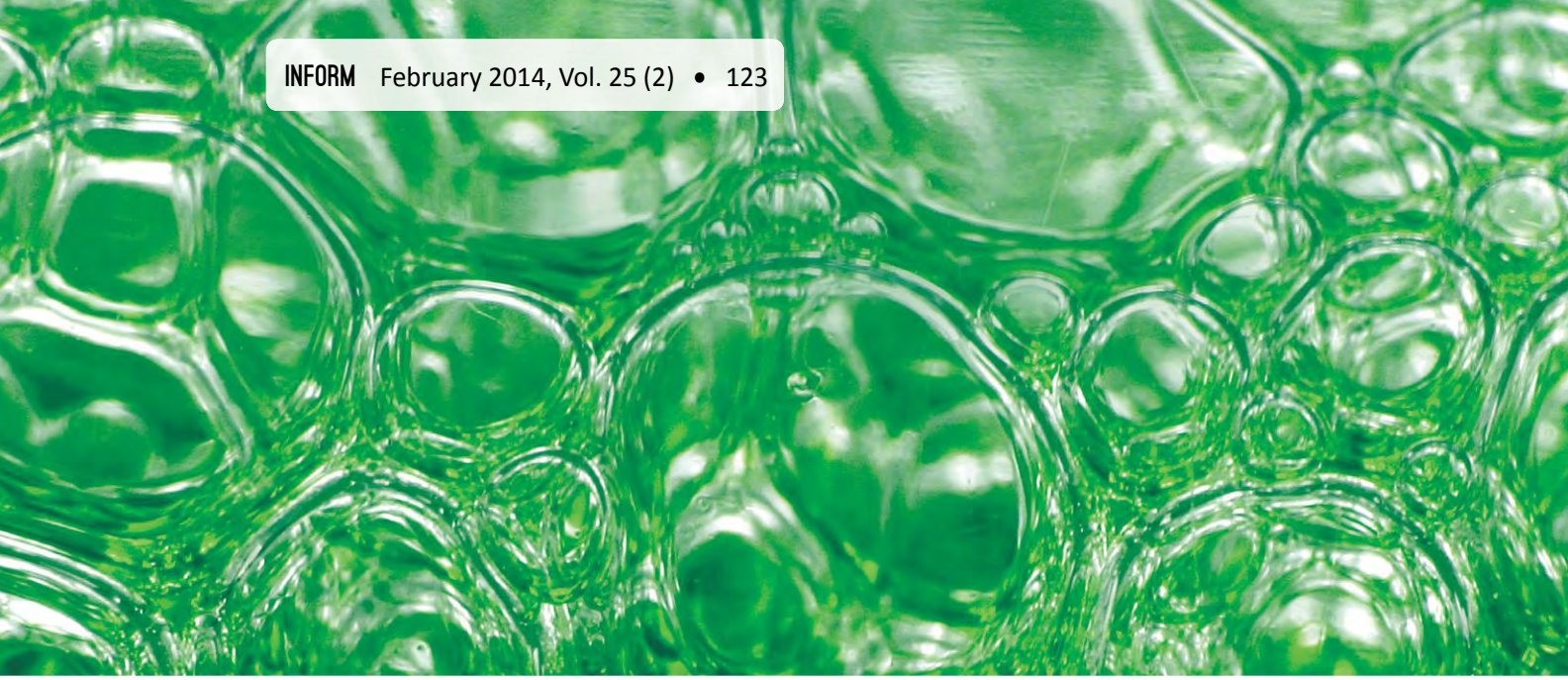

An integral approach to fluid-bed coating and matrix-encapsulation of sensitive systems for the detergent industry

Zoltán Szilágyi and Andreas Baranyai

The fluidized-bed spray-granulation process is already since 40 years in standardized industrial use as a thermal drying technology, complementary to spray-drying. Both drying processes use the intensive heat exchange between a liquid with high specific surface (as sprayed droplets with a diameter between 20 µm and 80 µm) and the environment at well-defined temperatures in order to transform liquid one- or multicomponent systems into dried powder or granular particles (Uhlemann and Mörl, 2000).

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- **Fluid-bed technology has been recognized for years as the most appropriate process tool for drying sensitive liquid systems by transforming them into free-flowing particles without loss of the proprietary characteristics of the active ingredient.**
- **The technology can also be used to protect liquid or solid sensitive ingredients or actives of a specific detergent formulation against internal or external effects, or to achieve the most efficient performance of an active ingredient in a well-determined time or place of action.**
- **Several examples within the cleaning and detergent industry illustrate the versatility of this technology.**

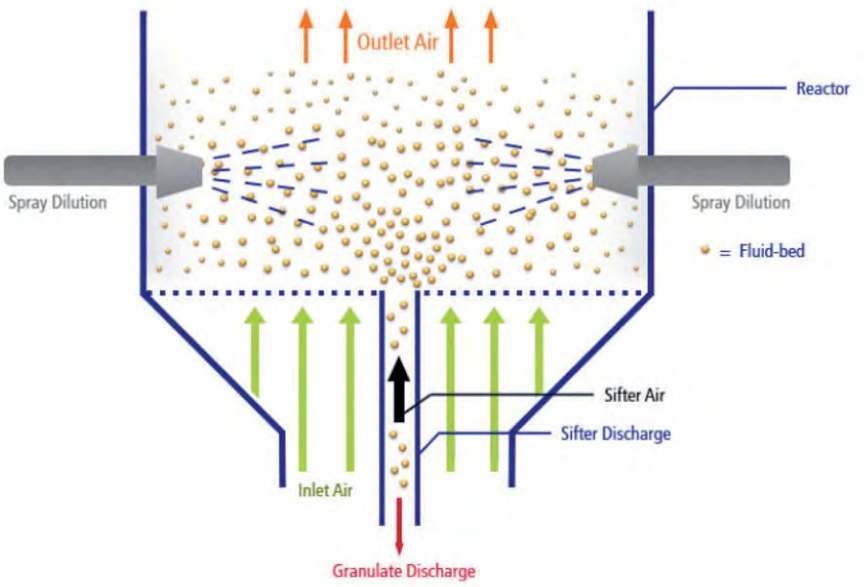

FIG. 1. Principle of fluid-bed technology.

A short-term and heavy interaction between the liquid droplets and the heated air of the environment is taking place during the drying period of a classical spray-drying process: A well-defined temperature is generated at the surface of the drying droplets and of the dried particles, which enables the smooth and gentle drying of sensitive systems. On the other hand, this timely limited period determines the final particle size of the generated powder particles, keeping them usually within the range between 50 µm and 150 µm.

Fluidized-bed spray granulation can be seen as an extension of the spray-drying technology, as—without changing the basic principles of intensive heat transfer interaction during the drying of sprayed liquid droplets—it provides through the optimization of the process technology a flexible and very differentiated tool for individual handling of the generated particles.

