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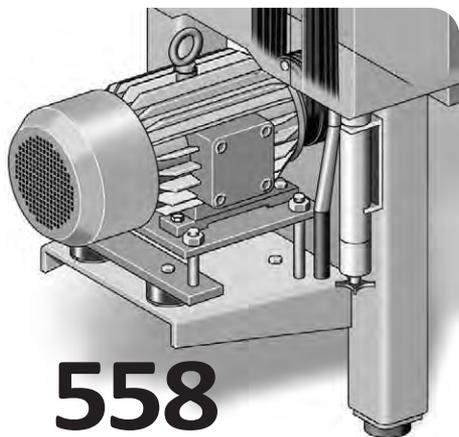
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International News on Fats, Oils, and Related Materials
ISSN: 0897-8026 IFRMEC 24 (9) 545-608
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Formerly published as *Chemists' Section*, *Cotton Oil Press*, 1917-1924; *Journal of the Oil and Fat Industries*, 1924-1931; *Oil & Soap*, 1932-1947; news portion of *JAACS*, 1948-1989. The American Oil Chemists' Society assumes no responsibility for statements or opinions of contributors to its columns.

Inform (ISSN: 0897-8026) is published 10 times per year in January, February, March, April, May, June, July/August, September, October, November/December by AOCS Press, 2710 South Boulder Drive, Urbana, IL 61802-6996 USA. Phone: +1 217-359-2344. Periodicals Postage paid at Urbana, IL, and additional mailing offices. **POSTMASTER:** Send address changes to *Inform*, P.O. Box 17190, Urbana, IL 61803-7190 USA.

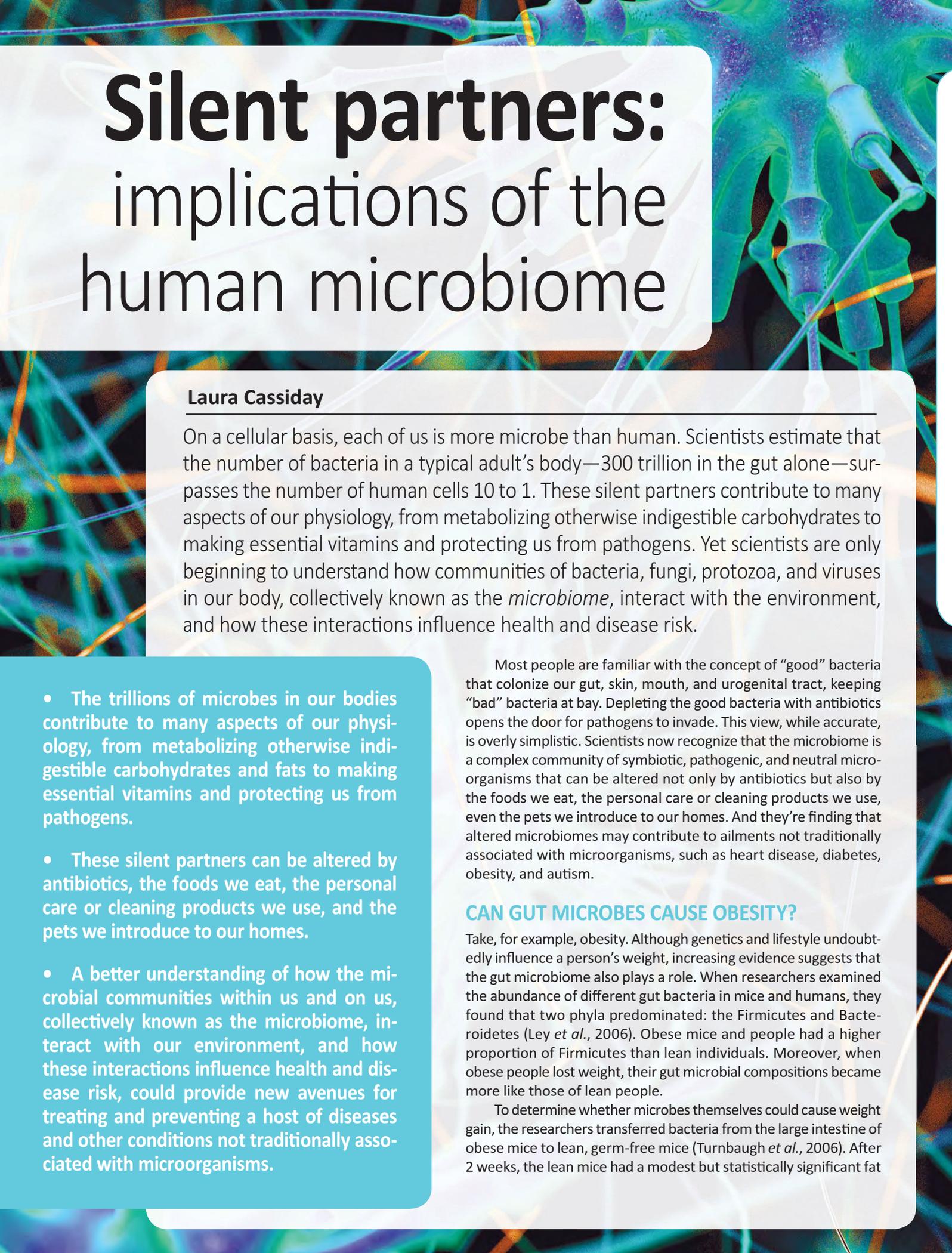
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Silent partners: implications of the human microbiome

Laura Cassiday

On a cellular basis, each of us is more microbe than human. Scientists estimate that the number of bacteria in a typical adult's body—300 trillion in the gut alone—surpasses the number of human cells 10 to 1. These silent partners contribute to many aspects of our physiology, from metabolizing otherwise indigestible carbohydrates to making essential vitamins and protecting us from pathogens. Yet scientists are only beginning to understand how communities of bacteria, fungi, protozoa, and viruses in our body, collectively known as the *microbiome*, interact with the environment, and how these interactions influence health and disease risk.

- The trillions of microbes in our bodies contribute to many aspects of our physiology, from metabolizing otherwise indigestible carbohydrates and fats to making essential vitamins and protecting us from pathogens.
- These silent partners can be altered by antibiotics, the foods we eat, the personal care or cleaning products we use, and the pets we introduce to our homes.
- A better understanding of how the microbial communities within us and on us, collectively known as the microbiome, interact with our environment, and how these interactions influence health and disease risk, could provide new avenues for treating and preventing a host of diseases and other conditions not traditionally associated with microorganisms.

Most people are familiar with the concept of “good” bacteria that colonize our gut, skin, mouth, and urogenital tract, keeping “bad” bacteria at bay. Depleting the good bacteria with antibiotics opens the door for pathogens to invade. This view, while accurate, is overly simplistic. Scientists now recognize that the microbiome is a complex community of symbiotic, pathogenic, and neutral microorganisms that can be altered not only by antibiotics but also by the foods we eat, the personal care or cleaning products we use, even the pets we introduce to our homes. And they're finding that altered microbiomes may contribute to ailments not traditionally associated with microorganisms, such as heart disease, diabetes, obesity, and autism.

CAN GUT MICROBES CAUSE OBESITY?

Take, for example, obesity. Although genetics and lifestyle undoubtedly influence a person's weight, increasing evidence suggests that the gut microbiome also plays a role. When researchers examined the abundance of different gut bacteria in mice and humans, they found that two phyla predominated: the Firmicutes and Bacteroidetes (Ley *et al.*, 2006). Obese mice and people had a higher proportion of Firmicutes than lean individuals. Moreover, when obese people lost weight, their gut microbial compositions became more like those of lean people.

To determine whether microbes themselves could cause weight gain, the researchers transferred bacteria from the large intestine of obese mice to lean, germ-free mice (Turnbaugh *et al.*, 2006). After 2 weeks, the lean mice had a modest but statistically significant fat

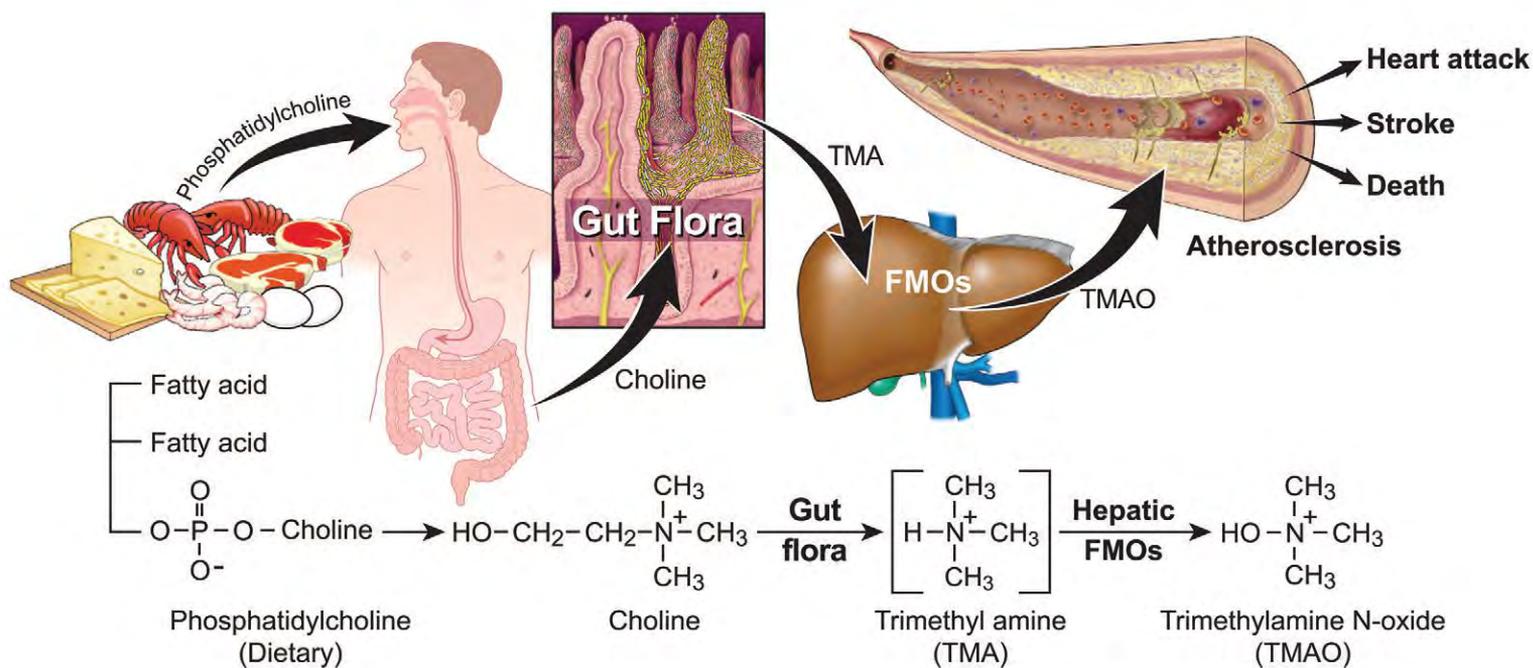


FIG. 1. Microbes may contribute to atherosclerosis by metabolizing dietary choline into TMA, which liver enzymes then convert to the atherosclerosis-promoting compound TMAO. Abbreviations: TMA, trimethylamine; TMAO, trimethylamine-N-oxide; FMO, flavin monooxygenase. Reprinted by permission from Macmillan Publishers Ltd: I (Wang, Z., et al., Nature 472:57-63, 2011).

gain and were able to extract more calories from their food. As it turns out, the microbiome of obese mice contains an abundance of enzymes for breaking down indigestible polysaccharides, increasing the efficiency of energy harvest and possibly fueling weight gain.

The involvement of the microbiome in a growing list of diseases suggests new avenues for therapy. For years, yogurt companies have touted the ability of live *Lactobacillus* cultures in their products to ease digestive ailments. Indeed, a few studies have shown that yogurt can improve symptoms of inflammatory bowel disease.

Yet yogurt is limited in the species of bacteria that it can introduce, and the microbes within yogurt often don't survive the acidity of the stomach. In contrast, fecal transplants—which involve transferring a small amount of feces from a healthy donor into the intestine of the recipient by enema or duodenal infusion—aim to repopulate entire bacterial ecosystems. The procedure has already proven effective for curing gastrointestinal infections caused by the bacterium *Clostridium difficile* (van Nood *et al.*, 2013), which are difficult to treat because of the microbe's resistance to common antibiotics. More surprisingly, fecal transplantation from lean donors increased the insulin sensitivity of obese recipients with metabolic syndrome (Vrieze *et al.*, 2012), a condition that is characterized by obesity and insulin resistance and that is a precursor of diabetes.

Researchers are now investigating fecal transplants for the treatment of a host of conditions, including obesity, diabetes, and Parkinson's disease. However, this crude approach does have limitations. In addition to the "yuck" factor, the general safety and long-term effects of transferring fecal matter from one person to another are still unknown.

AS UNIQUE AS A FINGERPRINT

A more refined approach to treating disease will require a greater understanding of the "normal" human microbiome. However, this task is complicated by the variability of microbiomes among people—evidence so far suggests that a person's microbiome is as unique as his or her fingerprint. In 2008, the US National Institutes of Health (NIH) launched the Human Microbiome Project, a \$157 million effort to catalog the microbial species that inhabit humans.

To establish a reference human microbiome, researchers in the Human Microbiome Project Consortium sampled microbes from 242 healthy US volunteers (129 male, 113 female) at different sites of the body, including the nose, mouth, skin, lower intestine (stool), and urogenital tract. The researchers purified DNA from the samples, sequenced it with high-throughput

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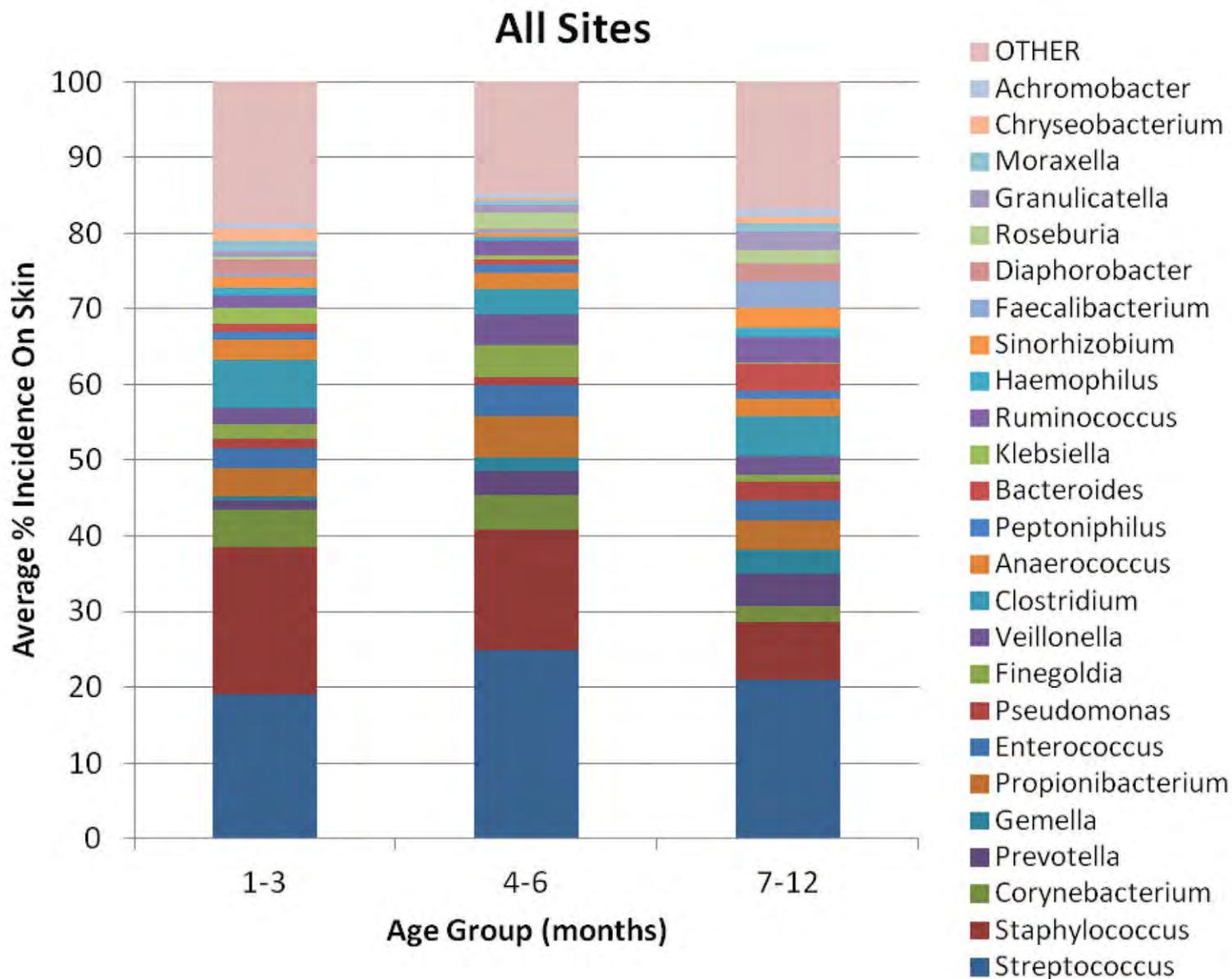


FIG. 2. Relative abundance of the most predominant genera in the various age groups, averaged over all body sites. Reprinted with permission from Capone, K.A., et al., *Diversity of the skin microbiome early in life*, *Journal of Investigative Dermatology* 131:2026–2032, 2011.

genome sequencing, and identified microbial species from the sequence of their 16S ribosomal RNA.

The analysis allowed researchers to define the boundaries of normal microbial variation in humans, which experts now say encompasses more than 10,000 microbial species. As expected, microbiomes differed substantially among people, and among body sites within the same person (Human Microbiome Project Consortium, 2012). However, metabolic functions were generally conserved. For example, each person had a population of bacteria in the gut to help digest fats, but

the bacterial species varied among individuals. Interestingly, almost all of the healthy volunteers carried pathogens in their microbiome that caused no disease, presumably because they were held in check by beneficial microbes. The reference microbiome has provided a foundation for researchers to explore under what conditions these pathogens turn deadly and how the human microbiome changes with disease.

According to Lisa Helbling Chadwick, program administrator at the National Institute of Environmental Health Sciences (NIEHS) in Research Triangle Park, North Carolina (USA), char-

acterizing the “normal” human microbiome will also enable researchers to determine how environmental exposures, such as chemical spills or industrial waste, alter it. “You can imagine that environmental chemicals change our microbiome in a similar way to antibiotics or diet, but there have been very few studies examining this question,” says Chadwick. Later this year, the NIEHS will award \$2 million in grants to fund the study of environmental influences on the microbiome.

CONTAMINANTS, TOXINS, AND OTHER ENVIRONMENTAL INFLUENCES

A recent study supports the idea that chemicals in the environment can perturb the microbiome (Choi *et al.*, 2013). Polychlorinated biphenyls (PCBs) are environmental contaminants that were widely used in dielectric and cooling fluids prior to their ban in the United States in 1979. PCBs can cause cancer in animals and humans; and because of their relatively long half-life (8–15 years), some PCBs still linger in the environment. When researchers fed a mixture of PCBs to mice, they detected a significant decrease in the abundance of Proteobacteria in the gut. The researchers speculate that this altered microbiome may compromise the mouse immune system and contribute to some symptoms of PCB poisoning.

Mice allowed to exercise on a running wheel for five weeks prior to PCB exposure were protected from the PCB-induced changes in their microbiomes. The researchers hypothesized that exercise may cause certain compounds to be excreted into the gut or cause changes in the intestinal immune system that help preserve the microbial status quo. “This study provides hope that we might be able to mitigate some of the effects of environmental exposures with behavioral changes,” says Chadwick.

Other evidence suggests that our microbiome could influence our response to environmental toxins. For example, arsenic enters the environment from both natural and human sources. Some species of bacteria in the guts of rodents and humans can metabolize and chemically modify ingested arsenic, making it more or less toxic and bioavailable (Pinyayev *et al.*, 2011; Alava *et al.*, 2012). Therefore, it’s possible, although as yet unproven, that a person’s unique microbiome could make them more or less susceptible to arsenic poisoning.

Emerging knowledge of the microbiome could also improve our understanding of the environmental origins of disease. “As we learn more about how chemicals and the microbiome interact, we might gain new insights into things like how early life exposures can cause diseases later in life,” says Chadwick.

If, as some researchers suspect, a person’s microbiome influences his or her response to toxic chemicals and vice versa, an overhaul of toxicological testing may be in order. Yet the US Environmental Protection Agency (EPA) remains confident in its current process for toxicological risk assessment. In a statement provided to *Inform*, the EPA said, “Research into

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the microbiome promises to add much to our understanding of the toxicological processes that lead to human disease, but traditional methods have been and will continue to be important. We have learned much from traditional toxicity testing, and research into new areas like the microbiome will expand and complement, not negate, what we can still learn from continued toxicity testing of chemical agents.”