TABLE 1. Overview of fluid-bed technology

Precursors	Liquids, Aqueous solutions, Oils, Melts, Powders, Filter cakes, Flakes, Granulates, Suspensions, Aqueous emulsions, Melt suspensions, Melt emulsions, Bulk goods
Fluid-Bed Processes	Fluid-Bed Spray Granulation, Coating, Agglomeration, Matrix Encapsulation, Spray Drying, Spray Freezing, Drying
Final Products	Granule sizes between 100 µm and 5 mm Powder sizes between 20 µm and 100 µm **with following properties** dispersible, Direct compression, Reduced hygroscopicity, Homogeneous component distribution in each particle, Equivalent stoichiometry in each granule, Prevention of demixing, Precise release of active substances, Protection of granulate from environmental effects, Prevention of storage incompatibilities

FLUID-BED TECHNOLOGY

The foundation of fluid-bed technology is the solid filling (generated through drying of the smallest droplets sprayed into the reactor chamber) streamed by a fluid medium (air). The streaming allows the solid particles to be brought into a fluidized state and to be mixed up with the fluid medium, a free-flowing, loosened mass of fine-grained particles. The main characteristic of the fluid-bed process is the continuous drying, which allows the setup of a specific residual moisture in the treated system. Additionally, the strong heat and material transfer intrinsic to the process hinders any overheating during drying.

Fluid-bed technology provides some important advantages when compared to classical spray-drying, such as:

- Simultaneous drying and particle formation in the same process chamber
- Individual particle handling through fluidization of generated particles
- Flexible particle size generation between 100 and 5000 µm
- Development of new techniques allowing the treatment of particle surfaces by means of spray-agglomeration, coating and matrix-encapsulation

Figure 1 shows the principle of fluid-bed technology.

In the past, fluid-bed technology was mainly used for the production of specific granular raw materials and actives at the pharmaceutical and agrochemical industries. Today it is widely spread across several different segments and areas, also in the cleaning and detergent industries.

Spray-drying equipments have been used for a long time for the production of washing and cleaning powders. In the last two decades, however, fluid-bed techniques are increasingly being in place for the generation of "special" granulates, as a differentiated product form when compared to "classical" dusty powders. On the one side driven by the improvement in the product form providing handling and application advantages, for example, easy-to-dose or direct compression capability. On the other side due to legal requirements on safety, health and environmental aspects when processing and using fluid-bed derived products: granulates show here several inherent advantages over powders, especially dust freedom and free flowing ability.

Table 1 provides an overview on the manifold starting material forms that may be handled by means of the different processes within the fluid-bed technology in order to result in solid particles with specific and targeted properties (Inprotec, 2013).

The fluid-bed technology through its versatility provides the platform for additional processes, bringing sensitive actives and specific raw materials into their most efficient physical form for their optimal performance, due to the need of :

- Protection against external or internal influences such as humidity, hygroscopicity, oxidation, volatility, toxicology, light
- Isolation against internal incompatibilities or even chemical reactions with other components within a formulation
- Masking of taste, odor and color
- Improvement of particle stability (handling, storage and transport), particle size and appearance
- Release control in the targeted environment until final release by means of different mechanisms

There are different technological processes available to achieve some of these targets. For instance, chemical methods such as coacervation or *in situ* polymerization of liquid systems may provide the isolation of individual active droplets. Such an isolation may be achievable also with the help of "Gel-Caps" or by using polyvinylalcohol (PVA) foils. The fluid-bed technology, however, seems to be the most universal of all methods when targeting the protection of sensitive systems without interfering in their mode of action.

An increase in stability can be already reached when changing the state of matter of sensitive materials from liquid to solid (drying). A short introduction to spray drying and fluid-bed spray granulation is given next.

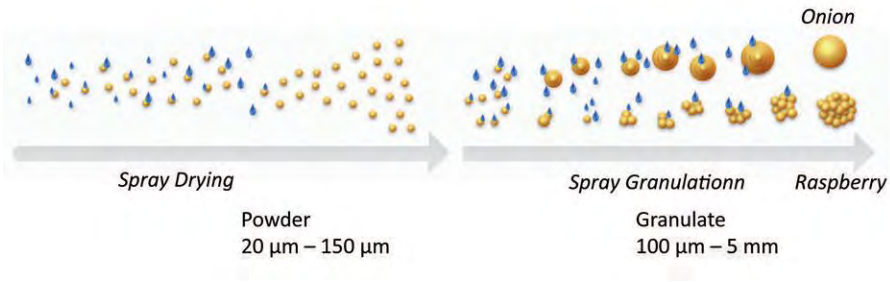

FIG. 2. Spray drying and spray granulation.

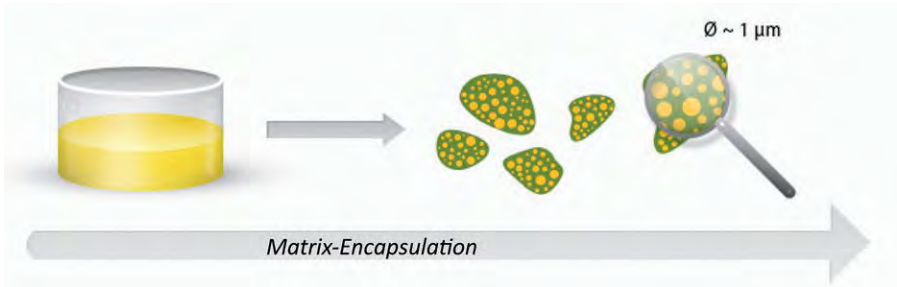

FIG. 3. Matrix-encapsulation.

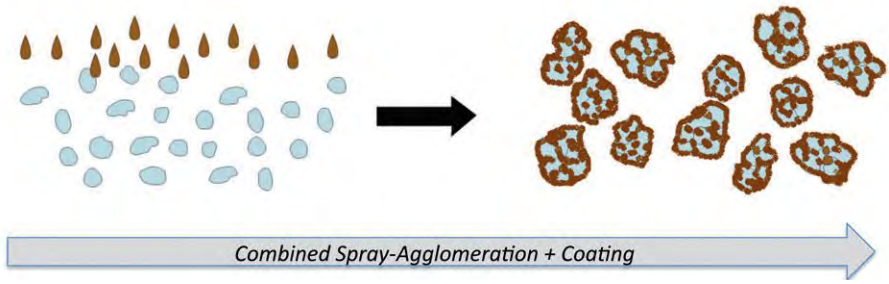

FIG. 4. Combined spray agglomeration + coating.

SPRAY DRYING/SPRAY GRANULATION

Small droplets of liquid sprayed through nozzles solidify into particles with sizes between 20 µm and 150 µm ("seeds") through direct heat transfer with hot inlet-air (spray drying). These seeds are held under fluidization and provide the surface for the layer-by-layer adsorption and drying for the following droplets generating granulates with adjustable sizes between 100 µm up to 5 mm (spray granulation). Depending on physical properties and drying parameters, specific particle shapes ("onion" and "raspberry") with different final characteristics and attributes can be produced (Fig. 2).

A batchwise or continuous production of granulates is possible, depending on the spray granulation equipment used.

In several cases, however, there is a need for further particle treatment in order to achieve an even better protection of actives or to suppress unwanted properties, for example, hygroscopicity. At the same time, it is vital to keep the efficiency target of actives, that is, their ability to perform optimally. Coating and matrix-encapsulation by means of fluid-bed processing allow the systematic manipulation of the surface morphology of treated active particles without loss of the required performance at the targeted "places" and "times." Depending on the size and state of

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TABLE 2. Overview of different coating/encapsulation processes depending on precursor materials.

Fluid-bed process	Precursor actives	Final product form
Matrix-encapsulation	Liquid droplets or solid particles < 20 µm	Particles 100–5000 µm
Combined agglomeration + coating	Solid particles 20–200 µm	Particles 200–5000 µm
Pure coating	Solid particles > 20 µm	Particles 200–5000 µm no substantial size increase

matter of the active precursors, three processes can be applied: matrix-encapsulation, combined agglomeration + coating, and pure coating (Table 2).

Matrix-encapsulation. Matrix-encapsulation is the technology of choice when long-term stability and protection of sensitive liquids (e.g., fragrances, essential oils) or powders are required. The precursor liquids or powders added to aqueous solutions containing specific soluble coating/encapsulation material are emulsified or brought into suspension and sprayed as 20 µm to 50 µm emulsion or suspension droplets for further processing (Fig. 3, page 125). As an alternative to aqueous solutions, hot melts of waxes also can be used to form sprayable hot-melt emulsions or suspensions containing the precursor actives.

Liquid actives can thus be embedded as 1–2 µm droplets ("inner phase") within solid matrix particles of adjustable size from 20 µm up to 5 mm. Loading up to 50% of liquids in solid granulates can be achieved.

Combined spray-agglomeration + coating. This technique is applied whenever the particle sizes are too small for their individual coating, but by being water-soluble no matrix-encapsulation with water-soluble materials is possible. In a first spray-agglomeration step, where the sprayed coating liquid acts as a "binder," fluidized powder particles with a particle size below 100 µm adhere to each other forming so-called "snow-crystal" friable agglomerates. When reaching a particle size above 150–200 µm, a coating of the individual agglomerates with the coating material occurs, as shown in Figure 4 (page 125).

Pure coating. Macroscopic particles from 150 µm to 5 mm diameter can individually be coated with specific hydrophilic or hydrophobic coating materials in a fluid-bed, as shown in Figure 5.

A careful selection of the most suitable coating candidate is a determining factor when aiming at the following specific objectives:

- Targeted release of the active in the desired medium
- Active stability against internal and external influences

Additionally, coating materials may also alter the substrate characteristics and morphology by:

- Viscosity adjustment
- Emulsion stabilization
- Taste and odor modification
- Active load increase
- Providing a diffusion barrier
- Improvement of the free-flowing ability

Three release models can be targeted depending on the choice of the most suitable coating material:

- *Controlled-release*—the release of the coated active is triggered by a specific factor such as a change of pH, T, p, H_2O/electrolyte-concentration
- *Slow-release*—through the slow and continuous dissolving of the more or less water-soluble coating layer
- *Diffusion*—through the semipermeable coating walls

Coating materials can be selected from two main systems:

- Water soluble—hydrophilic coating materials
- Water-insoluble—hydrophobic coating materials

Water-soluble coating systems are usually:

- Aqueous solutions of :
  - » Natural gums (gum arabica, acacia gum, carrageenan, . . .)
  - » Gelatin, peptides
  - » Modified, water-soluble starches
  - » Dextrins (maltodextrins, cyclodextrins)
  - » Glucose derivatives (di-, oligo-, polysaccharides)
  - » Cellulose ethers
  - » Polyols
  - » Synthetic polymer systems (polyacrylates)
  - » Silicates and other inorganic, water-soluble salts
- Hot melts of:
  - » Polyethylene glycol (PEG), surfactants (e.g., fattyalcohol-ethoxylates), urea

Hydrophobic (water-insoluble) systems regularly used in coating processes are:

- Aqueous dispersions/suspensions of:
  - » Self-crosslinking polymeric systems such as polyurethane (PU), polyethylene (PE), and polyacrylamide (PA)
- Hot melts of:
  - » Hardened vegetable oils (soy, palm kernel, coconut)
  - » Fatty acids with a carbon-chain length >C16, for example, stearic, behenic, . . .

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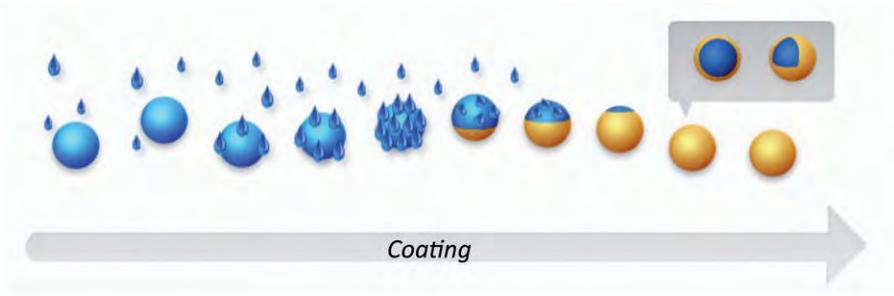

FIG. 5. Hydrophilic and hydrophobic coating materials in a fluid bed.

- » Fatty alcohols with a carbon-chain length >C14, for example, cetyl, stearyl, . . .
- » Paraffinic waxes
- » Natural waxes (carnauba, bees, . . .)

There are today already "hundreds" of specific, customized "coating formulations," suitable for specific targets and release requirements. And each day new candidates, mostly synthetic polymeric ones, join.

Following are some selected examples of fluid-bed technology generated coating and encapsulation for cleaning and detergency applications (*Jahrbuch für den Praktiker 2010*):

- Coating of inorganic salt particles with a self-cross-linked water-insoluble, semipermeable polymer system (controlled diffusion of the coated active through the semipermeable wall)
- Coating of enzymes with PEG-waxes (protection against internal incompatibility with the other formulation components)
- Coating of granular percarbonates, enzymes, and bleach activators, e.g., pthalimidoperoxycarboxylic acids (PAP) with special polymers (triggered controlled release) (Revolymer, 2013)
- Matrix-encapsulation of silicones with 15% silicone active loading (spray-granulated particles from an aqueous emulsion and spray-frozen granulates from a urea-based melt-emulsion)
- Matrix-encapsulation of essential oils/fragrances out of a water-soluble starch solution (powder and granulate)
- Combined spray-drying + spray-agglomeration + coating of a multicomponent system (each particle with the same stoichiometry protection from other components of the same formulation triggered controlled release).