INCREASED MODERNIZATION

To study how the human microbiome changes with increasing modernization, Maria Gloria Dominguez-Bello, associate professor of biology at the University of Puerto Rico, in San Juan, and her colleagues analyzed the microbiomes of people living deep in the Amazon hinterlands and in rural and urban settings. The researchers collected samples from the mouth, skin, and feces of 109 subjects: 70 Amerindians living in two remote villages in the Peruvian Amazon, 29 people living in a rural village by the Amazon River in Peru, and 20 subjects from San Juan, Puerto Rico as an example of a “Westernized” population at similar latitude.

The researchers found greater microbial diversity for the feces and skin of people living in the isolated Amazonian villages than in the rural and urban areas (unpublished results). The feces of people living in the remote villages contained the bacterial genera *Prevotella* and *Succinivibrio*, which were

undetectable in the feces of urban subjects. The participants living in Puerto Rico had a predominance of *Bacteroides* in their feces, similar to what has been observed for city dwellers in the United States (Yatsunenکو *et al.*, 2012).

According to Dominguez-Bello, these findings suggest that microbial diversity decreases with increased modernization. She also notes that most human microbiome studies involve Westernized subjects, which may not paint a complete picture of human microbiome diversity.

THE SKIN MICROBIOME

Although scientists don’t fully understand the consequences of reduced microbial diversity, the “hygiene hypothesis” posits that limited exposure to bacteria in early childhood could prime the immune system to overreact to harmless antigens such as pollen or peanuts. If true, the hygiene hypothesis could help explain the alarming rise in allergies and inflammatory diseases in recent decades.

To find out how cleansers and beauty products affect the skin microbiome, some large companies in the personal care industry are launching their own microbiome-related studies. Kimberly Capone, research manager and fellow at Johnson & Johnson Consumer Companies, Inc., with headquarters in New Brunswick, New Jersey, USA, is spearheading a skin microbiome project.



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In 1953, Johnson & Johnson pioneered the development of gentle cleansers with the introduction of JOHNSON'S® Baby Shampoo, the first product as mild to a baby's eyes as pure water. "As we continue to innovate in the area of mildness, we extensively study the effects of cleansers and their impact on skin, including the skin microbiome," says Capone.

Initial microbiome-related research at Johnson & Johnson has focused on understanding how an infant's skin microbiome develops and changes over time. In a study published in the *Journal of Investigative Dermatology*, Capone and her colleagues analyzed bacteria from three different skin sites (arm, buttock, and forehead) in 31 healthy infants, ranging in age from 3 to 52 weeks (Capone *et al.*, 2011).

The researchers found that the skin microbiome evolves over the first year of life, showing increasing diversity with age (Fig. 2, page 552). While staphylococci were prevalent on the skin of the youngest infants, their relative abundance decreased with time as the abundance of other genera increased. In contrast to that of adults, infant skin was colonized predominantly by bacteria from the phylum Firmicutes, of which *Staphylococcus* is a member. The researchers are now extending their study to evaluate the development of the skin microbiome throughout childhood.

Knowledge of the microbiome could help product formulators combat skin inflammation, which some bacterial species can modulate. In addition, a healthy skin microbiome could deter colonization by disease-causing species, such as the *Propionibacteria* that cause acne. Manufacturers may even start to rethink the preservatives they use to curb microbial growth in skin care products, as they could upset the natural balance of the skin microbiome.

Understanding the skin microbiome also opens the door for probiotic and prebiotic skin care products. By adding a mixture of beneficial bacterial species (or compounds that promote the growth of these species) to beauty products, product formulators may be able to reconstitute the optimal skin microbiome. However, the diversity of the human microbiome could preclude a "one-size-fits-all" approach. "One day, scientists may be able to create a product that is customized based on a person's microbial fingerprint," says Capone.

THE REAL BEEF WITH BEEF

Diet is another lifestyle factor that has the potential to affect our microbiomes. For example, vegetarians have different gut microbiomes than meat eaters. Stanley Hazen, chair of the Department of Cellular and Molecular Medicine at the Cleveland Clinic (Ohio, USA), discovered that gut microflora can contribute to cardiovascular disease by metabolizing dietary phosphatidylcholine (PC). PC, a major component of cell membranes, is naturally abundant in meat and egg yolk. Also known as lecithin, PC is commonly added to foods as an emulsifier.

Hazen and his colleagues didn't set out to study gut microflora. Instead, they wanted to identify metabolites in the blood of patients that could predict cardiovascular disease. The researchers used mass spectrometry to compare metabolites in archived

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blood plasma from an initial 50 people who experienced a heart attack, stroke, or death within three years of plasma collection vs. 50 age- and gender-matched people who did not (Wang *et al.*, 2011).

Much to the researchers' surprise, the metabolite that most strongly predicted cardiovascular risk was one that they'd

CONTINUED ON NEXT PAGE

never heard of before: trimethylamine *N*-oxide, or TMAO. When Hazen surveyed the literature, he discovered that some gut microflora can cleave a trimethylamine (TMA) group from dietary PC. Then, flavin monooxygenases in the liver convert TMA to TMAO.

When Hazen's team fed TMAO directly to mice, the metabolite promoted the deposition of cholesterol in artery walls and the formation of atherosclerotic plaques. Importantly, feeding choline to mice only promoted heart disease if the mice had intact intestinal flora: Mice treated with broad-spectrum antibiotics prior to the feeding did not make TMAO or go on to develop atherosclerosis (Fig. 1, page 551).

In a related study, Hazen and his colleagues showed that L-carnitine, a nutrient abundant in red meat, also promotes atherosclerosis through a microbiome-mediated mechanism (Koeth *et al.*, 2013). L-Carnitine and PC share similar structures, and gut microflora can metabolize both compounds into TMA. When the researchers fed mice chow supplemented with L-carnitine, the mice produced more TMA and TMAO and had increased atherosclerosis. However, these effects did not occur when the mice were treated with antibiotics. These results suggest that the intestinal microbiome may contribute to the well-established link between high red meat consumption and cardiovascular disease risk.

This study also provided evidence that a diet high in L-carnitine shifts the gut microbial composition to favor TMA-producing species, further increasing a person's risk of developing atherosclerosis. When human subjects were asked to eat a sirloin steak together with an L-carnitine supplement, regular red meat eaters produced more TMAO than vegetarians or vegans (Koeth *et al.*, 2013). Antibiotics suppressed TMAO production. Analysis of fecal microbial compositions revealed bacterial genera that were significantly associated with both meat-eating status and TMAO levels.

Hazen and his colleagues recently confirmed the association between TMAO and cardiovascular disease risk in an independent cohort of 4,007 people. Blood TMAO levels strongly predicted the risk of heart attack, stroke, and death over the ensuing 3-year period (Wilson Tang *et al.*, 2013). Hazen thinks that in the future, TMAO will be a useful biomarker to screen people in the clinic for elevated cardiovascular risk. "The risk factors we currently use have room for improvement," he says. For example, people with high low-density lipoprotein (LDL) cholesterol don't always develop heart disease, whereas some people with normal LDL cholesterol levels do.

According to Hazen, the association of TMAO with atherosclerosis helps explain this once-puzzling observation. Studies in mice have shown that TMAO enhances the rate of cholesterol transport into the artery wall, while simultaneously reducing the level of reverse transport. "The net effect is more cholesterol in the artery, even though LDL levels themselves are not changing," says Hazen. "This helps explain how two people with the same LDL cholesterol level can have different risks of developing heart disease."

If TMAO-producing microbes contribute to atherosclerosis, then selectively killing these species could help prevent or treat the disease. However, Hazen cautions that antibiotics are not the answer. Although antibiotics initially suppressed TMAO pro-

duction in mice, within several months the bacteria developed resistance. Treating patients with several antibiotics simultaneously may avoid resistance, but obliterating the microbiome over the long term would likely cause other serious health problems.

THE PERFECT MICROBE COCKTAIL

As researchers continue to catalog microbiomes from normal and disease states, makers of probiotic supplements will try to develop blends of microbes that combat diseases by replacing harmful bacteria with beneficial ones. Yet Hazen is skeptical of this approach. "The idea that we're going to find the perfect cocktail of microbes based on analyzing DNA in the stool is extremely simplistic," he says.

Hazen notes that the intestines are more than 40 feet (12 meters) long, and the microbial composition varies in every nook and cranny. "Looking at the bacteria in feces is like looking at the rubble from the World Trade Center and trying to figure out what was in the janitor's closet on the 63rd floor," says Hazen. "To say that we're going to come up with this perfect blend of microbes that you can ingest and repopulate the janitor's closet is probably not realistic." Other challenges of a probiotic approach include delivering the bacteria to the proper location and keeping them alive while passing through the acidic environment of the stomach.

A more practical approach would be targeting specific bacterial enzymes or receptors with drugs, Hazen says. "This is how we've always treated diseases, but until now we've focused on *Homo sapiens* enzymes. I think our future is going to be targeting bacterial enzymes." Unlike antibiotics, this new class of drugs wouldn't kill microbes, largely avoiding the issue of resistance, but would instead inhibit specific biochemical pathways.

For example, researchers might identify a compound that blocks the synthesis of trimethylamine by gut bacteria to prevent or treat cardiovascular disease. "Pharmaceutical companies already know how to drug a pathway," says Hazen. "In contrast, repopulating a whole organism with a bacterial community in the appropriate niche is an extremely daunting thing to do."

The invention of high-throughput DNA sequencing and approaches to identify metabolites in the body has only recently enabled scientists to probe the genetic and metabolic diversity of the microbiome. As such, knowledge of the microbiome and the consequences of disrupting it are still in its infancy. With time, perhaps we can learn how to better treat the other 90% of our body (the microbes), which until now has been neglected and even reviled.

In the 20th century, the advent of antibiotics and better hygiene dramatically improved human life expectancy and quality of life. Ironically, sustaining these improvements in the 21st century may require a return to a more bacteria-friendly approach. Learning how the human microbiome changes in response to our environment, diet, and disease will guide efforts to peacefully coexist with our silent partners.

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SBOG in 20 Years

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The Brazilian Society of Fats and Oils (“Sociedade Brasileira de Óleos e Gorduras” – SBOG) are organizing the SBOG’s 20th Anniversary Commemoration Event: **“SBOG 20 Years: New horizons for the Science and Technology of Oils and Fats”**, from **12 to 14 November, 2013** at the Convention Center Hotel Torres da Cachoeira, Florianópolis, SC, Brazil.

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Special Report on Aqueous Oil Extraction

Marguerite Torrey and Joseph Doolen

Olive oil extraction, carried out in the traditional manner, is the oldest green technology used to isolate edible vegetable oil. It is also the earliest instance of an oil extraction carried out with water.

- Olive oil has been produced through an aqueous oil extraction process for millennia. However, olive oil today constitutes only about 2% of commercial edible oil production, so the question arises: What other commodity oils can be extracted via a green, hexane-free, aqueous-based process?

- Roughly a third of the world's edible oil comes from the fruits of the oil palm, which are processed using technologies based on water to produce palm oil. In the past two decades, technology based on aqueous processes has been developed to produce culinary avocado oil. But applying such technology to seeds is more difficult because the seeds on which much of the world's vegetable oil trade is based have a high protein content. This leads to emulsion formation, making separation of oil from the aqueous phase difficult.

- This special report highlights advances in aqueous extraction of olive (this page), palm (559), avocado (560), soybean (593), corn (595), and camellia (599) oil, as well as in China (601) and using surfactants (604).

Historical evidence shows that olive oil production began about 2000 BCE (Amouretti, 1996). For centuries the basic batch processes remained unchanged—harvesting, crushing, and pressing the fruits at ambient temperatures to release the oil from the plant cells, and then decanting the oil from the vegetable water that was simultaneously extracted from the fruits.

Modern virgin olive oil production qualifies as a green, sustainable technology because extraction is carried out at or near ambient temperatures (cold-pressed). No heating is applied during the process, so no resources have to be expended for that. No solvent other than water is used in removing the oil from the plant tissues. And since the trees are perennials, their continuing presence helps to tie down the soil and lessen erosion for periods of years.

MODERN AQUEOUS OLIVE OIL EXTRACTION

Olives, which have a water content of 50% (Boskou, 2006), and oil palm fruits, which have a water content of perhaps 30% when they are ripe, are the primary fruits being extracted today for their oil. In modern olive oil processing, olives are collected, cleaned to remove leaves and twigs and other extraneous materials, and then washed. If the olives are to be processed batch-wise, they will likely be moved to the crushing step almost immediately. Otherwise, the olives are susceptible to rapid oxidative degradation if they are stored for more than a short time.

Larger mills are likely to run continuously. To ensure that enough cleaned olives are available to run for 12–24 hours a day without a break, the cleaned olives may be held for a period of time in stainless steel hoppers until they are fed into a continuous crushing system.

Hammer mills are commonly used in modern mills to crush the olives. They are especially well suited for reducing the heterogeneous mixture of soft fruit flesh and hard pits. The mills rupture the cellular and vacuolar membranes of the fruits, freeing the oil droplets inside. The mixture of fruit flesh and pits remains in the reduction zone until the particles in the resultant pulpy mass have been reduced to a predetermined size.

The olive pulp is then pumped to malaxers, or mixers. There, as the pulp is turned over and over for 60–90 minutes at under

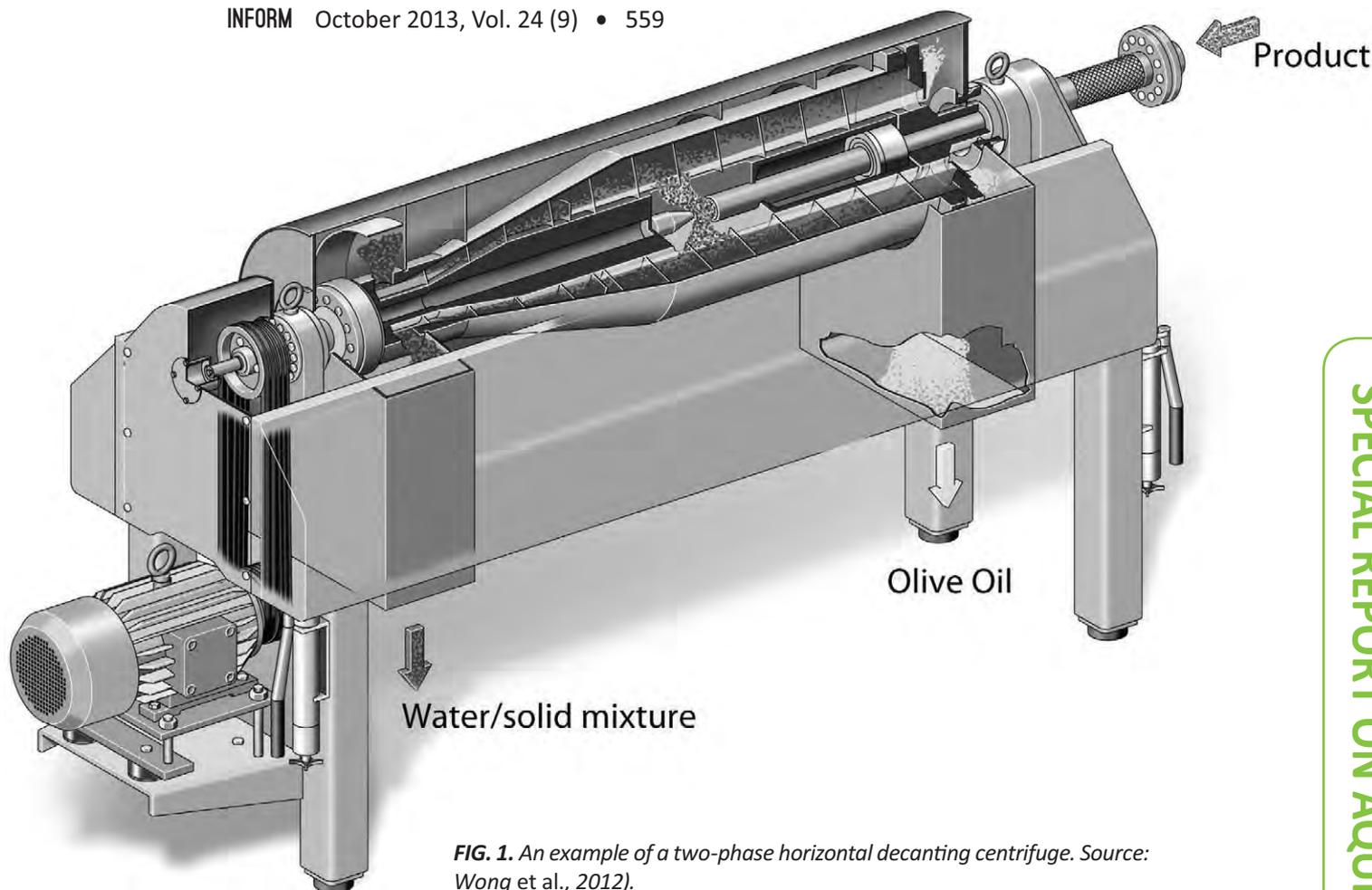


FIG. 1. An example of a two-phase horizontal decanting centrifuge. Source: Wong et al., 2012).

30°C, the freed oil gradually agglomerates into droplets having a wide range of sizes (Petrakis, 2006). These are in contact with the water released from the cells as well as any residual water from the washing step. Industrial enzymes can be added during malaxing to increase oil yield and enhance recovery of phenolic content of the final product (Chih *et al.*, 2012).

The equipment that defines modern aqueous oil extraction is the horizontal centrifuge decanter, first introduced by the Swedish company Alfa Laval in 1970 (Alba *et al.*, 2011). The two-phase horizontal centrifuge “decanter consists of a cylindrical-conical rotating bowl and a helical hollow-axis screw rotating coaxially inside it and at a slightly different speed to the bowl” (Fig. 1).

As malaxed olive paste is injected continuously into the two-phase horizontal centrifuge decanter through an opening in the hollow screw axis, water is added to the mix. Centrifugal force presses the heavy wet olive solids (pomace) against the wall, and they are slowly moved toward the end by the difference in rotation speed between the auger and the bowl. The lighter oil layer forms a ring nearest to the centrifuge axis, and is drained at the opposite end of the device.

(A so-called three-phase horizontal centrifuge decanter—which separates the paste into pomace, water, and oil—is also used commercially. Its greatest disadvantage compared with a two-phase system is that it generates water with a much higher contaminant level that requires greater pretreatment before it can be disposed of.)

The olive oil exiting the two-phase horizontal decanting centrifuge is actually a mixture, containing mostly oil and some water, together with fine particles of olive-flesh. The three phases can be separated by natural settling followed by decantation, centrifugation, or a combination of both process. Sieving through a vibrating screen to remove solids; passage of the oil-water mixture through a vertical centrifuge, and a final settling or decantation process to remove the air added to the oil during centrifugation are widely used processes before sending the oil to storage.

AQUEOUS PROCESSING OF PALM OIL PRODUCTS

The history of human use of palm oil may run as far back as Egypt 7000 years ago. European explorers and traders sailing along the coast of West Africa from 1400 A.D. onward found palm groves being harvested, and the Portuguese began to trade in palm oil around 1506 A.D.

Traditional extraction methods for palm oil were labor intensive and resulted in poor recovery and poor quality of the oil products. They included either pounding the mesocarp (the soft tissue between the skin and the kernel of the fruit) or else treading bunches that had been split, sometimes boiled, and left to soften for several days. Expelled oil was separated from the

CONTINUED ON NEXT PAGE



FIG. 2. Freshly processed avocado oil. Photo courtesy of A. Logan, Avocado Health Limited, New Zealand/Mexico.

resultant mash by skimming. The kernels were then picked out by hand, dried, and hand-cracked for subsequent production of palm kernel oil.

The first power-operated mills were constructed in Africa before 1914, but oils mills similar to those used today did not appear until the late 1960s.

Present-day oil palm milling processes recapitulate earlier processes, using both physical methods to segregate the kernels from the mesocarp and heat to hasten the processing. Harvested fresh fruit bunches (FFB) need to be processed quickly to minimize the formation of free fatty acids (FFA) that adversely affect oil quality. To this end, bunches are steamed for 60–90 minutes at 140°C and 3 kg/cm² pressure to stop the increase of FFA and make it easier to strip the fruits from the bunches (MPBO, 2012). Steaming also pre-conditions the kernels to prevent their breaking during subsequent pressing and nut cracking.

After heating, the FFB are fed to a rotary drum thresher, where the fruits are separated from the spikelets on which they grew in the bunch. The fruits are collected (the empty fruit bunches are segregated for other uses) then reheated with live steam to loosen the mesocarp from the kernels. The fruits are mashed, and heating ruptures the oil-bearing cells of the mesocarp.