The fluid-bed technology and related processes such as spray granulation, coating and matrix-encapsulation have been described. Simultaneous drying and particle forming, individual particle handling paired with selective and particular treatment of the granulated surfaces are the key features of the different fluid-bed processes. Particles can be protected, modified, and manipulated to deliver the best performance through different mechanisms. Several examples of the described processes of actives and components of cleaning and detergent formulations are presented.

Andreas Baranyai is CEO and Zoltán Szilágyi is in sales and marketing at inprotec AG in Heitersheim, Germany. Baranyai can be contacted at aby@inprotec-ag.de.

This article was reprinted with permission from the August 2013 issue of *SOFW Journal*, which tracks developments in the soap, detergent, perfume, personal care, and cosmetics industries.

[FAST FACT]

William (Bill) Christie, founder and editor of The AOCS Lipid Library, posted this explanation of a new study involving the lipid cardiolipin on The AOCS Lipid Library blog (http://lipidlibrary.aocs.org/news/blog.htm): "Cardiolipin is a unique lipid in many ways. For example, it is located only in mitochondria, where it is a key component of the oxidative phosphorylation system, and it has twice as many phosphate and fatty acid moieties as the conventional phospholipids. Also, depending on tissue, it has a restricted range of fatty acid components, resulting in a few relatively symmetrical molecular species. It has been accepted up until now that the two phosphate groups have very different acidities, with pK_1 = 2.8 and pK_2 = 7.5–9.5. A new study has come up with very different results; i.e. pK_1 = 2.15, similar to that of phosphoric acid, with pK_2 about one unit greater. The paper is open access (Olofsson, G. and E. Sparr, "Ionization constants pK(a) of cardiolipin. *PLOS One*, 8, e73040 (2013); doi:10.1371/journal.pone.0073040). The data are important for understanding how cardiolipin interacts with enzymes and proteins in membranes to affect their biological functions."

For more information, visit http://tinyurl.com/cardiolipin.

Novel chemicals (cont. from page 81)

How customized algal oils led to improved high performance fiber lubricants

Kevin Quon

In October 2013, Solazyme jointly announced a commercial supply agreement with Goulston Technologies Inc., a leading producer of high-performance fiber lubricants. The agreement is a perfect case study of the way the San Francisco-based renewable oils company uses its novel technology platform to increase the sustainability of triglycerides. This collaboration will focus on the utilization of Solazyme's oils to supply a sustainable substitute to currently utilized ingredients and the development of new products with enhanced functionality and performance.

Textiles may not have been the first addressable market Solazyme had in mind, but along the journey to designing a renewable source of fuel, the company adapted its corporate direction to align with the strengths of its technology platform. Over the past decade, the company has refined its ability to control microorganisms, both in regards to scaling up production and in terms of modifying product output.

Solazyme was among the first to realize that heterotrophic microalgae could be harnessed as a highly productive organism, efficiently serving as a flexible host for triglyceride oil production. The company has since developed the ability to adjust the carbon chain length of fatty acids, modify the degree of chemical saturation in a triglyceride, and control the position of a fatty acid on the glycerol moiety. Today, this unique combination of tailoring capabilities enables a wide array of customized solutions for companies looking to increase their product functionality.

The fiber lubricants market is just one example of how this technology is now being implemented. One challenge facing this industry is that formulators are limited to very few classes of ingredients capable of meeting specific conditions.

Compounding this market restriction is the growing demand for biobased raw materials that reduce environmental impact. According to industry estimates, the fiber lubricants market has grown to more than 1 billion pounds [450 million kilograms] annually. These lubricants have a relatively short lifespan of functionality, making it important to secure stable raw material supply chains. In many cases, such lubricants quickly become part of the waste stream - they are applied to the fiber before high speed processing and are subsequently removed during the fabric dyeing process.

The agreement announced in October 2013 called for the supply of Solazyme's Tailored™ Algal Oils to be used in a new class of sustainable products being developed by Goulston Technologies. Solazyme is expected to fulfill this agreement utilizing its customized high-oleic oil. These high-oleic algal oils will provide Goulston with excellent lubrication, viscosity, and stability properties while operating at elevated temperatures, and speed. In addition they are enabling Goulston to deliver more sustainable solutions without compromising the performance demanded by the textile industry.

Fiber lubricants are not the only application for which Solayzme's high-oleic algal oils have been shown to improve product properties. In 2012, the Dow Chemical Co. signed a contingent off-take agreement with Solazyme for the use of this oil in its dielectric insulating fluids. The oil provides higher fire resistance and increased longevity along with a nontoxic alternative to mineral oils. The Archer Daniels Midland Company also conducted a frying study on these high-oleic oils for food applications. The company found that after 10 days of use Solazyme's high-oleic oil had more frying life left than the premium high-oleic canola oil had at the start of the test.

High-oleic oils are one of the first products of Solayzme's ability to customize solutions using a proprietary technology platform. Such a capability allows companies with oil needs to effectively address two challenges simultaneously. Not only are these forward-thinking companies establishing sustainable supply chains through the use of renewable resources, they are also doing so by improving the overall functionality of the products in their target markets. This opens the door for innovation where, at least in the case of fiber lubricants, an new class of products is now being developed for the global industry by Goulston Technologies.

Kevin Quon is an independent trader and author who writes for Seeking Alpha, an online platform for investment research that provides insight from investors and industry experts rather than sell-side analysts.

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SUPPLEMENT

A representative list of other companies involved in commercializing novel pathways to biobased commodity chemicals

A figure showing the complicated nature of fluxolipidomics

A complete reference list for biodegradable lamellar systems in skin care

More Extracts & Distillates

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REPRESENTATIVE LIST OF OTHER COMPANIES INVOLVED IN COMMERCIALIZING NOVEL PATHWAYS TO BIOBASED COMMODITY CHEMICALS[a]

Company	Feedstock[b]	Technology	Products	Web site
Amyris Inc.	Plant sugars	Microbial engineering, fermentation	Farnesene	www.amyris.com
BioAmber Inc.	Sugars	Yeast fermentation	Succinic acid, 1,4-butanediol	www.bio-amber.com
Calysta Energy, Inc.	CH_4	Enzymes, microorganisms	Liquid fuels, alcohols, esters, oxides, olefins	www.calystaenergy.com
Coskata, Inc.	CH_4, wood, trash, industrial gases	Syngas production followed by fermentation	Ethanol, ethylene, propanol	www.coskata.com
Evonik Industries AG Joint with OPXBIO	Syngas, vegetable oils, grains, sugars	Enzymes, microorganisms	2-Hydroxyisobutyric acid, methyl methacrylate, C_4 alcohols, DL-methionine, L-lysine, L-threonine, L-tryptophan, isophorone	www.evonik.com
Genomatica, Inc.	Sugars, biomass, syngas	Engineered microorganisms, fermentation	Butanediol; butadiene	www.genomatica.com
Gevo, Inc.	Sugars	Yeasts, fermentation	Isobutanol	www.gevo.com
Glycos Biotechnologies Inc. ("GlycosBio")	Crude glycerin, waste fatty acids, non-food sugars	Fermentation using engineered microorganisms	Isoprene, propanediols, acetone, lactic acid, ethanol, isopropanol, succinic acid	www.glycosbio.com
Greasoline GmbH	Non-food fats and oils; waste from palm oil biodiesel production	Catalysis based on activated carbon	High-cetane fuel; road fuels, jet fuels; C_1–C_4 gases, C_5–C_{18} liquid hydrocarbons	www.greasoline.com
LanzaTech NZ Ltd.	CO	Microbial fermentation	Ethanol, 2,3-butanediol	www.lanzatech.com
Mango Materials Inc.	Waste CH_4	Fermentation	Polyhydroxybutyrate	www.mangomaterials.com
Myriant Corp.	Sugars (sorghum or corn)	Anaerobic fermentation; bioengineered organisms	Succinic acid; acrylic acid, lactic acid, muconic acid; fumaric acid	www.myriant.com
Newlight Technologies LLC	CH_4 or CO_2	Engineered biocatalyst	Polyhydroxyalkanoate, resins	www.newlight.com
Next Fuels, LLC	Biomass (palm oil residues); sugarbeet residues, sugarcane bagasse	Hydrothermal liquefaction	Crude biofuel	www.nextfuels.com
OPX Biotechnologies. Inc. ("OPXBIO") Joint with Dow Chemical	H_2/CO_2 or sugar Corn sugar, sugarcane; cellulosic	Genetic manipulation of microbes	Fatty acids; renewable diesel, jet fuel Acrylic acid	www.opxbio.com
Siluria Technologies Inc.	CH_4	Catalyst formed by coating metal and metal oxide crystals onto a proprietary virus	Ethylene, gasoline blend stock	www.siluria.com
Verdezyne, Inc.	Plant oils, sugars	Engineered yeasts, fermentation	Adipic acid, dodecanedioic acid, sebacic acid	www.verdezyne.com

[a]Modified from a presentation by Andrew Soare of Lux Research (Singapore), "How novel materials and technologies will transform the bio-based industry through 2020," given November 21, 2013, at the Malaysian Palm Oil Board International Palm Oil Congress (PIPOC) held in Kuala Lumpur, Malaysia. . For more information, visit www.luxresearchinc.com," or use the email address, info@luxresearchinc.com.

[b]Syngas: hydrogen and CO (or CO_2). Biogas: CO_2 and CH_4.

A FIGURE SHOWING THE COMPLICATED NATURE OF FLUXOLIPIDOMICS

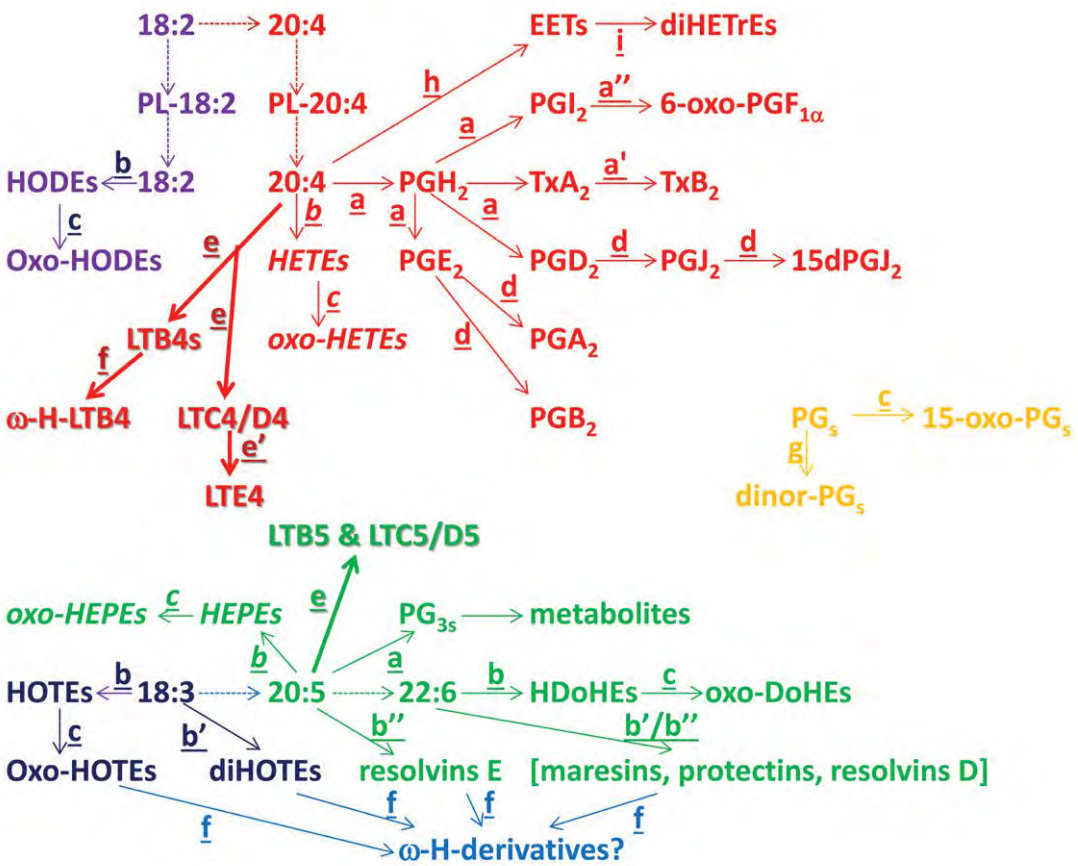

Figure: Main metabolic pathways from the major essential fatty acids linoleic (18:2), arachidonic (20:4), alpha-linolenic (18:3, eicosapentaenoic (20:5) and docosahexaenoic(22:6) acids. The different colors relate to different pathways. The metabolism of 20:5 is simplified as it is similar to that of 20:4.
PL: phospholipids; PG: prostaglandin; Tx: thromboxane; LT: leukotriene; HETE: hydroxyeicosatetraenoate; HEPE: hydroxyeicosapentaenoate; HODE: hydroxyoctadecadienoate; HOTE: hydroxyoctadecatrienoate; HDoHE: hydroxydocosahexaenoate; EET: epoxyeicosatrienoate.
Small letters next to the arrows indicate the enzymes or non-enzyme modifications (*e.g.* dehydation, hydrolysis) with different letters indicating different kinetics.
Accounting for the different kinetics, measuring as many metabolites as possible at different time points is promising to better approach phenotypes.