Most commonly, palm oil is obtained from the digested fruits by pressing using twin screw presses (MPOB, 2012). Hot water is added to enhance the flow of oil when necessary. The oil slurry is collected in the crude oil tank for purification, and the fiber and nuts go to the depericarper for separation. At this point, the crude palm oil from the presses contains 35–45% palm oil, 45–55% water, and varying proportions of fibrous materials.

The oil is then sent to a vibrating screen of 30–40 mesh to remove the major part of the fiber and residue. It is then pumped to a clarification tank, held at about 90°C, for oil separation. The

clarified oil then passes through a high-speed centrifuge to remove impurities from the oil. The oil is dried under vacuum, cooled to about 45°C, and sent to storage.

AVOCADO

As the popularity of avocado fruits for edible purposes has increased in the past few decades, so has global production. Growers and shippers seek to market only the best fruits, so as a corollary, the amount of fruit rejected from the fresh fruit trade has also increased. Growers have looked for alternative uses for discarded fruit—and discovered a burgeoning interest in using avocado oil extracted from the fruits for culinary and cosmetic purposes.

Marie Wong of Massey University, Auckland, New Zealand, pointed out: “The oil is in the flesh of the fruit; the flesh contains a relatively high percentage of water compared to seeds and pulses. Hence, the aqueous extraction . . . is more suitable for these oils,” which have a water content of 65% (Woolf *et al.*, 2009).

Culinary properties. Avocado oil for use in cooking resembles olive oil. Since both oils are extracted by physical means with little or no heating, they both retain the green color of the fruits they come from (Fig. 2). At least 60% of the fatty acids in the oil are monounsaturated (oleic and palmitoleic), and about 10% are polyunsaturated, making for a healthful oil. According to Andrew Logan, of AvoHealth (www.avohealth.com), avocado oil has “a high lutein content [important in eye health] and very high β -sitosterol content.”

α -Tocopherol, an antioxidant, is the major form of vitamin E found in avocado oil. The levels in avocado oil are similar to those in olive oil (0.1–0.4 mg g⁻¹), and the presence of this antioxidant helps extend the shelf life of both oils.

The smoke point of avocado oil is high (>250°C), indicating its suitability for frying. The oil from the ‘Hass’ cultivar, the most popular avocado, has been described as having the flavor of avocados, with a grassy and butter/mushroom-like component, whereas the ‘Fuerte’ cultivar produces oil with more mushroom and less avocado flavor (Wong *et al.*, 2010).

In comparing the cold-pressing procedures used to produce avocado oil with those for olive oil, there are several differences. For instance, seeds are not removed from olives before processing; their removal is essential for avocados. Water may need to be added to avocado flesh during malaxing to lower the viscosity. And Wolff and coworkers initially found that adding enzymes such as pectinase during malaxation improved oil yields by 5–40% in early-season fruits. Enzymes made no difference in mid-season avocados.

The top avocado producer in the world is Mexico (1.26 million metric tons in 2011). Mevi Avocados, one of Mexico’s

**SPECIAL REPORT
CONTINUES ON PAGE 592**

AOCS congratulates the Solvent Extractors' Association of India, which recently celebrated the 50th anniversary of its founding.

■■■

In late July 2013, the Chemical Safety Board (CSB) characterized the US Occupational Safety and Health Administration's (OSHA) response to seven recommendations CSB had made involving worker-safety standards—including the OSHA standard on combustible dust that has been in process since 2009—as “unacceptable.”

■■■

The Canadian government will provide \$15 million in new research funding for that country's canola industry; when combined with industry contributions, the total investment in research and innovation will be more than C\$20 million (about \$19 million) over five years. According to the Canadian Canola Council, projects scheduled to be funded involve oil and canola meal nutrition, integrated pest management, yield and quality optimization, sustainable production, supply surveillance and forecasting, and technology transfer.

■■■

DuPont Pioneer is constructing a regional research center in Delmas, a small farming town east of Johannesburg, South Africa. The facility is scheduled to be completed by 2017, according to a report by Albawaba.com. “The technology hub in South Africa would be similar to others the unit has established in Brazil, India, and China,” the online news site said.

■■■

On August 1, 2013, US President Barack Obama signed an executive order instructing key federal agencies to work together to reduce the risks of hazardous chemicals to workers and communities. (See <http://tinyurl.com/chem-safety-order>.)

■■■

Effective August 13, 2013, Health Canada's Food Directorate approved the use of gelatin at a maximum level of 1.0% in calorie-reduced margarine. Gelatin already was allowed as a gelling agent in unstandardized foods at levels “consistent with Good Manufacturing Practice (GMP)” as well as in a variety of standardized foods at levels consistent with GMP, or up to 0.75%. ■

NEWS & NOTEWORTHY



Shortage leads to green route to olefins

A shortage of acrylic acid attributed to its increased use by coatings manufacturing in India and China—and for the superabsorbent polymers in disposable diapers—has led to a green route to glycerol and long-chain olefins via ultraviolet irradiation of fats and vegetable oils.

Acrylic acid—a petroleum-based product with few available biobased alternatives—is an extremely versatile chemical feedstock. Used either on its own or after esterification with various alcohols, acrylic acid is the base for acrylic polymers used in paints, coatings, detergents, sealants, adhesives, thickeners, emulsifying agents, dispersing agents, and the like. About four million metric tons of acrylic acid are produced annually, according to an industry source, with a value of \$10 billion; demand is expected to increase.

“Our original interest was in the glycerol and its degradation products,” explains

Douglas C. Neckers, the lead researcher and president/chief executive officer (CEO) of Biosolar LLC in Millbury, Ohio, USA. His study appeared in *ACS Sustainable Chemistry & Engineering* (doi:10.1021/sc400135y, 2013). “We've only just started down the pathway.”

Neckers and co-author Maria Muro-Small investigated a number of vegetable oils and animal fats, including olive oil, canola oil, waste cooking oil (canola oil put through at least three cycles of a small deep fryer), lard, and tallow. Irradiation with UV light (using either a Hanovia mercury lamp immersed in a quartz sleeve or a Fusion system equipped with an H-bulb) produced long-chain alkenes, dienes, trienes (such as 1-tetradecene; 1-hexadecene; 1,7-hexadecadiene; and 1,7,10-hexadecatriene) and glycerol. “The glycerol can be transformed through catalytic processes into acrylic acid and other essential raw materials for the plastic industry,” Neckers and Muro-Small write in their article.

Irradiation produced a number of photoproducts, and the results suggest that the

CONTINUED ON NEXT PAGE

TABLE 1. Photoproducts and yields^a

Fats and/or model systems	Products (% yield)			
	1-Tetradecene	1-Hexadecene	1,7-Hexadecadiene	1,7,10-Hexadecatriene
Lard	5	12	7	7
Tallow	10	9.8	13	—
Ethyl palmitate	34	—	—	—
Glyceryl tripalmitate	36	—	—	—
Glyceryl dipalmitate	34	—	—	—
Glyceryl monopalmitate	32	—	—	—

^aAdapted from “A green route to petroleum feedstocks: photochemistry of fats and oils,” *Sustainable Chemistry & Engineering*, doi:10.1021/sc400135y, 2013.

saturated fatty acids (palmitic and stearic) are more photoreactive than the unsaturated fatty acids (oleic and linoleic); reactivity appears to depend on the position of the fatty acids on the glycerol backbone.

Neckers noted that there are two yields—the chemical yield and photochemical yield, or how much light is required. (See Table 1.) “We think the chemical yield will scale up nicely,” he added. “We are not sure about the photochemical yield.”

Next up on the docket: investigating the mechanism in order to increase the yield, producing the photoproducts from the glycerol instead of the glyceride, and processing them into a source of acrylic acid.

“I think the synthesis is well done,” commented W. Warren Schmidt, a consultant based in Cincinnati, Ohio, USA, and a member of the *Inform* Editorial Advisory Committee. “It may take years, but it may well become an alternate source of olefins.”

Neckers and Muro-Small are not alone in searching for a biobased route to acrylic acid. A number of companies, including Dow Chemical/OPXBIO and BASF/Cargill/Novozymes, have mounted similar efforts.

Dow Chemical and OPXBIO (see www.opxbio.com) signed a collaboration agreement in April 2011 to develop an industrial-scale process to produce BioAcrylic—acrylic acid from fermentable sugars such as corn and/or cane sugar. OPXBIO says that a

life-cycle analysis conducted by Symbiotic Engineering, a greenhouse gas (GHG) and sustainability consultant, concluded that OPXBIO’s fermentation process using engineered microbes can reduce GHG emissions by more than 70% when compared to traditional petroleum-based acrylic acid production.

“The BioAcrylic process converts sugar feedstock . . . into the intermediate 3-hydroxypropionic acid (3-HP), which is then chemically dehydrated in a second step to BioAcrylic,” OPXBIO’s CEO Charles Eggert told *Inform* in an email. “We have successfully produced 3-HP in 3,000 liter (L) fermenters, and we anticipate pre-commercial production at the 25,000-L scale or larger beginning in 2014.

“Our plan is to start commercial-scale production and sales of BioAcrylic in 2017,” Eggert added, “with an initial plant capacity in the range of 100 million pounds [about 45,000 metric tons] per year.”

The BASF/Cargill/Novozymes partnership was formed in August 2012. The companies announced in July 2013 that they had produced 3-HP at pilot scale. Further, they reportedly have developed several technologies to dehydrate 3-HP at laboratory scale. The companies expect to reach the next level of scale-up in 2014, according to the Green Chemical Blog (www.greenchemicalsblog.com), although they declined to release any specific capacity goal. ■

AOCS Board Petition to Nominate

For each annual election of AOCS Governing Board officers, the membership may nominate up to four additional member-at-large candidates by petition. Petitioned candidates receiving at least 50 AOCS member signatures will be added to the ballot approved by the Governing Board. Preference will be given to the first four petitioned candidates meeting the eligibility requirements as outlined here. Petitioned nominations must be received at the AOCS Headquarters no later than **October 30, 2013**.

Petition forms can be obtained by visiting www.aocs.org/BoardPetition. Please mail or fax completed petitions with at least 50 AOCS signatures to:

AOCS Nominations and Elections Committee
P.O. Box 17190
Urbana, IL 61803-7190 USA

Fax: +1 217-693-4852
Attn: Benjamin Harrison



SUSTAINABILITY WATCH

Researchers at ITERG and CETIOM in France studied the complete life cycle—from the farm through to the end-of-life of the packaging—of sunflower oil and rapeseed oil produced in France. “We focused heavily on the impacts of crushing and refining,” the scientists wrote in *Oilseeds & Fats Crops and Lipids* (doi:http://dx.doi.org/10.1051/ocl/2013004, 2013). “The production of 100 g of refined bulk sunflower and rapeseed emits 89 g and 127 g equivalent CO₂ and consumes 1.7 L and 0.8 L of water, respectively. . . . Oil supply chain operators can use these values to compare to their own process values and gauge the improvements brought about by their ecodesign strategies. For example, using a biomass boiler, using less packaging, and making different choices on seed suppliers can lead to a lower set of impact values.”

The crushing and refining stages account for the vast majority of greenhouse gas (GHG) emissions generated, the authors note, with bottling accounting for just 5%–6%. “These GHG emissions are primarily driven by pressurized steam generation and electricity consumption,” they write. “Direct in-plant water consumption accounts for the majority of total water consumption, although indirect water consumption

tied to crushing and refining production-process inputs (water used to produce electricity, natural gas, chemical agents, etc.) accounts for a non-negligible share.”



A new method for producing electricity from carbon dioxide released from industrial smokestacks has been described in an article in *Environmental Science & Technology Letters* (10.1021/ez4000059, 2013).

Bert Hamelers of Wageningen University in the Netherlands and colleagues explain that electric power-generating stations alone release about 12 billion metric tons (MT) of CO₂ annually worldwide from combustion of coal, oil, and natural gas. Home and commercial heating produce another 11 billion MT. Smokestack gas from a typical coal-fired plant contains about 10% CO₂, they note.

The team describes technology that would react the CO₂ with water and, with further processing, produce an electric current. The process reportedly could produce a total of about 1,570 kilowatts of additional electricity per year, or about 400 times the annual electrical output of the Hoover Dam in the United States. ■

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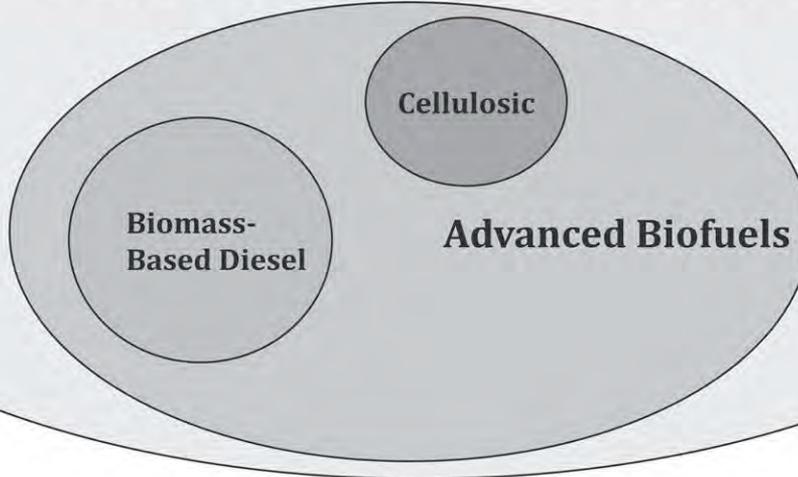
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BIOFUELS+

Total Renewable Mandate



EPA finalizes 2013 RFS

On August 6 the US Environmental Protection Agency (EPA) announced that it had finalized the 2013 percentage standards for four fuel categories (see Figure above) that are part of the Renewable Fuel Standard (RFS) program established by Congress.

The final 2013 overall volumes and standards require 16.55 billion gallons of renewable fuels to be blended into the US fuel supply (a 9.74% blend). Requirements of this standard are specified in Table 1. These standards reflect EPA's updated production projections, which

are informed by discussions with industry and by biofuels market analysis.

During this rulemaking, EPA received comments from a number of stakeholders concerning the "E10 blend wall." Projected to occur in 2014, the "E10 blend wall" refers to the difficulty in incorporating ethanol into the fuel supply at volumes exceeding those achieved by the sale of nearly all gasoline as E10 (i.e., 10% ethanol and 90% gasoline). Most gasoline sold in the United States today is E10. In its August 6 ruling, EPA said it will propose flexibilities in the RFS statute to reduce both the advanced biofuel and total renewable volumes in the forthcoming 2014 RFS volume requirement proposal.

EPA is also providing greater lead time and flexibility in complying with the 2013 volume requirements by extending the deadline to comply with the 2013 standards by four months, to June 30, 2014.

A January 2013 ruling by the US Court of Appeals required the agency to reevaluate projections for cellulosic biofuel to reflect market conditions; the final 2013 standard for cellulosic biofuel announced in August was developed in a manner consistent with the approach outlined in that ruling.

Canadian government support of biofuels falls

At a federal-provincial agriculture ministers meeting held in Halifax, Nova Scotia in July 2013, Minister of Agriculture and Agri-Food Gerry Ritz said the Canadian government would not increase its support for the biofuel industry, after having provided more than \$1 billion

TABLE 1. 2013 US Renewable Fuel Standards^a

Fuel	Volume ^b		Percentage
	Gallons	Liters	
Biomass-based diesel	1.28 billion	4.8 billion	1.13
Advanced biofuels	2.75 billion	10.8 billion	1.62
Cellulosic biofuels	6.00 million	2.27 million	0.004

^aSource: <http://tinyurl.com/2013-RFS-EPA>.

^bAll volumes are ethanol-equivalent, except for biomass-based diesel, which is actual volume.

already. Ritz said, “I think the investments made early on have built the industry as large as it’s going to get” (<http://tinyurl.com/Canada-biofuel-funding>).

The Canadian ethanol industry has been lobbying for an increase in ethanol content in gasoline to 10%, but Ritz said that is not feasible. The reason in part is that farmer investment in and local ownership of ethanol plants has not been as successful as the government had hoped, despite government action setting a mandate for biofuel content and provision of millions of dollars for plant construction.

Ritz commented that Canada cannot meet its current mandate of 5% ethanol in gasoline and 2% biofuel content in diesel without imports, which negates the government goal to help build another outlet for grain (see Table 2).

US corn oil production increasing

Bernie Hoffman, vice president of business development and minority owner of WB Services LLC, told *Ethanol Producer Magazine* that by the end of 2013 or early 2014, about 80% of US ethanol plants will be extracting corn oil from their by-product distillers dried grains with solubles (<http://tinyurl.com/corn-oil-EtOH>). Doing so is accounting for an increasing portion of total revenue for ethanol plants that have adopted and optimized the technology.

WB Services (Sedgwick, Kansas) is offering ethanol producers two separate technologies that offer them the opportunity to process corn oil into either biodiesel or renewable diesel on-site. The company has been involved in the construction of

two demonstration facilities in Sedgwick (neither of which is co-located with an ethanol plant). The first, a biodiesel plant, is already operating at a rate of 2 million gallons (7.6 million liters) per year. Construction is nearing completion on the second, a renewable diesel facility that will operate at 3 million gallons per year.

Although the two facilities are not co-located with an ethanol plant, WB Services says the plants are commercially viable, even though the corn oil must be purchased and shipped in. Co-location would allow for on-site feedstock production, lowered capital costs, and use of existing infrastructure.

Although both facilities are designed specifically to use corn oil as feedstock, the process allows for the use of oils or greases (www.servicesllc.com).

The company estimates that they can construct a biodiesel facility and bring it to commissioning in 6–8 months. The same process for a renewable diesel facility takes 12–14 months. Construction costs are lower for a biodiesel plant than for a renewable diesel plant.

The biodiesel technology used by WB Services is designed to cope with the higher free fatty acid content of corn oil that other biodiesel plants cannot handle.

Bacteria produce PHB

Researchers from the Polytechnic University of Catalonia (UPC; Spain) have found that *Bacillus megaterium* Uyuni S29, isolated from the water ‘eyes’ of the Uyuni salt flat in southwest Bolivia, stores large amounts of the polymer poly-β-butyrates

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In July 2013 Irish company Research and Markets released a report forecasting that the biofuels market in China will see a compound annual growth rate of 16.9% over the period 2012–2016. The growth of the transportation sector is a major contributor to this increase, as well as the increased demand for energy. High capital costs in the production of biofuels could act as a counterweight to this growth, however. For further information see <http://tinyurl.com/biofuels-China>.

■■■

INEOS Bio announced in August that its Indian River BioEnergy Center at Vero Beach, Florida, USA, had started producing cellulosic ethanol on a commercial scale. The company uses gasification and fermentation technology to produce renewable fuel as well as electricity. The company has already produced ethanol from vegetative and yard waste, citrus, oak, pine, and pallet wood waste. It has permits to use municipal solid waste, and will begin production of ethanol from it during 2014. Annual output will be 8 million gallons (30 million liters), or 24 kilo tons (22,000 metric tons), per year of cellulosic ethanol and six megawatts (gross) of renewable power.

■■■

Neste Oil, headquartered in Porvoo, Finland, announced it has added technical corn oil to its range of feedstocks used to produce its NExBTL renewable diesel. Matti Lehmus, Neste Oil’s executive vice president, oil products and renewables, commented, “Technical corn oil is an excellent addition to our feedstock base, as it is officially approved for producing renewable fuel intended for the growing North American market.” At present, Neste Oil is producing premium-quality renewable fuel from over 10 different feedstocks on an industrial scale. ■

TABLE 2. Biofuels in Canada

	Current production (liters/yr)	Current consumption (liters/yr)
Ethanol	~2 billion	2.8 billion
Biodiesel	155 million ^a	600 million

^aAn ADM biodiesel plant in Lloydminster, Alberta, scheduled to go on-stream in September 2013, is predicted to increase domestic production capacity to 400 million liters per year.

(PHB). This biodegradable plastic is used by the food, pharmaceutical, cosmetic, and packaging industries.

“[The Uyuni salt flats] are very extreme environments, which facilitate intracellular accumulation of PHB, a reserve material used by bacteria in times when nutrients are scarce,” said microbiologist Marisol Marqués-Calvo, one of the co-authors of the work (<http://tinyurl.com/bacteria-PHB>). Scientists from the UPC and the Graz University of Technology (Austria) have successfully made the bacillus produce significant quantities of the compound in the laboratory in cultivation conditions similar to those used in industry. The technique is published in the journals *Food Technology & Biotechnology* (51:123–130, 2013) and the *Journal of Applied Microbiology* (114:1378–1387, 2013).

“The resulting biopolymer has thermal properties different from conventional PHBs, which makes it easier to process, independently of its application,” Marqués added. The research team has managed to reduce PHB’s high molecular weight for the first time, using lipase enzymes, which break up fats, as well as using the biopolymer to form nano- and microspheres loaded with antibiotic to control their spread throughout the organism.

Malaysia expands use of palm biodiesel

The government of Malaysia and the Ministry of Plantation Industries and Commodities announced in July the expansion

of the country’s mandatory biodiesel program into the Southern Region. The Central Region, encompassing Putrajaya, Melaka, Negeri Sembilan, Selangor, and Kuala Lumpur, first introduced mandatory biodiesel use in January 2011. The Central Region has since used 113,000 metric tons (MT) of palm biodiesel annually.

Implementation in the Southern Region should lead to the use of 37,270 MT of palm diesel per year, or a savings of 43.14 million liters per year of petrodiesel consumption. In July palm diesel was being sold in the Southern Region for RM1.80 per liter, or \$0.55 per liter.

The biodiesel program contributed to the 44% increase in the nation’s palm biodiesel production, that is, from 173,220 MT in 2011 to 249,213 MT in 2012. The program also has contributed toward stabilizing the crude palm oil price through increased demand for palm biodiesel.

The successful implementation of the biodiesel program in the Central and Southern Regions will result in the increase in palm biodiesel consumption in the country to 149,630 MT per year. This accounts for 30% of the total target of 500,000 MT a year when the B5 (5% biodiesel + 95% petrodiesel) program is implemented nationwide by July 2014. To strengthen the crude palm oil price through stock reduction, the Government has decided to increase the percentage of palm biodiesel blend above the 5% threshold. ■



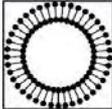
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“Evening primrose oil (EPO) is widely used as a dietary supplement from which beneficial effects have been reported in rheumatic and arthritic conditions, premenstrual and menopausal syndromes, and atopic dermatitis,” write researchers led by Sergio Montserrat-de la Paz of the University of Seville in Spain. Their study was designed to examine the effects of diets supplemented with EPO on generalized chronic pain, including fibromyalgia syndrome, induced by intermittent cold stress in mice. “The results indicate that dietary EPO is suitable to improve mechanical and thermal allodynia [a painful response to a normally innocuous stimulus] and hyperalgesia [increased pain response to a noxious stimulus] and it is also . . . able to improve behavioral disturbances, anxiety, and depression,” the team reports. In addition, EPO—which is high in γ -linolenic acid—significantly reduced proinflammatory mediators, they say. The work appeared in the *Journal of Functional Foods* (doi: <http://dx.doi.org/10.1016/j.jff.2013.04.012>, 2013).

■■■

A new report from Mintel, a market research firm based in Chicago, Illinois, USA, reveals that for just more than half (51%) of all adult consumers in the United States, the favorite type of plain chocolate is milk chocolate, followed by 35% who favor dark chocolate, and 8% who prefer white chocolate. In contrast, Mintel’s 2011 report found that 57% of consumers favored milk chocolate and 33% of consumers preferred dark chocolate. For more details about the report, see <http://tinyurl.com/Mintel-Choco>.

■■■

European consumers can understand the information presented on nutrition labels, but labels have little effect on purchasing decisions. That is the conclusion of the FLABEL project (Food Labeling to Advance to Better Education for Life), a three-year study of nutrition labeling on 37,000 food products sold across Europe. The researchers found that the average consumer spends between 25 and 100 milliseconds looking at nutrition labels—too brief a span of time for the information to be processed meaningfully. More information about the project is available at <http://www.flabel.org/en>. ■

FOOD, HEALTH & NUTRITION



Omega-3 fatty acids and oral and skin cancers

Omega-3 fatty acids (omega-3s), contained in oily fish such as salmon and sardines, selectively inhibit growth and induce cell death in early and late-stage oral and skin cancers, according to new research from scientists at Queen Mary, University of London.

In vitro tests showed omega-3s induced cell death in malignant and premalignant cells at doses that did not affect normal cells, suggesting omega-3s have the potential to be used in both the treatment and prevention of certain skin and oral cancers—the cancers of interest to the Queen Mary researchers.

In particular, the team was interested in studying omega-3s and squamous-cell carcinomas (SCC). Squamous cells are the main part of the outermost layers of the skin, and SCC is one of the most prevalent forms of skin cancer. However, squamous cells also occur in the lining of the digestive tract, lungs, and other areas of the body. Further, oral SCC are the sixth most common cancer

worldwide and are difficult and very expensive to treat.

In the experiments, the researchers grew cultures from several different cells lines, to which they added fatty acids. The cell lines included both oral and skin SCC, along with premalignant cells and normal skin and oral cells. Kenneth Parkinson, head of the Oral Cancer Research Group at Queen Mary’s Institute of Dentistry, said of the finding that the omega-3s selectively inhibited the growth of the malignant and premalignant cells: “Surprisingly, we discovered this was partly due to an overstimulation of a key growth factor (epidermal growth factor), which triggered cell death. This is a novel mechanism of action of these fatty acids.”

While previous research has linked omega-3s with the prevention of a number of cancers, there has been very little work done on oral cancers or normal cells, the authors noted.

Zacharoula Nikolakopoulou carried out the research while studying for her doctorate at Queen Mary, under the supervision of Parkinson and Adina Michael-Titus.

CONTINUED ON NEXT PAGE

Nikolakopoulou said: “It may be that those at an increased risk of such cancers—or their recurrence—could benefit from increased omega-3 fatty acids. Moreover, as the skin and oral cancers are often easily accessible, there is the potential to deliver targeted doses locally via aerosols or gels. However, further research is needed to define the appropriate therapeutic doses.”

The research appeared in *Carcinogenesis* (10.1093/carcin/bgt257, 2013).

Omega-3 insufficiency may have worsening effects over time

In another study on omega-3 fatty acids, US researchers found that diets lacking omega-3 fatty acids (omega-3s) can have worsening effects over consecutive generations of rats, especially affecting adolescent animals. The study was led by Bitu Moghaddam, a professor of neuroscience in the Kenneth P. Dietrich School of Arts and Sciences at the University of Pittsburgh (Pitt; Pennsylvania).

The Pitt team found that in their model, second-generation insufficiencies of omega-3s caused elevated states of anxiety and hyperactivity in adolescents and affected the young rats’ memory and cognition.

“We have always assumed that stress at this age is the main environmental insult that contributes to developing these conditions in at-risk individuals, but this study indicates that nutrition is a big factor, too,” said Moghaddam. “We found that this dietary deficiency can compromise the behavioral health of adolescents, not only because their diet is deficient but because their parents’ diet was deficient as well. This is of particular concern because adolescence is a very vulnerable time for developing psychiatric disorders, including schizophrenia and addiction.”

The Pitt team administered a set of behavioral tasks to study the learning and memory, decision making, anxiety, and hyperactivity of both adult and adolescent rats. Although subjects appeared to be in general good physical health, there were behavioral deficiencies in adolescents that were more pronounced in second-generation subjects with omega-3 insufficiencies. Overall, these adolescents were more anxious and hyperactive, learned more slowly, and had impaired problem-solving abilities. The team found that the insufficiency affected expression of dopamine-related proteins in both age groups. Further, the adolescents exhibited an increase in tyrosine hydroxylase expression that was selective to the dorsal striatum. Current thinking suggests that the dorsal striatum contributes directly to decision making.

“Our study shows that, while the omega-3 deficiency influences the behavior of both adults and adolescents, the nature of this influence is different between the age groups,” said Moghaddam.

The team is now exploring epigenetics, or how environmental events influence genetic information, as a potential cause of the differences. Likewise, the team is exploring markers of inflammation in the brain, since omega-3 insufficiencies cause an increase of omega-6 fats, which are in certain cases proinflammatory molecules in the brain and other tissues.

“It’s remarkable that a relatively common dietary change can have generational effects,” said Moghaddam. “It indicates that our diet does not merely affect us in the short term but also can affect our offspring.”

The study appeared in *Biological Psychiatry* (doi: 10.1016/j.biopsych.2013.06.007, 2013).

Vitamin D and immune-mediated disorders

Vitamin D deficiency has been linked to immune-mediated diseases by a number of studies. There is some evidence that vitamin D deficiency plays a role in multiple sclerosis, type 1 diabetes (T1D), inflammatory bowel disease, rheumatoid arthritis, systemic lupus erythematosus, and Sjogren’s syndrome, all of which are considered to be immune-mediated conditions.

Does vitamin D deficiency correlate with immune-mediated diseases in the hospital setting? That was the question that researchers led by Sreeram V. Ramagopalan of the University of Oxford set out to answer in a cross-sectional study that appeared in *BMC Medicine* (doi:10.1186/1741-7015-11-171, 2013).

The team searched medical records for all patients admitted to a particular hospital, specifically looking for patients who were medically coded for vitamin D deficiency or rickets and osteomalacia, which are linked to vitamin D deficiency. They next examined what diseases these vitamin D-deficient patients suffered from, matching the deficient patients with controls—patients admitted to the hospital that were not suffering from vitamin D deficiency, rickets, or osteomalacia. In total, the researchers had records for 19,679 patients with vitamin D deficiency (13,260 with vitamin D deficiency only, 1,228 with rickets, and 5,191 with osteomalacia).

- The researchers found an extensive list of immune-mediated diseases associated with vitamin D deficiency. Compared to controls, patients coded for vitamin D deficiency had an increased risk of having Addison’s disease (which causes adrenal hormone insufficiency), ankylosing spondylitis (a type of arthritis), autoimmune hemolytic anemia, chronic active hepatitis, celiac disease, Crohn’s disease, T1D, pemphigoid (a rare autoimmune skin blistering disease), pernicious anemia, primary biliary cirrhosis, rheumatoid arthritis, Sjogren’s syndrome, and other autoimmune disorders. All of these associations were statistically significant.
- A diagnosis of vitamin D deficiency, however, also pointed to a decreased risk of having asthma and myxedema, and decreased activity of the thyroid gland.

“Regardless of whether vitamin D deficiency causes these diseases or these diseases cause vitamin D deficiency, vitamin D deficiency should be treated and monitored,” said Brant Cebulla, writing about the study on the Vitamin D Council’s blog (www.vitaminDcouncil.org). “At the very least, this study provides evidence that patients with these diseases are at risk for vitamin D deficiency and should be treated accordingly. Further research will clarify if vitamin D sufficiency has any prevention or treatment effect for these diseases.” ■

Monsanto Co. (St. Louis, Missouri, USA) announced in July 2013 that it would withdraw all its pending approval requests to grow new types of genetically modified organisms (GMOs) in the European Union. The reason given was the lack of commercial interest in growing the crops there. Instead the company said it will concentrate on its conventional seeds business in Europe and also on approvals to import to Europe its genetically modified crops grown in the United States and South America. The withdrawn requests covered five corn GMOs, one soybean, and one sugar beet. The company does not plan to withdraw its application to renew the approval for its insect-resistant MON810 corn—the only GMO crop currently cultivated commercially in Europe (<http://tinyurl.com/Monsanto-withdrawal>).

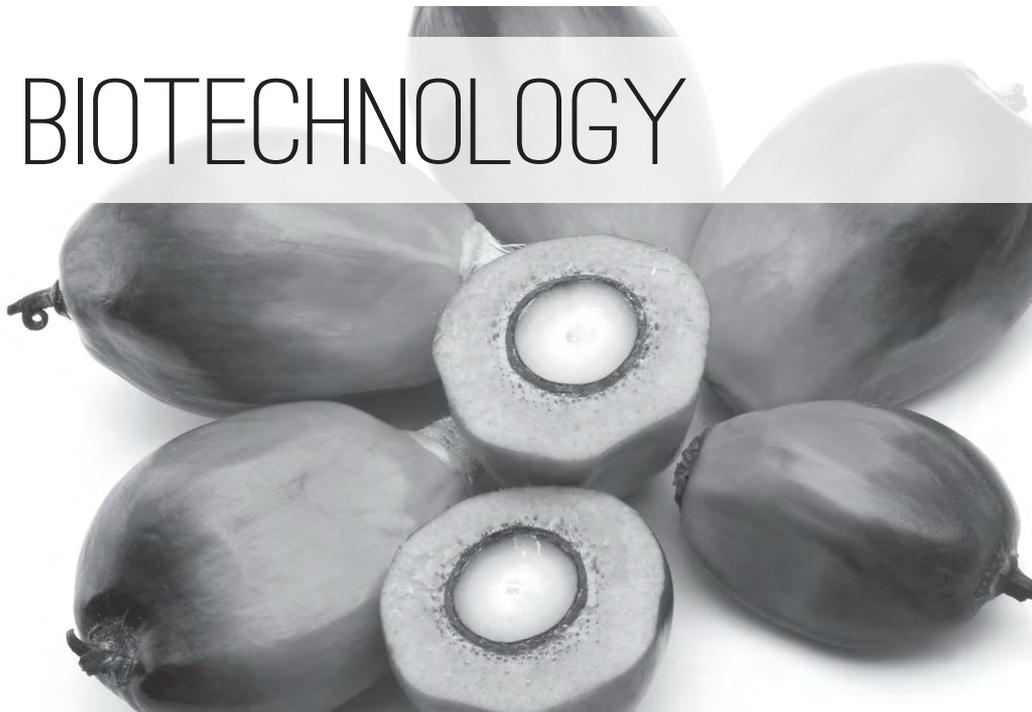
■■■

The Council for Biotechnology Information—including BASF, Dow AgroSciences, DuPont, Bayer CropScience, Syngenta, and Monsanto—created an online information portal concerning genetically modified organisms (GMO). The site, www.gmoanswers.com, contains information about GMO uses and history and the health and safety of GMO crops. Representatives from participating companies and supporting partners will also answer questions about GMO through public message boards.

■■■

As of July 29, 2013, Japan is accepting US wheat exports again after the discovery of a genetically modified (GM) wheat plant on a farm in the state of Oregon (see *Inform* 24: 510–511, 2013). Earlier that day, the US Department of Agriculture (USDA) announced that this contamination with Monsanto's Roundup-resistant wheat was an isolated case. But questions remain as to how the contamination occurred. According to the *St. Louis Post-Dispatch* newspaper, regulators are comparing this case to a 2006 GMO contamination, when Bayer CropScience's GM LibertyLink rice got into the US market. In that case, the source of contamination was never found. Monsanto tested its GM wheat from 1998 to 2005. It claimed all of the seed was destroyed at the end of the experiment. ■

BIOTECHNOLOGY



Genome of oil palm mapped

Genomes of two oil palm species—*Elais guineensis* and *E. oleifera*—have been mapped by researchers with the Malaysian Palm Oil Board (MPOB; Kuala Lumpur), Orion Genomics (St. Louis, Missouri, USA), and Cold Spring Harbor Laboratory (New York, USA). The results appeared in the journal *Nature* in late July.

The research identified a single gene in *E. guineensis* that is critical for yield of the oil palm, a crop that accounts for nearly half of the world's edible vegetable oil. The gene is called *SHELL*. Mutations in the gene explain the single most important economic trait of the oil palm: how the thickness of the shell surrounding the seed correlates to the fruit size and oil yield.

Three shell forms are known: *dura* (thick), *pisifera* (shell-less), and *tenera* (thin). The latter, which is a hybrid between *dura* and *pisifera* palms, has a distinct fiber ring surrounding the coconut-like shell of the oil palm seed.

Until now, mapping of the *SHELL* gene has been very challenging because the genome of the plant is large; generation times are long; and phenotyping in experimental oil palm plantations, which are widely distributed geographically, is difficult.

The *tenera* palm produces 30% more oil per hectare than *dura* palms and is commercially the most desirable form. Currently, it

can take six years to identify whether an oil palm plantlet has the desired shell form. The genetic marker will also be important for quality control in commercial seed production; allowing for reduction or elimination of *dura* forms in the nursery, long before the plants are set out in the field.

Being able to genotype the oil palm will enable the achievement of higher oil yields, and may lead to reduction in land area devoted to oil palm plantations. In turn, genotyping could then provide opportunities for conservation and restoration of dwindling reserves of tropical rain forests, which have been destroyed in favor of oil palm plantations.

According to the *St. Louis Post-Dispatch* newspaper (<http://tinyurl.com/diagnostics-patents>), the MPOB has not decided how this information will be transferred to oil palm producers. The likelihood is the MPOB will license a diagnostic test, for which it will receive royalties. The MPOB has been filing gene patents during the past year pertaining to applications of the gene and diagnostic approaches.

The researchers also revealed a draft sequence of *E. oleifera*, which has the same number of chromosomes ($2n = 32$) and produces fertile interspecific hybrids with *E. guineensis* but seems to have diverged about 51 million years ago in the Old World–New World split. Of the two, *E. guineensis* has a higher oil yield, but *E. oleifera* has higher

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unsaturated fatty acid content, lower height, and resistance to disease.

For further information, see the original papers:

- Singh, R., E.L. Low, L. Cheng, L. Ooi, M.O. Abdullah, *et al.*, The oil palm *SHELL* gene controls oil yield and encodes a homologue of SEEDSTICK, *Nature*, doi: 10.1038/nature.12356.
- Singh, R., M. Ong-Abdullah, E.L. Low, M.A.A. Manaf, R. Rosli, *et al.*, Oil palm genome sequence reveals divergence of interfertile species in Old and New worlds, *Nature*, doi: 10.1038/nature12309.

Brazilian farmers' suit against Monsanto dropped

Aprosoja, Brazil's largest soybean cooperative, dropped its lawsuit against US seed producer Monsanto in August 2013. Aprosoja filed the lawsuit on behalf of Brazilian farmers to reclaim royalties on Roundup Ready seeds paid since 2010.

Both sides came to an agreement outside of court that led to Aprosoja ending its legal action. As part of the agreement, Monsanto is offering discounts of 16% per hectare on Intacta RR2 PRO™ insect-resistant seeds for farmers who sign contracts agreeing not to sue the GM seed company for the royalties they had paid.

Monsanto had claimed that its seed patent on its Roundup Ready crop extended into 2014. However, Brazil's Upper Tribunal of Justice rejected the company's claim in February 2013, declaring that the patent expired in 2010. Soybean farmers, who were still charged royalties between 2010 and 2013 as the case was proceeding, wanted their paid monies returned, and they enlisted the help of Aprosoja to prosecute the company.

The company's genetically modified technology is present in 85% of Brazil's soyfields, according to Reuters news service.

Farming of GM crops slowly accepted in Africa

Many African countries remain skeptical of the use of genetically modified (GM) crops. However, some nations have been swayed to a more positive outlook.

South Africa has grown GM corn, soybeans, and cotton commercially since 1996 and is both the first GM crop sower and largest producer in Africa, growing 3 million hectares of GM crops annually. Egypt began planting GM corn in 2008; Burkina Faso, GM cotton in 2009; and Sudan, GM cotton in 2012.

The rest of the continent has been slow to make the shift. The reason for the resistance involves political and economic pressures, particularly those of the largest importer of Africa's crops, the European Union (EU). Strict EU regulations on GM food and feed products have discouraged African leaders from investing. Several leaders are also concerned that using GM crops gives seed companies such as DuPont and Monsanto too much influence on African economies.

Despite these concerns, Cameroon, Kenya, Malawi, Ghana, and Uganda have approved confined trials of GM crops. Also, in July 2013, DuPont Pioneer completed its acquisition of South African seed company Pannar Seed Ltd. As part of the investment, DuPont will invest 62 million in South African Rand (\$6.0

million) in building a regional research center similar to the DuPont Innovation Centers elsewhere in the world. Pannar will also receive access to DuPont's genetics library and technology to improve African farming techniques.

Bacteria coating may eliminate need for fertilizer for oilseed crops

The University of Nottingham (England, United Kingdom) licensed bacterial coating technology for crops to British sustainable nitrogen research company Azotic Technologies Ltd. The coating, developed by university professor Edward Cocking, allows plants to absorb nitrogen from the surrounding atmosphere through nitrogen fixation.

Most plants—the exceptions being legumes such as peas, beans, and lentils, which can perform nitrogen fixation on their own—use fertilizer to efficiently supply nitrogen to build nucleotides for DNA and RNA and create amino acids. The bacterial coating makes it possible for plants to take nitrogen out of the air the same way legumes do. This bacterium can incorporate itself into the cellular structure of all major crops, including soybean, cotton, and others, forming a commensal relationship.

Azotic will handle acquiring initial regulatory approval in the UK, Europe, the United States, and other countries. The company expects the coating to be available in two to three years under the name N-Fix.

For more information, go to www.azotictechnologies.com/index.php/.

Discovery of gene in rice plants that increases yield in drought conditions

In a paper appearing in the August 2013 issue of *Nature Genetics* (doi: 10.1038/ng.2725) scientists report they have found a gene in rice plants that can be altered to increase root length, helping the plant absorb more nutrients and survive intense drought conditions.

Rice plants with a higher expression of the DEEPER ROOTING 1 (DRO1) gene will have roots that point downward and grow up to twice as deep into the ground. Rice plants with the DRO1 gene had 3.6 times the yield of non-altered rice plants in severe drought conditions. Even in moderate growing conditions, the deeper roots led to twice the yield of rice plants without DRO1.