a: Cyclooxygenases & PGH isomerases
a' & a'': Hydrolysis of thromboxane A & prostacyclin (PGI_2)
b: Lipoxygenases
b' & b'': Double & triple lipoxygenation/hydroxylation
c: Hydroxy-dehydrogenases
d: Dehydration
e: Leukotriene synthases (LTA synthase + LTB & LTC/D synthases)
e': Dipeptidase (inactivation of LTD into LTE)
f: Omega hydroxylases
g: β-oxidation
h: Epoxygenases
i: Epoxide hydrolases

S4

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EXTRACTS & DISTILLATES

Model studies on the key aroma compounds formed by an oxidative degradation of ω-3 fatty acids initiated by either copper(II) ions or lipoxygenase

Hammer, M., and P. Schieberle, *J. Agric. Food Chem. 61*:10891–10900, 2013, http://dx.doi.org/10.1021/jf403827p.

Due to the high number of double bonds, ω-3-polyunsaturated fatty acids such as eicosapentaenoic aid (EPA) or docosahexaenoic acid (DHA) are prone to rapid oxidation, leading to the formation of intense taints often described as "fishy." To clarify the identity of the compounds responsible for such off-flavors, EPA, DHA, and α-linolenic acid (ALA) were oxidized singly either in the presence of copper ions or in the presence of lipoxygenase. The autoxidation of EPA and DHA led to a mixture of odorants eliciting an overall fishy odor quality, whereas neither the oxidation of ALA by copper ions nor that by lipoxygenase led to an unpleasant odor. Application of aroma extract dilution analysis (AEDA) on the volatiles generated by autoxidation of EPA revealed *trans*-4,5-epoxy-(*E,Z*)-2,7-decadienal, identified for the first time as a fatty acid degradation product, (Z)-1,5-octadien-3-one, (*Z*)-3-hexenal, (*Z,Z*)-2,5-octadienal, (*Z,Z*)-3,6-nonadienal, and (*E,E,Z*)-2,4,6-nonatrienal with the highest flavor dilution (FD) factors. The autoxidation as well as the enzymatic oxidation of all three acids led to the same odorants, but with different FD factors depending on the acid and/or the type of oxidation applied. Thus, the results suggested that a defined ratio of a few odorants is needed to generate a fishy off-flavor.

Membrane microdomains, rafts, and detergent-resistant membranes in plants and fungi

Malinsky, J., *et al., Annu. Rev. Plant Biol. 64*:501–529, 2013, http://dx.doi.org/10.1146/annurev-arplant-050312-120103.

The existence of specialized microdomains in plasma membranes, postulated for almost 25 years, has been popularized by the concept of lipid or membrane rafts. The idea that detergent-resistant membranes are equivalent to lipid rafts, which was generally abandoned after a decade of vigorous data accumulation, contributed to intense discussions about the validity of the raft concept. The existence of membrane microdomains, meanwhile, has been verified by unequivocal independent evidence. This review summarizes the current state of research in plants and fungi with respect to common aspects of both kingdoms. In these organisms, principally immobile microdomains large enough for microscopic detection have been visualized. These microdomains are found in the context of cell-cell interactions (plant symbionts and pathogens), membrane transport, stress, and polarized growth, and the data corroborate at least three mechanisms of formation. As documented in this review, modern methods of visualization of lateral-membrane compartments are also able to uncover the functional relevance of membrane microdomains.

Analysis of free phytosterols/stanols and their intact fatty acid and phenolic acid esters in various corn cultivars

Esche, R., *et al., J. Cereal Sci. 58*:333–340, 2013, http://dx.doi.org/10.1016/j.jcs.2013.07.011.

The contents and the compositions of free sterols/stanols, steryl/stanyl fatty acid esters, and steryl/stanyl phenolic acid esters were determined in 25 corn cultivars grown at the same location and in the same year. The lipid extracts obtained from the flours of whole kernels were subjected to solid-phase extraction for the isolation and separation of the sterol classes, followed by capillary gas chromatography. The study provides for the first time information on the variability of individual steryl/stanyl fatty acid esters in corn cultivars. The analysis revealed pronounced differences in total contents of each sterol class. The levels of free sterols/stanols ranged from 298 to 433 μg/g dry matter flour, those of steryl/stanyl fatty acid esters from 430 to 1187 μg/g, and those of steryl/stanyl phenolic acid esters from 46 to 240 μg/g. Free sterols/stanols were mainly composed of sitosterol and campesterol; up to 11% were made up of stanols. Steryl/stanyl esters of C18-fatty acids were dominating, with sitosteryl linoleate as the most abundant ester (44.7–53.5% of total fatty acid esters). Among the three investigated sterol classes, the distribution patterns of steryl/stanyl phenolic acid esters showed the greatest variability between the cultivars, even though *trans*-sitostanyl ferulate was the dominating ester in all samples.

High-resolution imaging mass spectrometry reveals detailed spatial distribution of phosphatidylinositols in human breast cancer

Kawashima, M., *et al., Cancer Sci. 104*:1372–1379, 2013, http://dx.doi.org/10.1111/cas.12229.

High-resolution matrix-assisted laser desorption/ionization imaging mass spectrometry (MALDI IMS) is an emerging application for lipid research that provides a comprehensive

CONTINUED ON NEXT PAGE

and detailed spatial distribution of ionized molecules. Recent lipidomic approach has identified several phospholipids and phosphatidylinositols (PI) that are accumulated in breast cancer tissues and are therefore novel biomarker candidates. Because their distribution and significance remain unclear, we investigated the precise spatial distribution of PI in human breast cancer tissues using high-resolution MALDI IMS. We evaluated tissues from nine human breast cancers and one normal mammary gland by negative ion MALDI IMS at a resolution of 10 μm. We detected 10 PI with different fatty acid compositions, and their proportions were remarkably variable in the malignant epithelial regions. High-resolution imaging enabled us to discriminate cancer cell clusters from the adjacent stromal tissue within epithelial regions; moreover, this technique revealed that several PI were specifically localized to cancer cell clusters. These PI were heterogeneously distributed within cancer cell clusters, allowing us to identify two different populations of cancer cells that predominantly expressed either PI (18:0/18:1) or PI (18:0/20:3). Tracing the expression level of PI during cancer cell progression suggested that the latter population is associated with the invasion. Our study documents a novel model for phospholipid analysis of breast cancer tissues by using high-resolution MALDI IMS and identifies candidate PI that can describe a specific phenotype of cancer cells.