The DRO1 gene occurs naturally in more than 60 rice varieties. The gene was altered through traditional crossbreeding techniques until the team produced a rice crop in which DRO1 expression was uniformly present.

The authors of the paper pointed out in their conclusion that: "Other economically important monocots, such as maize, contain DRO1 homologs that may be useful for enhancing drought avoidance in other crops. Our results open the way for new breeding strategies using genes influencing root system architecture to develop crop cultivars with high adaptability to drought."

Other important oil-bearing monocots include corn and oil palm. ■

The US Environmental Protection Agency (EPA) has added more than 130 compounds—including 119 chemicals used as fragrances in home and institutional cleaning products—to the agency's safer ingredients list. This marks the first time EPA has added fragrances to the list, which is used as a tool for deciding which products can use EPA's voluntary Design for the Environment ecolabel. For more information, visit www.epa.gov/dfe/saferingredients.htm.

■ ■ ■

In August 2013, Solazyme and Sasol Olefins & Surfactants GmbH (Sasol O&S; Hamburg, Germany) announced that they have finalized commercial terms for the supply by Solazyme to Sasol of an algal oil rich in erucic acid ($C_{22}H_{42}O_2$) that is under development at Solazyme for production of downstream derivatives such as behenyl alcohol ($C_{22}H_{45}OH$). Sasol O&S produces and sells C22 derivatives such as behenyl alcohol to serve a number of applications in markets that include the paper, water treatment, personal care, lubricants, paints, inks, coatings, and adhesives industries. In addition to the agreement on commercial supply terms, the companies also executed a letter of intent to investigate expanding to a broad collaboration.

■ ■ ■

Evonik Industries (Essen, Germany) announced in late July 2013 that it has been operating a pilot plant for production of ω -amino lauric acid (ALS) in Slovenská Ľupča, Slovakia, since the first of the year. The biobased ALS is an alternative to petroleum-based laurin lactam (LL). ALS replaces the monomer LL in the manufacture of sustainable high-performance plastics and yields an identical compound polyamide 12 (PA 12). Palm kernel oil (PKO), which Evonik has already been using as a base for various other chemical products, is the starting material. "It is hard to imagine that volumes would be high enough to disrupt the over 6 million metric ton market for PKO," commented Neil A. Burns, a consultant based in Freehold Township, New Jersey, USA. "I think it will be an excellent value-added renewable product that will make barely a ripple in the PKO supply chain," he added. ■

SURFACTANTS, DETERGENTS & PERSONAL CARE



ACI releases sustainability report

The American Cleaning Institute's (ACI) 2013 Sustainability Report, which was released in early August 2013 by the Washington, DC-based trade group, shows overall reductions by member companies in four environmental metric data points: energy use, greenhouse gas emissions, water use, and solid waste generation.

Twenty-four ACI member companies, including cleaning product makers and suppliers, submitted environmental metrics data reflecting US cleaning product-related production. To ensure year-to-year comparability in all data categories, only companies whose data spanned all three reporting years (2009, 2010, and 2011) are presented in the report.

The US cleaning product industry's report details continual adoption of sustainability practices within the cleaning products supply chain, said ACI. "[The report] also highlights how ACI and its members are improving product safety and transparency and giving back to those in need," the group said in a news release.

Among the findings of the 2012 ACI Sustainability Report:

- Overall, 2011 showed a reduction in the industry's footprint for all

four environmental metrics compared to 2009. During this period, energy use decreased 9%, greenhouse gas emissions decreased 7%, water use decreased 5%, and solid waste generation decreased 17%.

- There was also improvement in the energy use profile per cleaning product produced. From 2009 to 2010, energy use saw a 6% decrease, and from 2010 to 2011, this metric decreased 4%.
- There was a 12% decrease in greenhouse gas (GHG) emissions from 2010 to 2011.

The report is available at <http://www.cleaninginstitute.org/sustainability2013>.

World demand for lubricants rises

World demand for lubricants—a key end use for surfactants—is expected to increase 2.3% per year to 43.9 million metric tons (MMT) in 2017. The fastest growth will be in Asia, supported by rising vehicle ownership rates and ongoing industrialization in large countries such as China. Above-average increases will also occur in South America, the Middle

CONTINUED ON NEXT PAGE

East, and Africa. These regions will each experience healthy economic growth, rising manufacturing output, and expanding motor vehicle ownership—all of which will contribute to gains in lubricant consumption.

In contrast, demand will remain nearly flat in the developed countries of North America and Western Europe, where efficiency gains will offset the effects of rising economic and industrial output. Although volume growth will be restrained in these regions, lubricant suppliers will benefit from increasing demand for premium, high-value products such as synthetic and bio-based lubricants. These and other trends are presented in *World Lubricants*, a new study from The Freedonia Group, Inc., a Cleveland, Ohio, USA-based industry market research firm.

Demand for lubricants used in the manufacturing market will see healthy growth, driven by rising manufacturing output especially in developing countries. Asia, Eastern Europe, South America, the Middle East, and Africa will benefit from the continued shift of global manufacturing activity to these regions. Gains in other markets, including agriculture and construction, will in the aggregate outpace those in both the motor vehicle and manufacturing markets through 2017. Healthy economic growth, investment in infrastructure and other construction, and the increasing mechanization of the economies in developing countries will result in strong performance in these markets.

For more information about the report, visit www.freedoniagroup.com.

Study: AE demonstrate low environmental risk

Field research on key detergent ingredients demonstrates low environmental risk to waterways and river sediments, according to a study by the American Cleaning Institute (ACI), a trade group based in Washington, DC.

The research, “Occurrence and Risk Screening of Alcohol Ethoxylate Surfactants in Three US River Sediments Associated with Wastewater Treatment Plants,” appeared in the *Science of the Total Environment* (463–464:600–610, 2013). The complete article is available at <http://tinyurl.com/AES-ACI>.

Alcohol ethoxylates (AE) are high production volume chemicals used around the world in detergent and personal care products. The chemical backbone of AE—aliphatic alcohols or, simply, fatty alcohols—represents a special interest in the context of environmental risk, as fatty alcohols are also abundant and ubiquitous naturally occurring compounds that may be found in animal and human waste, plant matter, runoff, and the like.

Hence, in a risk assessment, researchers need to distinguish between the natural fatty alcohol concentrations and the added contribution from human activities, according to Kathleen Stanton, ACI director of technical and regulatory affairs and one of the paper’s co-authors.

“The major disposal route of AE is down the drain through sewage systems and municipal wastewater treatment plants into receiving surface waters,” said Stanton. “This makes the fate and effects of residual AE in treated sewage effluent of interest to industry and regulators alike.

“We found through a weight-of-evidence risk assessment that AE and fatty acids associated with detergent use present a

low risk to the environment. It also highlights the need to carefully consider the procedures for environmental risk assessment of compounds such as fatty alcohols, as they are found naturally in the environment as well as associated with consumer product use.

“This latest article ties a bow on field work first conducted in 2003,” added Stanton.

Pres. Obama calls for improved chemical safety

US President Barack Obama on August 1 signed an executive order instructing key federal agencies, including the Department of Labor, to work together to reduce the risks of hazardous chemicals to workers and communities.

The order directs federal agencies to initiate “innovative approaches” for collaborating with stakeholders to improve chemical facility safety and security through agency programs and private-sector initiatives, federal guidance, standards, and regulation. Included in the order is a requirement for the department to review, along with the Environmental Protection Agency (EPA), the Occupational Safety and Health Administration’s Process Safety Management (PSM) standard and the EPA’s Risk Management Program (RMP). The review will determine if the PSM standard or RMP can and should be expanded to address additional regulated substances and types of hazards. In a conference call with stakeholders, Assistant Secretary of Labor David Michaels, the head of the Occupational Safety and Health Administration, said his agency strongly supports the efforts of the president to improve and enhance chemical plant safety.

More information on the executive order is available at <http://tinyurl.com/Chem-Safety-Order>.

Wilmar and Elevance partner

Singapore’s Wilmar International Ltd. and Elevance Renewable Sciences, Inc., a specialty chemicals company in Woodridge, Illinois, USA, announced in July 2013 that they have begun shipping commercial products, including novel specialty chemicals, to customers from their first world-scale joint venture biorefinery in Indonesia. The biorefinery is the first based on Elevance’s proprietary metathesis technology. (For more about metathesis, see www.elevance.com/technology/metathesis/.)

The commercial-scale manufacturing facility produces novel specialty chemicals, including multifunctional esters such as 9-decenoic methyl ester, biobased alpha and internal olefins such as decene, and oleochemicals. The facility has a capacity of 180,000 metric tons (MT) per year with the ability to expand up to 360,000 MT.

The specialty chemicals, olefins, and oleochemicals will be used in personal care products, detergents and cleaners, lubricants and additives, engineered polymers, and other specialty chemicals markets.

The new plant is located within Wilmar’s integrated manufacturing complex in Gresik, Indonesia. The biorefinery will initially operate using palm oil, but is capable of running on multiple renewable oil feedstocks, including mustard, soybean, and, when they become commercially available, jatropha and algal oils. ■

PEOPLE/INSIDE AOCS

Erhan recognized

The Oil Technologists' Association of India and CSIR-Indian Institute of Chemical Technology have named AOCS member **Sevim Erhan** recipient of the Dr. K.T. Achaya Award for her significant contributions made in the area of lubricants. The award was presented during the inaugural function of the International Conference on Emerging Trends in Oleochemicals & Lipids Expo 2013 in Hyderabad, India on August 8.

Holmes takes new position

After 16 years with Mississippi State University (USA), **William Holmes** moved in June to the University of Louisiana at Lafayette (USA), where he is director of mass spectrometry and instrument development for the College of Engineering. He is presently working in the areas of energy and oils.

New general manager for Crown Companies

CPM, the world's leading supplier of process engineering, process equipment, and aftermarket parts for the oilseed, animal feed, bio-fuels and food processing industries, announced the appointment of **Todd Fierro** to Crown Companies' general manager. Fierro fills a new executive management position responsible for global coordination of all sales, operations, engineering, strategy, and administrative

functions. He will be based in Roseville, Minnesota, USA, and will report to **Ted Waitman**, CEO of CPM.

"Todd will complete the global alignment of resources and technologies to strengthen Crown's leadership position in

preparation, extraction, refining and oleochemical solutions," said Waitman.

New IFT Fellows

AOCS members **Gary List**, retired from the US Department of Agriculture, National Center for Agricultural Utilization Research, Peoria, Illinois, and **Lekh Juneja**, executive vice president of Taiyo Kagaku Co. Ltd., Yokkaichi, Mie, Japan, were named Fellows of the Institute of Food Technologists at the IFT Annual Meeting + Food Expo held in Chicago, Illinois, USA on July 13–16, 2013. ■

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AOCS MEETING WATCH

November 6–8, 2013. Australasian Section AOCS Biennial Meeting and Workshops, NOAH's on the Beach, Newcastle, New South Wales, Australia. www.aocs.org/australasian.

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>.

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.

IN MEMORIAM

ROBERT GEORGE ACKMAN

Internationally renowned marine oil and lipid chemist Robert G. Ackman died in Dartmouth, Nova Scotia, Canada on July 16, 2013, at the age of 85. He had been a member of AOCS for 46 years.



He is survived by his wife Catherine, his daughters Elizabeth and Margaret, four grandchildren, and his sister.

Born in Dorchester, New Brunswick, Ackman completed his B.A. degree in organic chemistry at the University of Toronto in 1950, his M.S. in organic chemistry from Dalhousie University in 1952, his Ph.D. in organic chemistry from the University of London (England) in 1956 and a D.I.C. (Diploma of the Imperial College, London, England) in organic chemistry. At Imperial College, he was introduced to "vapor phase chromatography (VPC)" (as gas-liquid chromatography, or GLC, which had been developed only a couple of years previously, was then known) and he saw its potential for fatty acid analysis.

Ackman started his career in marine oils in 1950 as a research chemist at the Halifax Laboratory of the Fisheries Research Board of Canada. In 1979 he helped establish the Canadian Institute of Fisheries Technology and later the Department of Food Science and Technology at the Technical University of Nova Scotia (TUNS; since April 1997 a part of Dalhousie University). In 1995, he was appointed Professor Emeritus.

Ackman is best known for his pioneering work on lipid analytical chemistry, particularly in the capillary GLC of fatty acids and the chemistry and biochemistry of marine lipids. He edited the classic text *Marine Biogenic Lipids, Fats and Oils*, Volumes I and II (CRC Press, 1989) and was author on over 550 scientific papers. His work in the early 1960s established the relationship between the retention time of fatty acid methyl esters and their structure on GLC columns of different polarity, and on the response of the flame ionization detector to fatty acid structure, thus providing a mechanistic basis for both the qualitative and quantitative analysis of fatty acids. He developed a number of techniques and procedures in the analysis of marine oils especially the omega-3 fatty acids of fish oils, that are now widely used to analyze complex fatty acid mixtures from marine organisms.

Through his expertise in fatty acid analysis Ackman played an important role in the development of low-erucic acid rapeseed oil (canola oil).

Members of AOCS were well served by Dr. Ackman's skills and knowledge. He was an associate editor for *Lipids* from 1980 to 2006, and he reviewed articles for the *Journal of the American Oil Chemists' Society* as well. He participated in the activities of the Health and Nutrition Division and the AOCS Canadian Section, and he served as a member-at-large of the AOCS Governing Board from 1989 to 1991. He received the Supelco–American Oil Chemists' Society Award in 1994 for original research in fats, oils, lipid chemistry, or biochemistry.

The International Society for Fat Research presented Ackman with its H.P. Kaufmann Memorial Lecture Medal in 1980. He received an honorary Doctor of Laws degree from Dalhousie University in 2000; and in 2001 he was made an Officer of the Order of Canada, in recognition of his outstanding level of talent and service to Canadians. Ackman also received Canada's Diamond Jubilee Medal in 2012.

Recollections by co-workers are available in this issue's supplement (digital and mobile editions only).

JEAN A. BÉZARD

French oil chemist Jean Bézard died on March 4, 2013, in Saint Apollinaire at the age of 82. He is survived by his wife Arlette and his sons Bruno, Thierry, and Hervé, all of whom became scientists like their father.

Bézard received his doctorate of natural sciences in 1965, then became professor and subsequently chair of the animal physiology program at the Université de Bourgogne, president of the Association française de nutrition, and an Officier of the Palmes Académiques for his service to French education. He was awarded emeritus status in 1996.

Some of his early research interests included the hepatic metabolism of cholesterol and omega-3 fatty acids. His research also involved investigations on the composition of palm kernel oil and chromatographic analysis of triglycerides.

Bézard joined AOCS in 1973. He served as president of the European section of AOCS in 1995–1996 and continued as a section officer through 2001. ■



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PATENTS

Chewable soft capsule having improved ingestion property and method for producing same

Shodai, H., *et al.*, Shionogi & Co., Ltd., US8372428, February 12, 2013

A soft capsule in which a shell is filled with fill material, and the fill material is in a solid or semi-solid form at room temperature. The soft capsule may be a chewable capsule, and the fill material may comprise a low melting point additive. The content of the low melting point additive may be 10% or more with respect to the total weight of the fill material, and may have a melting point of about 20–50°C. The low melting point additive may be selected from the group consisting of chocolate base, lard, coconut oil, and macrogol (polyethylene glycol) as well as a combination thereof.

Expression of fatty acid desaturases in corn

Ursin, V., *et al.*, Monsanto Technology LLC, US8378186, February 19, 2013

The invention relates generally to the expression of desaturase enzymes in transgenic corn plants and compositions derived therefrom. In particular, the invention relates to the production of oils with improved omega-3 fatty acid profiles in corn plants and the seed oils produced thereby. Such oils may contain stearidonic acid, which is not naturally found in corn plants and has been shown to have beneficial effects on health.

Laminating adhesives based on triglyceride-derived polyols

Simons, J.B., Henkel Corp., US8382937, February 26, 2013

Natural oils, fats, and other fatty acid triglycerides are transesterified with polyalcohols such as trimethylolpropane or glycerol to yield polyhydroxyl-functionalized reaction products that are useful materials in the formulation of two-component laminating adhesives.

Dielectric fluid composition containing vegetable oils and free of antioxidants

Aranda Cotero, J., *et al.*, US8383020, February 26, 2013

A dielectric fluid composition containing 60–63% in weight of monounsaturated fatty acid, of 20–23% in weight of di-unsaturated fatty acid, of 5–7% in weight of tri-unsaturated fatty acid, and of 6–8% in weight of saturated fatty acid, such that the dielectric

fluid is free of antioxidants and/or external additives and has the following properties: a dielectric strength of 40 kv to 70 kv at a separation of 2 mm, a dielectric constant of 2.5–3.1 at 25°C, and a dissipation factor of 0.05–0.15% at 25°C. The dielectric fluid composition can be obtained by a combination of 95.5% to 99.25% in weight of at least one high-oleic vegetable oil, of 0.25% to 1.5% in weight of grapeseed oil, of 0.25% to 1.5% in weight of sesame seed oil, and of 0.25% to 1.5% in weight of rice bran oil.

Process for producing methyl esters

Lemke, D.W., US8378132, February 19, 2013

Transesterification systems and methods for producing methyl ester are disclosed. In one embodiment, a method for producing methyl ester includes introducing a first charge into a reactor. The first charge contains a triglyceride-containing fat and an alcohol. The method can also include performing a first transesterification reaction in which the triglyceride-containing fat is reacted with the alcohol to produce a first product. The method can further include settling the first product into a methyl ester-containing layer and a glycerol-containing layer, decanting the glycerol-containing layer after settling from the reactor, mixing a second charge with the first product, and performing a second transesterification reaction after mixing the second charge with the first product.

Method for producing fatty acid alkyl ester

Abe, H., *et al.*, Kao Corp., US8378133, February 19, 2013

The present invention provides a method for producing a fatty acid alkyl ester, including step 1 of adding at least one surfactant selected from anionic surfactants, cationic surfactants, and amphoteric surfactants, and water to a crude fatty acid alkyl ester and stirring to obtain a mixture containing aggregates, and step 2 of separating the aggregates from the mixture from step 1 so that the removal rate of steryl glucoside is 60% or more.

Compositions for oral administration of active principles requiring masking of taste

Chacornac, I., and P. Probeck, Sanofi, US8383146, February 26, 2013

A process for the preparation of a composition intended for the oral administration of active principles with unacceptable taste, which comprises from about 15% to 30% of organoleptically unpleasant active ingredient (principle) that is mixed with from about 60% to about 80% of an ester of glycerol or of a fatty acid, to which a wax is optionally added and to which a surfactant is added, and in that it is prepared by a spray-cooling process which can produce a particle size of less than 350 µm.

EXTRACTS & DISTILLATES

Effect of dietary coriander oil and vegetable oil sources on fillet fatty acid composition of rainbow trout

Randall, K.M., *et al.*, *Can. J. Anim. Sci.*, DOI: 10.4141/cjas2013-001, 2013.

A 16-wk feeding trial was conducted to examine the effect of adding coriander oil to vegetable oil (VO) diets on the bioconversion of linoleic acid (LA; 18:2n-6) to arachidonic acid (ARA; 20:4n-6) and α -linolenic acid (ALA; 18:3n-3) to eicosapentaenoic acid (EPA; 20:5n-3) and docosahexaenoic acid (DHA; 22:6n-3) in rainbow trout. The experimental treatments were a 4×2 factorial arrangement of diets using four dietary oils (fish, flax, canola, and camelina oils) and two levels of coriander oil (0 and 5 g kg⁻¹ inclusion levels). Twenty-four tanks of triploid female rainbow trout (130 g initial weight; *n* = 3) were used in the experiment. The experiment lasted 112 d during which fish were fed to satiation twice per day. The fatty acid composition of fillets from coriander-fed fish had increased concentrations of 20:5n-3 and 22:6n-3 (*P* < 0.05). Furthermore, a trend to increased (20:5n-3 + 22:6n-3)/20:4n-6 ratios was seen when coriander oil was added to the diet (*P* = 0.067). These results suggest that the addition of coriander oil to VO diets can significantly increase the bioconversion of 18:3n-3 to 20:5n-3 and 22:6n-3 in rainbow trout.

Egg yolk: structures, functionalities and processes

Anton, M., *J. Sci. Food Agric.* 93:2871–2880, 2013.