Rice brans, rice bran oils, and rice hulls: composition, food and industrial uses, and bioactivities in humans, animals, and cells

Friedman, M., *J. Agric. Food Chem*. *61*:10626–10641, 2013, http://dx.doi.org/10.1021/jf403635v.

Rice plants produce bioactive rice brans and hulls that have been reported to have numerous health-promoting effects in cells, animals, and humans. The main objective of this review is to consolidate and integrate the widely scattered information on the composition and the antioxidative, anti-inflammatory, and immunostimulating effects of rice brans from different rice cultivars, rice bran oils derived from rice brans, rice hulls, liquid rice hull smoke derived from rice hulls, and some of their bioactive compounds. As part of this effort, this paper also presents brief summaries on the preparation of health-promoting foods including bread, corn flakes, frankfurters, ice cream, noodles, pasta, tortillas, and zero-*trans*-fat shortening as well as industrial products such bioethanol and biodiesel fuels. Also covered are antibiotic, antiallergic, anticarcinogenic, antidiabetic, cardiovascular, allelochemical, and other beneficial effects and the mechanisms of the bioactivities. The results show that food-compatible and safe formulations with desirable nutritional and biological properties can be used to develop new multifunctional foods as well as bioethanol and biodiesel fuel. The overlapping aspects are expected to contribute to a better understanding of the potential impact of the described health-promoting potential of the rice-derived brans, oils, and hulls in food and medicine. Such an understanding will enhance nutrition and health and benefit the agricultural and industrial economies.

Novel solid-phase extraction for epimer-specific quantitation of ergot alkaloids in rye flour and wheat germ oil

Köppen, R., *et al*., *J. Agric. Food Chem*. *61*:10699–10707, 2013, http://dx.doi.org/10.1021/jf403628q.

Ergot alkaloids and their epimer-specific determination have gained increasing importance for food safety. A solid-phase extraction and cleanup method based on sodium-neutralized strong cation exchange (Na^+-SCX) was developed to quantitate 12 priority ergot alkaloids in rye flour and wheat germ oil by HPLC [high-performance liquid chromatography] fluorescence analysis. Sample preparation is achieved by omitting acidic and alkaline conditions enabling minimized epimerization, which is necessary to determine ergot alkaloids according to their natural distribution in foods. Ergot alkaloids are eluted from SCX-column by forming ion pairs using a sodium hexanesulfonate-containing solution, which prevents epimerization for at least 96 h. Method validation yielded recoveries of 80–120% (rye flour) and 71–96% (wheat germ oil) with a maximum limit of quantitation (LOQ) of 2.0 μg kg^{-1} per ergot alkaloid for both matrices. The applicability of the developed method was demonstrated by analyzing 16 samples from German retail markets: 9 rye flours (max. 178 ± 5 μg kg^{-1}) and, reported for the first time, 7 wheat germ oils (max. 56.8 ± 2.7 μg kg^{-1}) expressed as the sum of 12 ergot alkaloids.

Cholesteryl ester species differently elevate plasma cholesterol in hamsters

Jiao, R., *et al*., *J. Agric. Food Chem. 61*:11041–11047, 2013, http://dx.doi.org/10.1021/jf4039293.

This study was to examine the effect of free cholesterol (C) and individual cholesteryl ester (CE) species, namely cholesteryl palmitate (CP), cholesteryl stearate (CS), cholesteryl oleate (CO), and cholesteryl linoleate (CL) on plasma total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), non-HDL-C, and triacylglycerols (TG) in hamsters. Results showed that addition of dietary CE species into diet at 0.1% differently raised plasma TC concentrations, with CO elevating plasma TC to 331 mg/dL, while CS raised plasma TC only to 220 mg/dL. It was found that CS was a poor substrate of pancreatic cholesterol esterase, while CO was a good substrate. The fecal analysis showed CS-fed hamsters had the highest fecal cholesterol concentration, while RT-PCR [real-time polymerase chain reaction] analysis found CS feeding was associated with down-regulations of intestinal Niemann-Pick C1 like 1 (NPC1L1) and acyl-CoA:cholesterol acyltransferase 2 (ACAT2) as well as microsomal triacylglycerol transport protein (MTP). It was therefore concluded that the plasma cholesterol-raising activity of CE species was partially governed by their hydrolysis rates in the intestine, and the relatively low raising activity associated with CS was

mediated by down-regulation of intestinal NPC1L1, ACAT2, and MTP.

Enzymatic synthesis of extra virgin olive oil based infant formula fat analogues containing ARA and DHA: one-stage and two-stage syntheses

Pande, G., *et al., J. Agric. Food Chem. 61*:10590–10598, 2013, http://dx.doi.org/10.1021/jf4036468.

Structured lipids (SL) with high palmitic acid content at the *sn*-2 position enriched with arachidonic acid (ARA) and docosahexaenoic acid (DHA) were produced using extra virgin olive oil, tripalmitin, ARA and DHA single cell oil free fatty acids. Four types of Ss were synthesized using immobilized lipases, Novozym 435, and Lipozyme TL IM, based on one-stage (one-pot) and two-stage (sequential) syntheses. The SL were characterized for fatty acid profile, triacylglycerol (TAG) molecular species, melting and crystallization profiles, tocopherols, and phenolic compounds. All the SL had >50 mol% palmitic acid at the *sn*-2 position. The predominant TAG in all SL were PPO and OPO [where P = palmitic and O = oleic]. The total tocopherol s of SL1-1, SL1-2, SL2-1, and SL2-2 were 70.46, 68.79, 79.64, and 79.31 μg/g, respectively. SL1-2 had the highest melting completion (42.0°C) and crystallization onset (27.6°C) temperatures. All the SL produced in this study may be suitable as infant formula fat analogues.

High-density lipoproteins: A consensus statement from the National Lipid Association

Toth, P.P., *et al., J. Clin. Lipidol. 7*:484–525, 2013, http://dx.doi.org/10.1016/j.jacl.2013.08.001.