Hen egg yolk is an ideal example of natural supramolecular assemblies of lipids and proteins with different organization levels. These assemblies are mainly due to interactions between proteins and phospholipids, and these interactions are essential in understanding and controlling the production of food made with yolk, and particularly emulsions. Furthermore, these assemblies can be modulated by external constraints among which are thermo-mechanical and high-pressure treatments. This review focuses on multiscale structures present in egg yolk, and their modulation by processes, in relation with their emulsifying properties. Egg yolk is mainly composed of two fractions—plasma and granules—which are natural nano- and micro-assemblies. These two fractions possess different composition, structures, and functionalities and exhibit specific behavior under treatments such as high pressure and temperature. Plasma contains a large quantity of lipids structured as lipoproteins (low-density lipoproteins),



Journal of the American Oil Chemists' Society (September)

- Performance of regular and modified canola and soybean oils in rotational frying, Przybylski, R., E. Gruczynska, and F. Aladedunye
- Determination of aroma profiles of olive oils from Turkish olive cultivars, Bayrak, A., M. Kiralan, and H.H. Kara
- Physicochemical properties, volatile compounds and phospholipid classes of silver carp brain lipids, Wang, C., W. Xia, Y. Xu, Q. Jang, and P. Yu
- Synthesis of infant formula fat analogs enriched with DHA from extra virgin olive oil and tripalmitin, Pande, G., J.S.M. Sabir, N.A. Baeshen, and C.C. Akoh
- Strain dependence of the uniaxial compression response of vegetable shortening, Gonzalez-Gutierrez, J., and M.G. Scanlon
- Brewer's yeast as a new source of palmitoleic acid—analysis of triacylglycerols by LC–MS, Řezanka, T., D. Matoulková, I. Kolouchová, J. Masák, and K. Sigler
- Soybean meal retains its nutritional value as an animal feed following *in situ* transesterification, Haas, M.J., R.L. Stroup, and D. Latshaw
- Effect of tocopherols on the anti-polymerization activity of oryzanol and corn steryl ferulates in soybean oil, Winkler-Moser, J.K., K.A. Rennick, H.-S. Hwang, M.A. Berhow, and S.F. Vaughn
- The effect of olive leaf addition on antioxidant content and antioxidant activity of “Memecik” olive oils at two maturity stages, Sevim, D., O. Tuncay, and O. Koseoglu
- Chemical and physical characterization of donkey abdominal fat in comparison with cow, pig and sheep fats, Xu, F., L. Zhang, Y. Cao, L. Yang, H. Zhang, Y. Li, H. Zhao, and Y. Li
- Enhancement of antioxidants in olive oil by foliar fertilization of olive trees, Tekaya, M., B. Mechri, A. Bchir, F. Attia, H. Cheheb, M. Daassa, and M. Hammami
- Corrosion inhibition of mild steel in acidic medium by linseed oil-based imidazoline, Chen, Y., Y.Y. Jiang, H. Chen, Z. Zhang, J.Q. Zhang, and C.N. Cao
- Influence of solid supports on acyl migration in 2-monoacylglycerols: purification of 2-MAG via flash chromatography, Compton, D.L., J.A. Laszlo, and K.O. Evans
- Synthesis of bio-based polyurethane from modified *Prosopis juliflora* oil, Tathe, D.S., and R.N. Jagtap
- Soybean oil-based polyurethane networks: shape-memory effects and surface morphologies, Miao, S., N. Callow, P. Wang, Y. Liu, Z. Su, and S. Zhang
- Impacts of refining and antioxidants on the physico-chemical characteristics and oxidative stability of watermelon seed oil, Wani, A.A., D.S. Sogi, P. Singh, and A. Götz
- Effect of metal type on partial hydrogenation of rapeseed oil-derived FAME, Numwong, N., A. Luengnaruemitchai, N. Chollacoop, and Y. Yoshimura



JOURNAL OF SURFACTANTS AND DETERGENTS

Journal of Surfactants and Detergents (September)

- How to attain an ultralow interfacial tension and a three-phase behavior with a surfactant formulation for enhanced oil recovery: a review. Part 2. Performance improvement trends from Winsor's premise to currently proposed inter- and intramolecular mixtures, Salager, J.-L., A.M. Forgiarini, L. Márquez, L. Manchego, and J. Bullón
- Synthesis and characterization of novel surfactants 1,2,3-tri(2-oxypropylsulfonate-3-alkylether-propoxy) propanes, Zhou, M., J. Zhao, X. Wang, J. Jing, and L. Zhou
- Comparative analysis of rhamnolipids from novel environmental isolates of *Pseudomonas aeruginosa*, Rikalovic, M.G., A.M. Abdel-Mawgoud, E. Déziel, G.D. Gojgic-Cvijovic, Z. Nestorovic, M.M. Vrvic, and I.M. Karadzic
- A process model for approximating the production costs of the fermentative synthesis of sophorolipids, Ashby, R.D., A.J. McAloon, D.K.Y. Solaiman, W.C. Yee, and M. Reed
- Micellization studies of dicationic Gemini surfactants (m-2-m type) in the presence of various counter- and co-ions, Aslam, J., U.S. Siddiqui, W.H. Ansari, and Kabir-ud-Din.
- Structure and biological behaviors of some metallo cationic surfactants, Adawy, A.I. and M.M. Khowdiary
- Antimicrobial catanionic vesicular self-assembly with improved spectrum of action, Chaouat, C., S. Balor, C. Roques, S. Franceschi-Messant, E. Perez, and I. Rico-Lattes
- Micellization and interfacial interaction behaviors of gemini cationic surfactants-CTAB mixed surfactant systems, Mohamed, D.E., N.A. Negm, and M.R. Mishrif
- Gemini cationic surfactants: synthesis and influence of chemical structure on the surface activity, Negm, N.A., M.A. El-Hashash, D.E. Mohamed, J.M. Marquis, and M.M. Khowdiary
- Influence of spacer on association behavior and thermodynamic parameters of dimeric cationic surfactants, Kumar, S. and K. Parikh
- Interactions of glycols with dodecyl isothiuronium cationic surfactant on the surface active parameters, Hefni, H.H. and N.A. Negm
- A corrosion inhibition study of a novel synthesized gemini nonionic surfactant for carbon steel in 1 M HCl solution, El-Tabei, A.S., and M.A. Hegazy
- Synthesis and inhibitory activity of Schiff base surfactants derived from tannic acid and their cobalt (II), manganese (II) and iron (III) complexes against bacteria and fungi, Negm, N.A., A.F. El Faragy, I.A. Mohammad, M.F. Zaki, and M.M. Khowdiary
- Ozonation of anionic and non-ionic surfactants in aqueous solutions: impact on aquatic toxicity, Lechuga, M., A. Fernández-Arteaga, M. Fernández-Serrano, E. Jurado, A. Burgos, and F. Ríos
- Counter-ion effect on micellization of ionic surfactants: a comprehensive understanding with two representatives, sodium dodecyl sulfate (SDS) and dodecyltrimethylammonium bromide (DTAB), Naskar, B., A. Dey, and S.P. Moulik

- On the applicability of the regular solution theory to multi-component systems, Schulz, E.P., J.L.M. Rodriguez, R.M. Minardi, D.B. Miraglia, and P.C. Schulz

Lipids

Lipids (September)

- Combined fish oil and high oleic sunflower oil supplements neutralize their individual effects on the lipid profile of healthy men, Hlais, S., D. El-Bistami, B. El Rahi, M.A. Mattar, and O.A. Obeid
- Improvement of major depression is associated with increased erythrocyte DHA, Meyer, B.J., B.F.S. Grenyer, T. Crowe, A.J. Owen, E.M. Grigonis-Deane, and P.R.C. Howe
- Dietary intake and food sources of EPA, DPA, and DHA in Australian children, Rahmawaty, S., K. Charlton, P. Lyons-Wall, and B.J. Meyer
- Fish oil decreases C-reactive protein/albumin ratio improving nutritional prognosis and plasma fatty acid profile in colorectal cancer patients, Mocellin, M.C., J.A. Pastore e Silva, C.Q. Camargo, M.E.S. Fabre, S. Gevaerd, K. Naliwaiko, Y.M.F. Moreno, E.A. Nunes, and E.B.S.M. Trindade
- The effectiveness of fish oil supplementation in asthmatic rats is limited by an inefficient action on ASM function, Miranda, D.T.S.Z., A.L. Zanatta, B.C.L. Dias, R.T.H. Fogaça, J.B.B. Maurer, L. Donatti, P.C. Calder, and A. Nishiyama
- Taurine supplementation of plant-derived protein and n-3 fatty acids are critical for optimal growth and development of co-bia, *Rachycentron canadum*, Watson, A.M., F.T. Barrows, and A.R. Place
- Nontargeted lipidomic characterization of porcine organs using hydrophilic interaction liquid chromatography and off-line two-dimensional liquid chromatography-electrospray ionization mass spectrometry, Cífková, E., M. Holčápek, and M. Lísa
- Fatty acid profile of the initial oral biofilm (pellicle): an *in-situ* study, Reich, M., K. Kümmerer, A. Al-Ahmad, and C. Hannig
- Streptozotocin-induced diabetes partially attenuates the effects of a high-fat diet on liver and brain fatty acid composition in mice, Levant, B., M.K. Ozias, B.L. Guilford, and D.E. Wright
- Quantitative analysis of phytosterols in edible oils using APCI liquid chromatography-tandem mass spectrometry, Mo, S., L. Dong, W.J. Hurst, and R.B. van Breemen



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whereas granules are mainly composed of proteins aggregated in micrometric assemblies. If plasma is responsible for the important emulsifying properties of yolk, granules bring interesting emulsifying properties when assemblies are in the form of micelles in presence of salts. High-pressure or thermal treatments, applied before or after emulsion fabrication, alter their functionalities and could be used to commercially exploit these fractions.

Thermal and chemical evaluation of naturally auto-oxidised virgin olive oils: a correlation study

Chiavaro, E., *et al.*, *J. Sci. Food Agric.* 93:2909–2916, 2013.

The nature of the relationship between differential scanning calorimetry thermal properties and the oxidation and hydrolysis compounds formed during a real auto-oxidation process in virgin olive oils has not been evaluated so far, as these samples are difficult to find. In this work, 21 samples of virgin olive oil, stored under ideal conditions since their years of production (production range 1991–2005) to develop the natural auto-oxidation process, were analyzed in order to evaluate this relationship. Oils stored the longest time showed the highest hydrolytic degradation while the others exhibited higher contents of oxidized fatty acids and triacylglycerols, instead. Thermal properties of transitions were differently influenced by degradation compounds with the onset of both the cooling and heating profiles particularly influenced by diacylglycerols and oxidized lipids. Chemical data and thermal properties were correlated by using principal component analysis. Twenty-three variables were selected for the analysis with the first component fully segregating samples into two groups according to the year of storage and the level of hydrolysis and/or oxidation, on the basis of selected thermal properties obtained by cooling and heating transitions. These preliminary findings showed that differential scanning calorimetry could be considered a useful tool to evaluate lipid degradation in virgin olive oils, indicating its value as a support to chemical techniques.

Methods to recover value-added coproducts from dry grind processing of grains into fuel ethanol

Liu, K., and F.T. Barrows, *J. Agric. Food Chem.* 61:7325–7332, 2013.

Three methods are described to fractionate condensed distillers solubles (CDS) into several new co-products, including a protein–mineral fraction and a glycerol fraction by a chemical method; a protein fraction, an oil fraction, and a glycerol–mineral fraction by a physical method; or a protein fraction, an oil fraction, a mineral fraction, and a glycerol fraction by a physicochemical method. Processing factors (ethanol concentration and centrifuge force) were also investigated. Results show that the three methods separated CDS into different fractions, with each fraction enriched with one or more of the five components

(protein, oil, ash, glycerol, and other carbohydrates) and thus having different targeted end uses. Furthermore, because glycerol, a hygroscopic substance, was mostly shifted to the glycerol or glycerol–mineral fraction, the other fractions had much faster moisture reduction rates than CDS upon drying in a forced air oven at 60°C. Thus, these methods could effectively solve the dewatering problem of CDS, allowing elimination of the current industrial practice of blending distiller wet grains with CDS for drying together and production of distiller dried grains as a stand-alone co-product in addition to a few new fractions.

Isolation and structure elucidation of avocado seed (*Persea americana*) lipid derivatives that inhibit *Clostridium sporogenes* endospore germination

Rodríguez-Sánchez, D.G., *et al.*, *J. Agric. Food Chem.* 61:7403–7411, 2013.

Avocado fruit extracts are known to exhibit antimicrobial properties. However, the effects on bacterial endospores and the identity of antimicrobial compounds have not been fully elucidated. In this study, avocado seed extracts were tested against *Clostridium sporogenes* vegetative cells and active endospores. Bioassay-guided purification of a crude extract based on inhibitory properties linked antimicrobial action to six lipid derivatives from the family of acetogenin compounds. Two new structures and four compounds known to exist in nature were identified as responsible for the activity. Structurally, most potent molecules shared features of an acetyl moiety and a *trans*-enone group. All extracts produced inhibition zones on vegetative cells and active endospores. Minimum inhibitory concentrations (MIC) of isolated molecules ranged from 7.8 to 15.6 µg/mL, and bactericidal effects were observed for an enriched fraction at 19.5 µg/mL. Identified molecules showed potential as natural alternatives to additives and antibiotics used by the food and pharmaceutical industries to inhibit Gram-positive spore-forming bacteria.

Increasing seed oil content in Brassica species through breeding and biotechnology

Rahman, H., *et al.*, *Lipid Technol.* 25:182–185, 2013.

Increasing the seed oil content of Brassica species and other major oilseed crops is of paramount importance in maintaining a future supply of vegetable oil for a growing global population. Currently, commercially available Brassica species with enhanced seed oil content have all been developed through plant breeding. Many quantitative trait loci including gene interactions are involved in the control of seed oil content. Despite this complexity, manipulation of specific steps in storage lipid biosynthesis using genetic engineering has resulted in transgenic lines of *Brassica napus* with increased seed oil content. Recent studies suggest

that engineering of seed oil content can be guided using methods in metabolic analysis.

The role of short-chain fatty acids in the interplay between diet, gut microbiota, and host energy metabolism

den Besten, G., et al., *J. Lipid Res.* 54:2325–2340, 2013.

Short-chain fatty acids (SCFA), the end products of fermentation of dietary fibers by the anaerobic intestinal microbiota, have been shown to exert multiple beneficial effects on mammalian energy metabolism. The mechanisms underlying these effects are the subject of intensive research and encompass the complex interplay between diet, gut microbiota, and host energy metabolism. This review summarizes the role of SCFAs in host energy metabolism, starting from the production by the gut microbiota to the uptake by the host and ending with the effects on host metabolism. There are interesting leads on the underlying molecular mechanisms, but there are also many apparently contradictory results. A coherent understanding of the multilevel network in which SCFAs exert their effects is hampered by the lack of quantitative data on actual fluxes of SCFAs and metabolic processes regulated by SCFAs. In this review we address questions that, when answered, will bring us a great step forward in elucidating the role of SCFAs in mammalian energy metabolism.

Fuel properties of methyl esters of borage and black currant oils containing methyl γ -linolenate

Knothe, G., *Eur. J. Lipid Sci. Tech.* 115:901–908, 2013.

In this work, the methyl esters of two oils enriched in γ -linolenic acid (6Z,9Z,12Z-octadecatrienoic acid) were prepared under the aspect of evaluating their properties, including potential fuel properties. One oil is black currant oil in which γ -linolenic and α -linolenic (9Z,12Z,15Z-octadecatrienoic acid) are relatively evenly distributed, and the other oil is borage oil, which mainly contains γ -linolenic acid as C18:3 species. The fatty acid profiles of both oils confirm literature results. The cetane number (CN) of neat methyl γ -linolenate was also determined for the first time as 29.2, which is slightly higher than that of the more common methyl α -linolenate. The methyl esters (biodiesel) from such oils meet most property specifications in biodiesel standards with the exception of feedstock restrictions on highly unsaturated fatty acid chains, although CNs are lower and antioxidants are required for oxidation stability. Although, due to their nutritional value and limited occurrence, these oils are unlikely biodiesel feedstocks themselves, their methyl esters may be seen as models for similar derivatives from other feedstocks with elevated levels of unsaturation, including algal oils. The ^1H and ^{13}C NMR spectra of black currant and borage methyl esters are also reported.

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



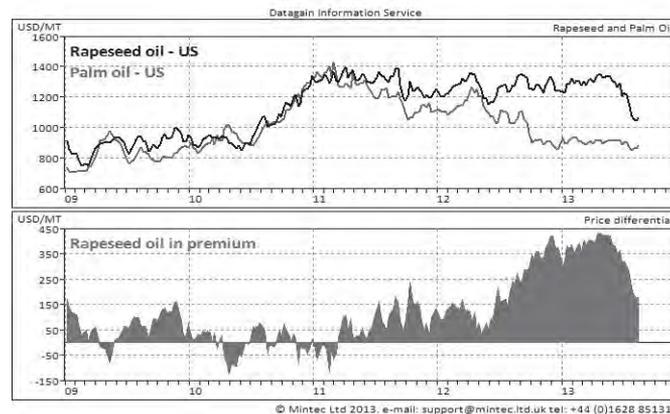
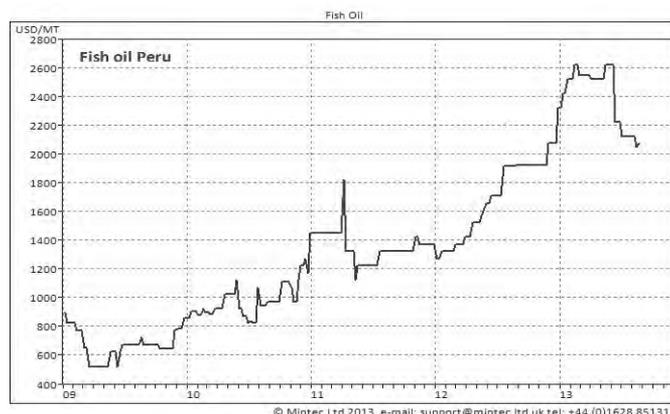
STATISTICAL ANALYSIS FROM MINTEC

James Hutchings at Mintec

Fish oil prices rose to a record level in the first half of 2013 due to restricted supply in the key producing countries of Peru and Chile and strong global demand. The Peruvian summer fishing quota has been reduced by 0.7 MMT to 2.05 MMT and fishing in Chile has been poor so far this year. Global fish oil production in Q1 2013 was down 23% year-on-year to 86,000 MMT. Prices have fallen back recently as the fish oil market has been influenced by the drop seen in the wider vegetable oil market, with fish feed manufacturers under pressure to reduce the fish oil proportions in their products.

Rapeseed (canola) oil is a relatively small market in the United States, however, prices have fallen sharply to their lowest level since 2010. The global rapeseed outlook for production in 2013/14 is now forecast to reach 66.4 MMT, up 7% year-on-year. Rapeseed production in Canada, the world's top exporter, is forecast to rise by 15% year-on-year to 15.3 MMT. Canadian exports are expected to rise to 7.5 MMT, an increase of 5% year-on-year, due to higher exportable supplies.

This sharp drop in rapeseed oil prices has significantly reduced its premium over palm oil, even despite the drop in palm oil prices. The drop in palm oil was caused by reduced demand from India (the world's largest palm oil consumer).



Mintec Limited Mintec is the principal independent source of global pricing information for commodities and raw materials. Mintec has a suite of unique procurement tools and training courses designed to assist supply chain professionals in their daily tasks of reporting, analyzing, and interpreting market information. Mintec supports sales and purchasing professionals in their commodity decisions, by improving efficiency, minimizing commodity risk and ultimately gaining a commercial advantage.

Mintec Ltd., 9 The Courtyard, Glory Park, Wooburn Green, High Wycombe, Buckinghamshire HP10 ODG, United Kingdom; Tel: +44 (0)1628 851313; Fax: +44 (0)1628 851321; Email: support@mintec.ltd.uk; Web: www.mintec.ltd.uk

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.

Vince Vavpot has more than 35 years of experience in oilseed processing. Though currently retired, he remains affiliated with Anderson International Corp., where he has worked for 39 years. His previous position before retirement was as vice president of Anderson's vegetable oil and animal feeds business group.



Describe your career path.

After graduating from the University of Dayton in Ohio with an associate degree in chemical engineering in 1963, I worked for three years at National Cash Register (NCR) as a laboratory technician in the company's Dayton location, which was where NCR's headquarters were at the time. I went back to school after leaving NCR and graduated in 1968 from the Cleveland State University's Fenn College of Engineering with a bachelors' degree in mechanical engineering. I then spent five years with Dow Chemical (Midland, Michigan, USA) as a process engineer with the plastic and packaging division, which was located in Cleveland, Ohio. In 1974, I joined Anderson International Corp (Stow, Ohio, USA). I was hired as a process

engineer for the dairy equipment product line for use of evaporators and spray dryers to make condensed and powdered milk.

Why did you join AOCS?

I got firmly involved in oilseed processing in 1978, when I became sales engineering manager at Anderson and learned about the design and specification for oilseed processing machinery. I thought it was a good idea to join and come to the exhibitions, get involved and meet people.

What do you love about your job?

What I've always enjoyed about this work is getting out there and helping people improve the performance of their businesses with technology. After a certain number of years, you've self-educated yourself to where you've become a consultant, a teacher. You travel around the world to plants that have your equipment and meet with people that want to expand. A lot of times they work with blinders on; they only have their way of doing things. We see a variety of ways to solve problems. It's a pleasure to assist and help these people.

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

When I started 39 years ago, if you invented a new process machine or made a considerable improvement, this would last for 10 years. Today, if you aren't continuously improving your process machinery you are falling behind the needs of the industry. And these improvements must reduce operating costs and improve oil and meal quality.

How has your industry changed since you entered the field? The most notable changes have been the vast consolidation of oil milling from small mills of 50–200 short tons per day to 1,000–5,000 tons/day. And processing changed by the addition of solvent extraction technology in the place of mechanical crush facilities. When I started, there were over 200 oil mills in the United States. Cottonseed was still king, but no more. Now mechanical extraction is returning as a "natural oil" supply.

Do you have any advice for those looking to enter your field? Be very attentive to what the processor is telling you, as within those comments you will determine their needs. Work hard to fulfill the needs and you will develop a true and honest business relationship. The worldwide oilseed industry speaks the same language no matter where your travels take you. Being respectfully attentive to processor needs crosses all cultural barriers.

How do you see the industry changing in the next five years? Consumers are becoming more knowledgeable about their foods in terms of health, dietary benefits, sanitary conditions, effects of contamination, and sources of their food. The trend is for more healthful oils and meals, growth in natural oils, higher nutritional values in their oils. The trend will be for niche market development and at the same time having commodity oils available.

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

My mentor was Maurice Williams, a biologist by education, who served Anderson in R&D for over 50 years as a technician, then pilot plant manager, then director of R&D. A man who was our teacher, who always told us the best methods and the impact our machinery had on oil and meal quality from the viewpoint of a biologist, he always made himself available to help every one of us understand the process working of our machinery. He authored over 35 articles for the feeds and oilseed industry, wrote oilseed processing chapters for several high-quality technical publishers such as Bailey's Industrial Oil and Fat Products, and was asked to edit many others, even from competitors. An honest man, a gentleman, a scholar, and a teacher. He was a multiple annual lecturer at Texas A&M short courses for oilseed and animal feeds processing for over 25 years.

Describe a memorable job experience.

During my stint as regional sales manager for East Asia, I commissioned a copra mill in the Philippines. On this occasion, I worked for 36 hours straight. The temperature and humidity were quite high. After the 36 hours I was found lying on a stack of burlap bags full of copra with copra bugs crawling all over my body. (Copra bugs do not bite, fortunately.) I was led to a primitive shower stall where I was helped to stand upright while three women poured water over my body to cool me down, then led to a cot where six fans were used to continue cooling my body and allowing me to sleep. During the 36 hours, I was given copious amounts of water, ice tea, soups, and bottled refreshments, yet throughout this time I never had to relieve myself. This prevented me from getting sick from heat stroke. It could have been quite a bad experience but for the care given us by the Philippine people on hand.

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

Association involvement is the best way. One of the best things I did was I let myself be talked into becoming the exhibitor committee chair for the 89th AOCS Annual Meeting & Expo in Chicago, Illinois, in 1998. I became friends with not only competitors but also other companies in the organization. You get to meet new contacts as well.

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

It's become quite difficult [to advance]. When I started with the company, there were a lot of oil mills in the US alone. They had engineers and salespeople, and there were more opportunities for advancement. Due to the [rise of] major processing plants in the US, you've lost that a bit. In our company, we have to know about the oilseeds and the products. Now, where do you find those people? You find them at the major companies. Your choices are limited. You almost have to start from scratch, but the opportunities are there.

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

Oilseed processing is quite specific. Ninety percent is specific and 10% is culturally rounded. [But] we're dealing with a global audience. You need to be pretty well-rounded in the oilseed industry, and if you look at the demographics [at AOCS meetings], there's a pretty good proportion of people from all around the world involved. You should be culturally educated.

What is your opinion towards the value of obtaining or possessing a graduate degree during a challenging economy? Graduate degrees in chemical and mechanical engineering are quite important, especially in the agricultural sciences. ■



2012–2013 AOCS Laboratory Proficiency Program Winners

The AOCS Laboratory Proficiency Program (LPP), formerly known as the Smalley Check Sample Program, is the world's most extensive and respected collaborative proficiency testing program for oil- and fat-related commodities, oilseeds, oilseed meals, and edible fats. More than 500 chemists participate to verify their lab's quality control. Participants use AOCS or similar methods for sample analysis and then compare their results with those from a large cross section of other laboratories using the same methods and samples. For more information, contact Evelyn King at AOCS Technical Services (phone: +1 217-693-4815; fax: +1 217-693-4859; email: evelynk@aoacs.org).

**Aflatoxin in Almonds
First Place**

Donna Dean-Zavala
Blue Diamond Growers
Sacramento, California
95812-1768
USA

**Aflatoxin in Corn Meal
First Place**

Kendrick Albert
Purdue University
West Lafayette, Indiana
47907-2063
USA

**Aflatoxin in Corn Meal
Honorable Mention**

Dr. Jim Balthrop Jr.
Office of the Texas State
Chemist
College Station, Texas
77843
USA

**Aflatoxin in Corn Meal
Test Kit
First Place**

Dr. Jim Balthrop Jr.
Office of the Texas State
Chemist

College Station, TX 77843
USA

**Aflatoxin in Corn Meal
Test Kit**

Honorable Mention
Wayne Adcock
State Chemical Laboratory
Auburn, Alabama
36832-1620
USA

**Aflatoxin in Corn Meal
Test Kit
Honorable Mention**

Denver Haynes
Tyson Foods Inc.
Wilkesboro, North Carolina
28697
USA

**Aflatoxin in Cottonseed
First Place**

John Wetmore
Wetmore Enviro Lab Ltd.
Casa Grande, Arizona 85122
USA

**Aflatoxin in Peanut Butter
First Place**

Albany Analytical Team
Jimmie Brown
JLA USA
Albany, Georgia 31721
USA

**Aflatoxin in Peanut Paste
First Place**

Prashant Tank
IEH Laboratories & Consulting
Group
Lost Hills, California 93249
USA

**Aflatoxin in Peanut Paste
Honorable Mention**

Mariana Astore
SGS Argentina S.A. Florencio
Varela
Buenos Aires C1430 DNN
Argentina

**Aflatoxin in Peanut Paste
Honorable Mention**

Mariana Astore
SGS Argentina SA Alejandro
Roca
Buenos Aires C1430 DNN
Argentina

**Aflatoxin in Peanut Paste
Honorable Mention**

JLA Argentina SA
General Cabrera, Cordoba X
5809 BAS
Argentina

**Aflatoxin in Peanut Paste
Test Kit**

First Place
Sharyn Passeretti, Matthew
Gilbert, & Dillon Oostendorp
ABC Research Laboratories
Gainesville, Florida 32607
USA

**Aflatoxin in Peanut Paste
Test Kit
Honorable Mention**

Headland Analytical Team
Kathy Poke
JLA I
Headland, Alabama 36345
USA

**Aflatoxin in Peanut Paste
Test Kit**

Honorable Mention
Dawson Analytical Team
Mary Jane Corley
JLA USA
Dawson, Georgia 39842
USA

**Aflatoxin in Pistachios
First Place**

Melissa Medina
DFA California—Kerman Lab
Kerman, California 93630
USA

**Cholesterol
First Place**

Ardin Backous, Anders
Thomsen, Keith Persons, &
Kent Karsjens
Eurofins Scientific
Des Moines, Iowa
50321-3157
USA

**Cholesterol
Honorable Mention**

Ann Lindgren & Darcy
Schroeder
Hormel Foods LLC
Austin, Minnesota 55912
USA

**Cholesterol
Honorable Mention**

Jocelyn Alfieri
Silliker Canada Co.
Markham, Ontario L3R 5V5
Canada

**Cottonseed Oil
First Place**

Donald Britton
Mid-Continent Laboratories
Memphis, Tennessee 38101
USA

**Edible Fat
First Place**

James Houghton
AAK
Louisville, Kentucky 40208
USA

**Edible Fat
Honorable Mention**

Felicia Melendez
Ag Processing, Inc.
Hastings, Nebraska 68901
USA

**Edible Fat
Honorable Mention**

Jack M. Stearns
AAK
Louisville, Kentucky 40208
USA

**Edible Fat
Honorable Mention**

Travis Patterson
Ag Processing, Inc.

Hastings, Nebraska 68901
USA

**Edible Fat
Honorable Mention**

Tracie McClure
Ag Processing, Inc.
Hastings, Nebraska 68901
USA

**Edible Fat
Honorable Mention**

Jerry Buttell
Ag Processing, Inc.
Hastings, Nebraska 68901
USA

**Feed Microscopy
First Place**

Marion Smith
Canadian Food Inspection
Agency
Ottawa, Ontario K1A 0C6
Canada

**Feed Microscopy
Second Place**

Yuan-te Fu
NCDA&CS Food & Drug
Protection
Raleigh, North Carolina
27607
USA

**Feed Microscopy
Third Place**

Evan Balaskas
Canadian Food Inspection
Agency
Ottawa, Ontario K1A 0C6
Canada

**Feed Microscopy
Honorable Mention**

Dr. Jim Balthrop Jr.
Office of the Texas State
Chemist
College Station, Texas 77843
USA

**Feed Microscopy
Honorable Mention**

Elizabeth Krzykwa
Canadian Food Inspection
Agency
Ottawa, Ontario K1A 0C6
Canada

**Fish Meal
First Place**

Pete Cartwright
NJ Feed Lab Inc.
Trenton, New Jersey 08638
USA

**Fish Meal
Honorable Mention**

Carmen Catter
International Analytical
Services
Lima Lima 32
Peru

**Fish Meal
Honorable Mention**

Paul Thionville, Boyce
Butler, Andre Thionville, &
Kristopher Williams
Thionville Laboratories, Inc.
New Orleans, Louisiana
70123
USA

**Fumonisin in Corn
First Place**

Dr. Jim Balthrop Jr.
Office of the Texas State
Chemist
College Station, Texas
77843
USA

**Fumonisin in Corn Test Kit
First Place**

Cheri Turman
Analytical Food Laboratories
Grand Prairie, Texas 75050
USA

**Gas Chromatography
First Place**

Dr. Hazimah Abu Hassan,
Mrs. Hajar Musa
Malaysian Palm Oil Board
B.B. Bangi, Selangor 43650
Malaysia

**Gas Chromatography
Honorable Mention**

Owensboro Grain Edible Oils
Owensboro, Kentucky
42301
USA

**Gas Chromatography
Honorable Mention**

Don Pepper
ADM Windsor F20
Windsor, Ontario N9C 4G9
Canada

**Gas Chromatography
Honorable Mention**

Carl Whitney
ConAgra Grocery Products
Memphis, Tennessee 38104
USA

**Gas Chromatography
Honorable Mention**

Ann Puvirajah
Canadian Grain Commission
Winnipeg, Manitoba R3C 3G7
Canada

**Gas Chromatography
Honorable Mention**

Ang Chee Loong
PGEO Edible Oils Sdn. Bhd.
Pasir Gudang, Johor 81707
Malaysia

**Gas Chromatography
Honorable Mention**

Kim Jennings-Wilson
Stratas Foods
Quincy, Illinois 62306
USA

**Gas Chromatography
Honorable Mention**

Jamie Ayton
NSW Department of Primary
Industries
Wagga Wagga, NSW 2650
Australia

**Gas Chromatography
Honorable Mention**

Randall Hoffman
ADM Valdosta 564/217
Valdosta, Georgia 31601
USA

**Gas Chromatography
Honorable Mention**

Abdul Bath
Adams Vegetable Oils Inc.
Arbuckle, California 95912
USA

**Gas Chromatography
Honorable Mention**

Pete Cartwright
NJ Feed Lab Inc.
Trenton, New Jersey 08638
USA

**Gas Chromatography
Honorable Mention**

Shirley Elliott
Darling International
Ankeny, Iowa 50021
USA

**Gas Chromatography
Honorable Mention**

Rudy Fulawka
Bayer CropScience Inc.
Saskatoon, Saskatchewan
S7K 3J9
Canada

**Marine Oil
First Place**

Otelia Robertson
Omega Protein Inc.
Reedville, Virginia 22539
USA

**Marine Oil
Honorable Mention**

Melissa V. Thrift
Omega Protein Inc.–Health
Science Ctr.
Reedville, Virginia 22539
USA

**Marine Oil
Honorable Mention**

Bertha Sulca
SGS del Peru S.A.C.
Lima 27-0125
Peru

**Marine Oil Fatty Acid Profile
First Place**

Analytical Serviced
POS Bio-Sciences
Saskatoon, Saskatchewan
S7N 2R4
Canada

**Marine Oil Fatty Acid Profile
Honorable Mention**

Pete Cartwright
NJ Feed Lab Inc.
Trenton, New Jersey 08638
USA

**Marine Oil Fatty Acid Profile
Honorable Mention**

Covance Labs
Madison, Wisconsin 53704
USA

**Mixed Seeds (Canola)
First Place**

Sovena Oilseeds Laboratory
Sovena Oilseeds
Almada 2801-801
Portugal

**Mixed Seeds (Safflower)
First Place (tie)**

Paul Thionville, Boyce Butler,
Andre Thionville & Kristopher
Williams
Thionville Laboratories, Inc.
New Orleans, Louisiana
70123
USA

**Mixed Seeds (Safflower)
First Place (tie)**

Cathy Sun
SGS Canada

Burnaby, British Columbia
V5A 4W4
Canada

**Mixed Seeds (Sunflower)
First Place**

Sovena Oilseeds Laboratory
Sovena Oilseeds
Almada 2801-801
Portugal

**NIOP Fats and Oils
First Place**

George Hicks
Dallas Group of America
Jeffersonville, Indiana 47130
USA

**NIOP Fats and Oils
Honorable Mention**

Joseph Caldwell
Dallas Group of America
Jeffersonville, Indiana 47130
USA

**Nutritional Labeling
First Place**

Ardin Backous, Anders
Thomsen, Keith Persons, &
Kent Karsjens
Eurofins Scientific
Des Moines, Iowa
50321-3157
USA

**Nutritional Labeling
Honorable Mention**

Sonia Bouchard
CFIA Food Lab
Lonqueuil, Québec J4K 1C7
Canada

**Oilseed Meal 100% Crude
Fiber**

First Place
Frank Hahn
Hahn Laboratories Inc.
Columbia, South Carolina
29201
USA

**Oilseed Meal 100% Crude
Fiber**

Honorable Mention
Brad Newton Beavers and
Jennie Stewart
Carolina Analytical Services
Bear Creek, North Carolina
27207
USA

**Oilseed Meal 100% Crude
Fiber**

Honorable Mention

Mumtaz Haider
Inspectorate America Corp.
Galena Park, Texas 77547
USA

**Oilseed Meal 100% Crude
Fiber**

Honorable Mention
John Reuther and
Eric de Ronde
Eurofins Central Analytical
Labs
Metairie, Louisiana 70001
USA

**Oilseed Meal 100% Crude
Fiber**

Honorable Mention
Lynn Hawkins and
Michael Hawkins
Barrow-Agee
Memphis, Tennessee
38116-3507
USA

**Oilseed Meal 100% Crude
fiber**

Honorable Mention
Frank Tenent and
Edgar Tenent
K-Testing Lab Inc.
Memphis, Tennessee 38116
USA

**Oilseed Meal 100% Moisture
First Place (tie)**

Ann Puvirajah
Canadian Grain Commission
Winnipeg, Manitoba R3C 3G7
Canada

**Oilseed Meal 100% Moisture
First Place (tie)**

Brad Newton Beavers and
Jennie Stewart
Carolina Analytical Services
Bear Creek, North Carolina
27207
USA

**Oilseed Meal 100% Moisture
Honorable Mention**

Trevor Meredith
Solbar Hatzor LTDCHS Israel
Ashdod 77121
Israel

**Oilseed Meal 100% Moisture
Honorable Mention**

Sandy Harrison
Illinois Crop Improvement
Assn.
Champaign, Illinois 61822
USA

Oilseed Meal 100% Moisture Honorable Mention

John Reuther and Eric de Ronde
Eurofins Central Analytical Labs
Metairie, Louisiana 70001
USA

Oilseed Meal 100% Nitrogen Ba 4d-90 Method First Place

Mumtaz Haider
Inspectorate America Corp.
Galena Park, Texas 77547
USA

Oilseed Meal 100% Nitrogen Ba 4d-90 Method Honorable Mention

Foong Ming Koh
SGS North America
Agricultural Div.
Deer Park, Texas 77536
USA

Oilseed Meal 100% Nitrogen Ba 4e-93 Method First Place

Paul Thionville, Boyce Butler, Andre Thionville, & Kristopher Williams
Thionville Laboratories, Inc.
New Orleans, Louisiana 70123
USA

Oilseed Meal 100% Nitrogen Ba 4e-93 Method Honorable Mention

Lynn Hawkins and Michael Hawkins
Barrow-Agee
Memphis, Tennessee 38116-3507
USA

Oilseed Meal 100% Nitrogen Ba 4e-93 Method Honorable Mention

Frank Tenent and Edgar Tenent
K-Testing Lab Inc.
Memphis, Tennessee 38116
USA

Oilseed Meal 100% Nitrogen Ba 4e-93 Method Honorable Mention

Ardin Backous, Anders Thomsen, Keith Persons, & Kent Karsjens
Eurofins Scientific
Des Moines, Iowa

50321-3157
USA

Oilseed Meal 100% Oil First Place

Robert Carr Jr.
Russell Marine Group-PNW, LLC
Portland, Oregon 97210
USA

Oilseed Meal 100% Oil Honorable Mention

John Reuther and Eric de Ronde
Eurofins Central Analytical Labs
Metairie, Louisiana 70001
USA

Oilseed Meal 100% Oil Honorable Mention

Melinda Graham
Hartsville Oil Mill
Darlington, South Carolina 29540-1027
USA

Oilseed Meal 100% Oil Honorable Mention

Renato M. Ramos
Admiral Testing Services
Luling, Louisiana 70070
USA

Oilseed Meal 100% Oil Honorable Mention

Mumtaz Haider
Inspectorate America Corp
Galena Park, TX 77547
USA

Oilseed Meal Overall First Place

Mumtaz Haider
Inspectorate America Corp
Galena Park, Texas 77547
USA

Oilseed Meal Overall Honorable Mention

Frank Tenent and Edgar Tenent
K-Testing Lab Inc.
Memphis, Tennessee 38116
USA

Oilseed Meal Overall Honorable Mention

Ardin Backous, Anders Thomsen, Keith Persons, & Kent Karsjens
Eurofins Scientific

Des Moines, Iowa 50321-3157
USA

Oilseed Meal Overall Honorable Mention

Frank Hahn
Hahn Laboratories Inc.
Columbia, South Carolina 29201
USA

Oilseed Meal Overall Honorable Mention

Lynn Hawkins and Michael Hawkins
Barrow-Agee
Memphis, Tennessee 38116-3507
USA

Olive Oil Part A First Place

Dr. Giorgio Cardone
Chemiservice SRL
Monopoli, Bari 70043
Italy

Olive Oil Part A Honorable Mention

Nikos Fakourelis
Eas Pezon
Peza Union of Agricultural Coop
Heraklion, Crete 70100
Greece

Olive Oil Part B First Place

Dr. Giorgio Cardone
Chemiservice SRL
Monopoli, Bari 70043
Italy

Olive Oil Part C First Place

Dr. Giorgio Cardone
Chemiservice SRL
Monopoli, Bari 70043
Italy

Omega 3 Nutraceutical Oils First Place

Dr. Edith Von Kries
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A simple and efficient one-step method for the extraction of lipid-soluble compounds from tissue and food

Adam Methereel

There are many reasons for measuring lipids in a wide array of sample matrices. Lipids such as omega-3 fatty acids, *trans* fatty acids, and cholesterol play significant roles in human health and are commonly determined in blood and tissue for biomarker assessments. Lipid analysis is further required for government-mandated food labeling and has the potential to support efforts in food traceability. Other important applications of lipid analyses include biodiesel production for greener fuels and basic biomedical and biomolecular research. This broad spectrum of applications is accompanied by an equally broad number of tissue and food samples on which to perform fatty acid and lipid analysis. Regardless of whether one is performing lipid assessments in humans, rats, mice, eggs, or milk, or whether that assessment is for purposes of omega-3 profiling, cell function quantification, or food labeling, the single most common theme among them all is the complex analytical process.