For >4 decades it has been recognized that elevated serum levels of high density lipoprotein cholesterol (HDL-C) are associated with reduced risk of cardiovascular disease (CVD) and its sequelae. Many prospective observational studies performed around the world have confirmed an inverse relationship between HDL-C and cardiovascular risk in people irrespective of sex, race, or ethnicity. Consequently, it was assumed that, by extension, raising HDL-C through lifestyle modification and pharmacologic intervention would reduce risk of CVD. Animal studies are consistent with this assumption. Lipid treatment guidelines around the world promoted the recognition of HDL-C as a therapeutic target, especially in high-risk patients. Some *post hoc* analyses from randomized controlled trials also suggest that raising HDL-C beneficially affects the risk of CVD. However, a number of recent randomized studies putatively designed to test the "HDL hypothesis" have failed to show benefit. The results of these trials have caused many clinicians to question whether HDL-C is a legitimate therapeutic target. In response to the many questions and uncertainties raised by the results of these trials, the National Lipid Association convened an expert panel to evaluate the current status of HDL-C as a therapeutic target; to review the current state of knowledge of HDL particle structure, composition, and function; and to identify the salient questions yet to be answered about the role of HDL in either preventing or contributing to atherosclerotic disease. The expert panel's conclusions and clinical recommendations are summarized herein. The panel concludes that, although low HDL-C identifies patients at elevated risk, and much investigation suggests that HDL may play a variety of antiatherogenic roles, HDL-C is not a therapeutic target at the present time. Risk stratified atherogenic lipoprotein burden (low density lipoprotein cholesterol and non-HDL-C) should remain the primary and secondary targets of therapy in patients at risk, as described by established guidelines. The National Lipid Association emphasizes that rigorous research into the biology and clinical significance of low HDL-C should continue. The development of novel drugs designed to modulate the serum levels and functionality of HDL particles should also continue. On the basis of an enormous amount of basic scientific and clinical investigation, a considerable number of reasons support the need to continue to investigate the therapeutic effect of modulating HDL structure and function.

Current issues surrounding the definition of *trans*-fatty acids: implications for health, industry and food labels

Wang, Y., and S.D. Proctor, *Br. J. Nutr. 110*:1369–1383, 2013, http://dx.doi.org/10.1017/S0007114513001086.

The definition of *trans*-fatty acids (TFA) was established by the Codex Alimentarius to guide nutritional and legislative regulations to reduce TFA consumption. Currently, conjugated linoleic acid (CLA) is excluded from the TFA definition based on evidence (primarily preclinical studies) implying health benefits on weight management and cancer prevention. While the efficacy of CLA supplements remains inconsistent in randomized clinical trials, evidence has emerged to associate supplemental CLA with negative health outcomes, including increased subclinical inflammation and oxidative stress (particularly at high doses). This has resulted in concerns regarding the correctness of excluding CLA from the TFA definition. Here we review recent clinical and preclinical literature on health implications of CLA and ruminant TFA and highlight several issues surrounding the current Codex definition of TFA and how it may influence interpretation for public health. We find that CLA derived from ruminant foods differ from commercial CLA supplements in their isomer composition/distribution, consumption level, and bioactivity. We conclude that health concerns associated with the use of supplemental CLA do not repudiate the exclusion of all forms of CLA from the Codex TFA definition, particularly when using the definition for food-related purposes. Given the emerging differential bioactivity of TFA from industrial vs. ruminant sources, we advocate that regional nutrition guidelines/policies should focus on eliminating industrial forms of *trans*-fat from processed foods as opposed to all TFA *per se*.

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Chemometric tools to highlight possible migration of compounds from packaging to sunflower oils

Maalouly, J., *et al., J. Agric. Food Chem. 61*:10565–10573, 2013, http://dx.doi.org/10.1021/jf402888e.

Polyethylene terephthalate (PET) could be considered for the packaging of vegetable oils taking into account the impact of its oxygen permeability on the oxidation of the oil and the migration of volatile organic compounds (VOC) from the polymer matrix. After accelerated aging tests at 40°C for 10, 20, and 30 days, the headspace of three sunflower oils packed in PET with high-density polyethylene caps was carried out using solid phase microextraction. Volatile organic compounds such as benzene hydrocarbons, ethylbenzene, xylene isomers, and diethyl phthalate were identified in vegetable oils by gas chromatography coupled to mass spectrometry. Chemometric tools such as principal components analysis (PCA), independent components analysis (ICA), and a multiblocks analysis, common components and specific weight analysis (CCSWA), applied to analytical data were revealed to be very efficient to discriminate between samples according to oil oxidation products (hexanal, heptanal, 2-pentenal) and to the migration of packaging contaminants (xylene).

Accumulated phosphatidylcholine (16:0/16:1) in human colorectal cancer; possible involvement of LPCAT4

Kurabe, N., *et al., Cancer Sci. 104*:1295–1302, 2013, http://dx.doi.org/10.1111/cas.12221.

The identification of cancer biomarkers is critical for target-linked cancer therapy. The overall level of phosphatidylcholine (PC) is elevated in colorectal cancer (CRC). To investigate which species of PC is overexpressed in colorectal cancer, an imaging mass spectrometry study was performed using a panel of nonneoplastic mucosal and CRC tissues. In the present study, we identified a novel biomarker, PC(16:0/16:1), in CRC using imaging mass spectrometry. Specifically, elevated levels of PC(16:0/16:1) expression were observed in the more advanced stage of CRC. Our data further showed that PC(16:0/16:1) was specifically localized in the cancer region when examined using imaging mass spectrometry. Notably, because the ratio of PC(16:0/16:1) to lyso-PC(16:0) was higher in CRC, we postulated that lyso-PC acyltransferase (LPCAT) activity is elevated in CRC. In an *in vitro* analysis, we showed that LPCAT4 is involved in the deregulation of PC(16:0/16:1) in CRC. In an immunohistochemical analysis, LPCAT4 was shown to be overexpressed in CRC. These data indicate the potential usefulness of PC(16:0/16:1) for the clinical diagnosis of CRC and implicate LPCAT4 in the elevated expression of PC(16:0/16:1) in CRC.

Online LC–GC analysis of free sterols/stanols and intact steryl/stanyl esters in cereals

Esche, R., *et al., J. Agric. Food Chem. 61*:10932–10939, 2013, http://dx.doi.org/10.1021/jf403046z.

The suitability of online liquid chromatography–gas chromatography for the analysis of free sterols/stanols, steryl/stanyl fatty acid esters, and *trans*-steryl/stanyl ferulic acid esters in cereals is demonstrated. The silylated lipid extracts were fractionated via liquid chromatography on a normal phase, and the fractions containing the sterol classes were transferred online to the gas chromatograph for the analysis of their individual compositions. The study provides for the first time data on free sterols/stanols and intact steryl/stanyl esters in sweet corn, popcorn, and proso millet. Sweet corn revealed the highest contents of free sterols/stanols and steryl/stanyl fatty acid esters, and popcorn, in turn, the highest amounts of *trans*-steryl/stanyl ferulic acid esters. The distribution patterns of the proso millet samples revealed pronounced differences from those of sweet corn and popcorn as they particularly exhibited high proportions of free cholesterol and cholesteryl fatty acid esters. Furthermore, no *trans*-steryl/stanyl ferulic acid esters could be detected. ■