Traditionally, lipid analysis is a tedious process requiring (i) sample collection, (ii) sample storage, (iii) sample preparation, (iv) measurement of analytes, and (v) data handling (Stark, 2008). Sample preparation can then be broken down further into (i) homogenization, (ii) extraction, and (iii) derivatization steps. Occasionally, and depending on the final goal of analysis, one or more of these steps can be combined into a single step, but these steps are rarely completely removed. Adding to the significant requirements of fatty acid analysis is the potential for additional analytical steps such as thin-layer chromatography for the separation of more specific fatty acid fractions. Although the analytical process for lipids is a challenge, significant advances have

been made over the past 10–15 years in collection (e.g., fingertip prick blood collection) (Marangoni *et al.*, 2004), extraction (e.g., ultrasonic energy) (Luque-Garcia and Luque de Castro, 2007), derivatization (e.g., microwave energy) (Armstrong *et al.*, 2008), and fast gas chromatographic steps (Masood *et al.*, 2008). Despite these advances, more traditional methods remain entrenched as standard procedure in many research institutions.

LIMITATIONS OF CURRENT METHODS

A comparison of the three most commonly used fatty acid extraction methods from food was carried out in flaxseed (Taha *et al.*, 2012). These three methods are (i) the International Organization for Standardization (ISO) method, (ii) the AOAC International method, used most commonly in industry for nutritional labeling purposes, and (iii) the Folch method, used most commonly in academic settings for agricultural, nutritional, and biochemical research. These methods commonly require between 4 and 24 hours to complete and can require up to 10 grams (g) of

CONTINUED ON NEXT PAGE

- **Traditional methods of analyzing lipids in tissue and food can take between 4 and 24 hours to complete and may require up to 10 grams of sample and as much as 500 milliliters of solvents for the extraction process.**
- **The Folch method—commonly used in agricultural, nutritional, and biochemical research—requires manual pipetting, for which yields may vary from day to day and from individual to individual, thus making comparative analysis difficult.**
- **The following article describes a simple and efficient one-step method for extracting cholesterol and fatty acids that requires no pipetting, no centrifugation, and no mixing of standards and solvents. The method increases analytical precision while reducing the time required to perform the separation technique.**

FIG. 1.
Image of extraction filter
equipped with syringe.

sample and as much as 500 milliliters (mL) of solvents for the extraction process. Specifically, the Folch method uses a 2:1:0.8 chloroform/methanol/aqueous buffer solvent mixture. Extraction of the lipids is performed with 2:1 chloroform/methanol, and the aqueous buffer is added to facilitate the separation of polar (methanol) and non-polar (chloroform) layers. This separation step requires centrifugation of the mixture followed by the manual pipetting of the bottom lipid-containing chloroform layer for the isolation of extracted lipids.

Manual laboratory techniques can yield significant variability from day to day and sample to sample within an individual. This variability is even larger between individuals within the same lab or between different labs and makes comparative analyses difficult. Methods and techniques designed to automate and simplify these procedures can reduce variability thereby improving analysis between multiple datasets.

A SIMPLE AND ACCURATE ONE-STEP EXTRACTION METHOD

Certo Labs has recently developed both a fatty acid and a cholesterol extraction kit to expedite the process of extracting lipids from various tissue

and food samples. These extraction kits are based on the Folch extraction method, which requires significant experience and skill to achieve accuracy and precision. The aims are to simplify the method without compromising this accuracy and precision and to allow non-experts to perform the lipid extractions with ease and confidence, and in significantly less time than would traditionally be required.

Using the Certo kit requires no pipetting, no centrifugation, and no mixing of standards and solvents. The pre-mixed extraction solvents are added to the tissue or food sample, and subsequently homogenized and extracted as normal. Following this, the aqueous buffer is added and the sample/solvent, inverted twice, added to the extraction syringe and passed through the treated filter (Fig. 1) by the gentle plunging of the syringe. The lipid-containing chloroform mixture is collected, and the non-lipid-containing water/methanol is selectively trapped and left behind in the filter. The Certo kit uses a 2:1:0.5 chloroform/methanol/aqueous buffer solvent mixture that is slightly different from the Folch method (2:1:0.8). The Folch solvent ratios are intended for optimal separation of polar and non-polar phases by centrifugation. Conversely, the modification of this ratio in the Certo kit is intended for optimal separation of polar and nonpolar phases by the aqueous filter.

The Certo kit has been tested for both cholesterol and fatty acids in a variety of foods and tissues and compared with the classic Folch method. Accuracy of lipid concentrations (mg/g) in egg and brain samples was similar (Fig. 2). The Certo kit also provided greater precision as the relative standard deviations averaged 4% in egg and 10% in brain, whereas Folch results averaged 6% in egg and 17% in brain. We hypothesize this is due to the removal of human error associated with manual pipetting, and it should be noted the Folch results were generated from a highly trained technician with 5+ years of experience in fatty acid determinations.

In addition to the increased analytical precision, using the Certo kit appreciably reduces the time required to perform the separation technique. The extraction time for the egg and brain data presented herein (6 samples each) required only 3 minutes using the Certo kit, while it took 12 minutes using the conventional Folch method. This correlates to an average of 30 seconds per extraction when using the Certo kit and 2 minutes per extraction for the Folch method.

Currently, the Certo kit has been assessed and developed for use in cholesterol and fatty acid extractions from approximately 100 mg of tissue or food samples; however, the kit has the potential for application to the preparation of any lipid-soluble compound including vitamins A, D and E; pesticides; and antibiotics. In addition, a larger-volume extraction kit that will allow for the analysis of up to 1 g of a sample and uses 20 mL of 2:1 chloroform/methanol for extraction is in development. These simple, easy, and high-throughput extraction kits promise to save academic and industry researchers' additional time and costs without compromising the accuracy and precision of the determination of lipid compounds.

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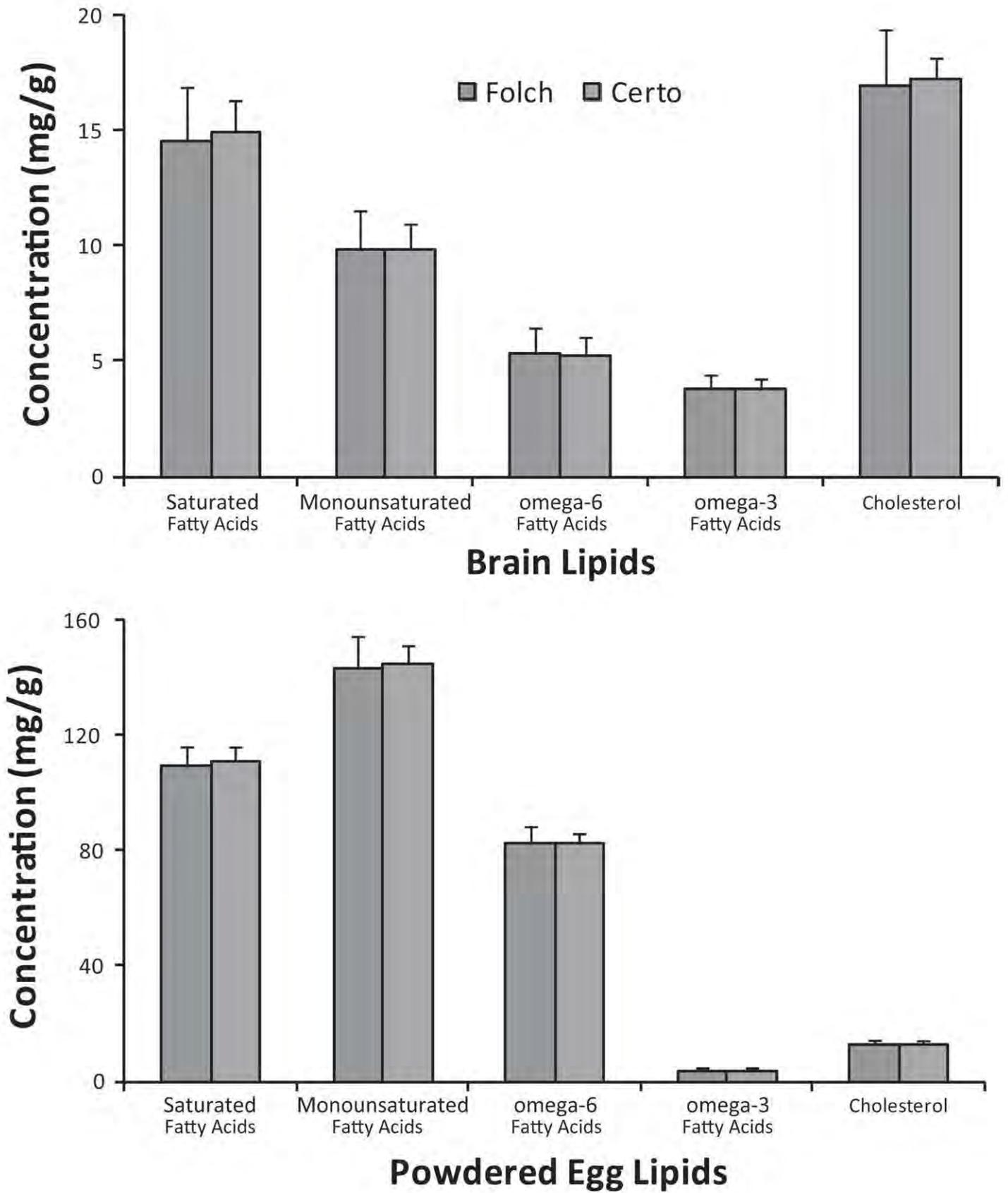


FIG. 2. Lipid determinations by Folch method vs. Certo kit in brain and egg.

Special report on aqueous oil extraction (cont. from page 560)

leading avocado growers, began commercial avocado oil production in a new 10,000 square meter processing and packaging plant in Jalisco, Mexico, in the second quarter of 2013. Mevi is operating the plant as a joint venture with AvoHealth, and according to Andrew Logan, they are using “cylindrical malaxers to eliminate dead spaces and reduce the opportunity for oxygen damage.” The temperature in the malaxers is kept at about 40–50°C, which does not adversely affect oil quality. Logan commented, “Cylindrical malaxers . . . maximize the heat transfer area, which in turn reduces residence time.”

OTHER APPLICATIONS OF AQUEOUS EXTRACTION

Keshavan Niranjana, a professor of food bioprocessing with the University of Reading (UK), points out the advantage that aqueous oil extraction has in processing fruits compared with seeds and pulses. “They don’t have significant levels of proteins

which would otherwise stabilize any emulsion formed, making separation of oil from the aqueous phase difficult,” said Niranjana. “This is a major issue with soybean.”

To counteract oil/water emulsification and the negative impacts on oil yield and quality, industrial processes for the extraction of edible oil from oilseeds generally involve a solvent extraction step, often using hexane. More importantly, this type of extraction is also often cheaper than using mechanical means. High oil yields have been reported for such processes, some in excess of 95% (Rosenthal *et al.*, 1996). However, the organic solvents used, especially hexane, contribute to emissions of volatile organic compounds.

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Olive, palm, and avocado

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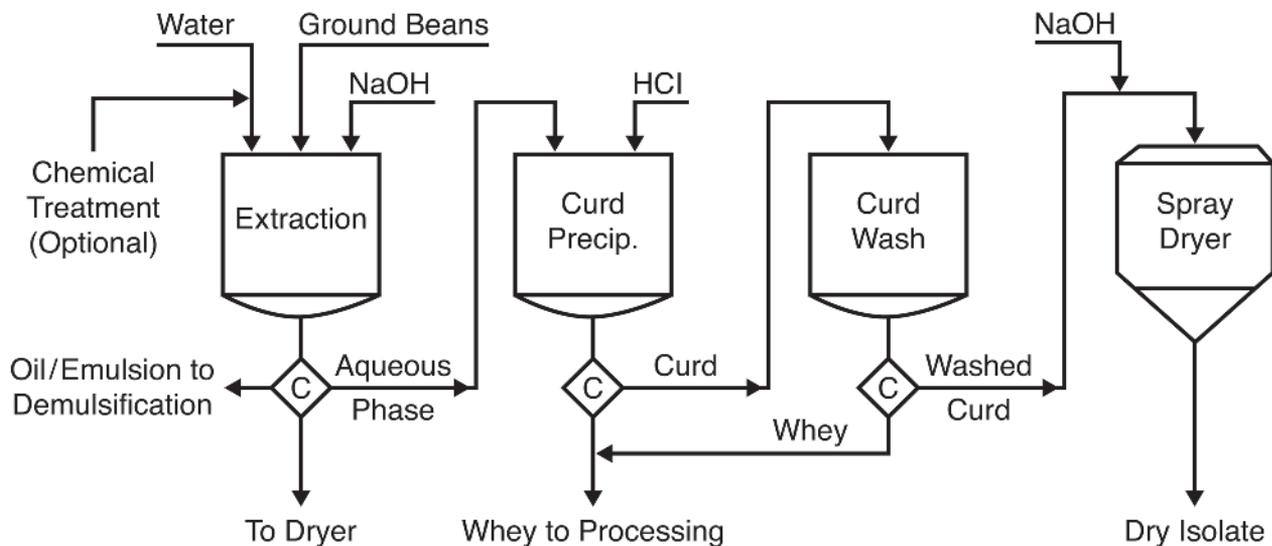


FIG. 1. Original concept for aqueous extraction of soybeans (Lawhon, J.T., K.C. Rhee, and E.W. Lusas, *J. Am. Oil Chem.* 58:377–384, 1981).

Advances in enzyme-assisted aqueous extraction of soybeans

Lawrence A. Johnson

Oil extraction from soybeans evolved from ancient batch pressing to continuous screw presses in the early 1900s and then to direct solvent extraction in the 1930s using primarily a petroleum distillate rich in *n*-hexane. Continuing concerns over worker safety, environmental pollution due to hexane emissions, and the safety of consuming hexane-extracted oils and proteins are driving the development of alternative soybean processing technologies. Industrial hexane is flammable, and occasional fires and explosions have caused considerable damage and loss of lives.

One such alternative is aqueous extraction (Fig. 1) in which water is used, not as a solvent to dissolve oil, but as an

extraction medium from which oil and protein are washed from the material to be separated and recovered. When soybeans are ground in water, a small amount of free oil and a larger amount of oil-rich cream (oil-in-water emulsion) float. The bulk of the underlying solution is rich in protein and sugar, and retains a small amount of oil emulsified by the protein; it is referred to as soy skim. Underneath the soy skim, an insoluble fiber-rich fraction settles out. After centrifuging the slurry of ground soybeans, about 45% of the oil can be recovered as a stable cream emulsion, which is difficult to break owing to large proportions of soluble protein and phospholipids that are potent emulsifiers. The skim contains approximately 19% of the seed oil, most of the sugars, and 92% of the protein. While aqueous extraction has been used to process olives and coconuts, the significantly lower yields of oil and dilute protein stream unacceptable for feed use have impeded commercial acceptance by the soybean industry.

Many recent advances deal with these challenges. Key advances include: (i) preparing soybeans by cracking, dehulling, flaking, and expanding (or extruding) to completely rupture cell walls and pseudomembranes around oil bodies, thereby eliminating barriers to extraction, improving oil extraction to 71%,

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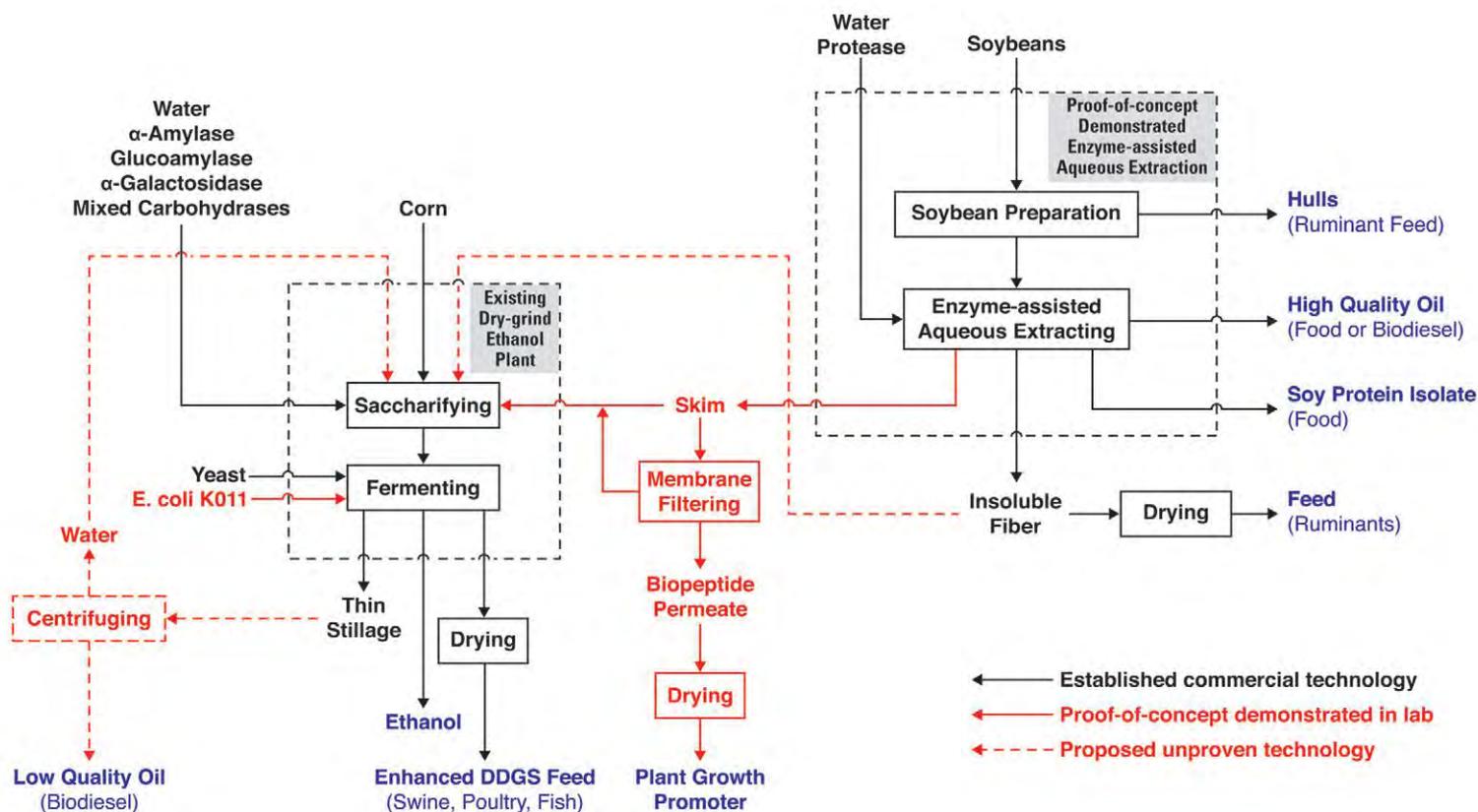


FIG. 2. An integrated corn and soybean biorefinery using enzyme-assisted aqueous processing.

and increasing oil droplet size to reduce emulsion stability; (ii) using a protease enzyme to enhance oil extraction to 96% (oil recovery is ~82% because some oil remains emulsified with the skim fraction); (iii) employing two stages of extraction with enzymes to improve oil extraction to 99% and protein extraction to 96% while reducing water use by 40%; (iv) using protease in the second or both stages to produce protein products with different extents of hydrolysis thereby enabling recovery of two types of protein ingredients with different functionalities; (v) breaking the cream emulsion by using the same protease as used for extraction while achieving 92% demulsification efficiency; (vi) recycling enzyme used for cream demulsification into one or more extraction stages to conserve enzyme; (vii) recovering protein by isoelectric precipitation when using enzyme in the second stage only or by membrane filtration when using enzyme in both stages; and (viii) using the soy skim to slurry with corn for fermentation in a soybean-corn biorefinery. The process employing these “best practices” can be viewed in this issue’s supplement (digital and mobile editions only). When these advances are integrated, the process is known as the “enzyme-assisted aqueous extraction process (EAEP)”; and when using these practices 98% oil extraction, 79% oil recovery, and 97% protein recovery are possible.

Rupturing all cells and pseudomembranes around oil bodies is key to successful extraction. Flaking and extruding

(or expanding) are much more effective in rupturing cell walls than most other alternatives although some have reported successfully using cellulases for that purpose. Using cellulases to disrupt cell walls makes sense when recovering oil as intact oil bodies. Proteases are critical to breaking down the hydrophobic protein oleosin, which along with phospholipids comprise the oil body pseudomembrane, to produce free oil.

The last remaining challenge is to capture maximum value of the skim, which contains about 19% of the original oil in the bean, soluble and partially hydrolyzed protein, and sugars, primarily oligosaccharides that are poorly digestible. The only recognized use of the skim is to recover protein isolates for use as food ingredients. Recently we demonstrated that EAEP can be integrated into a dry-grind corn ethanol plant (Fig. 2) to produce enhanced feed (potentially suitable for poultry, swine, and fish), speed fermentation, increase fuel ethanol production, and produce plant growth promoters. Also, such a process allows a second opportunity to remove oil and increase oil yield in dry-grind ethanol production.

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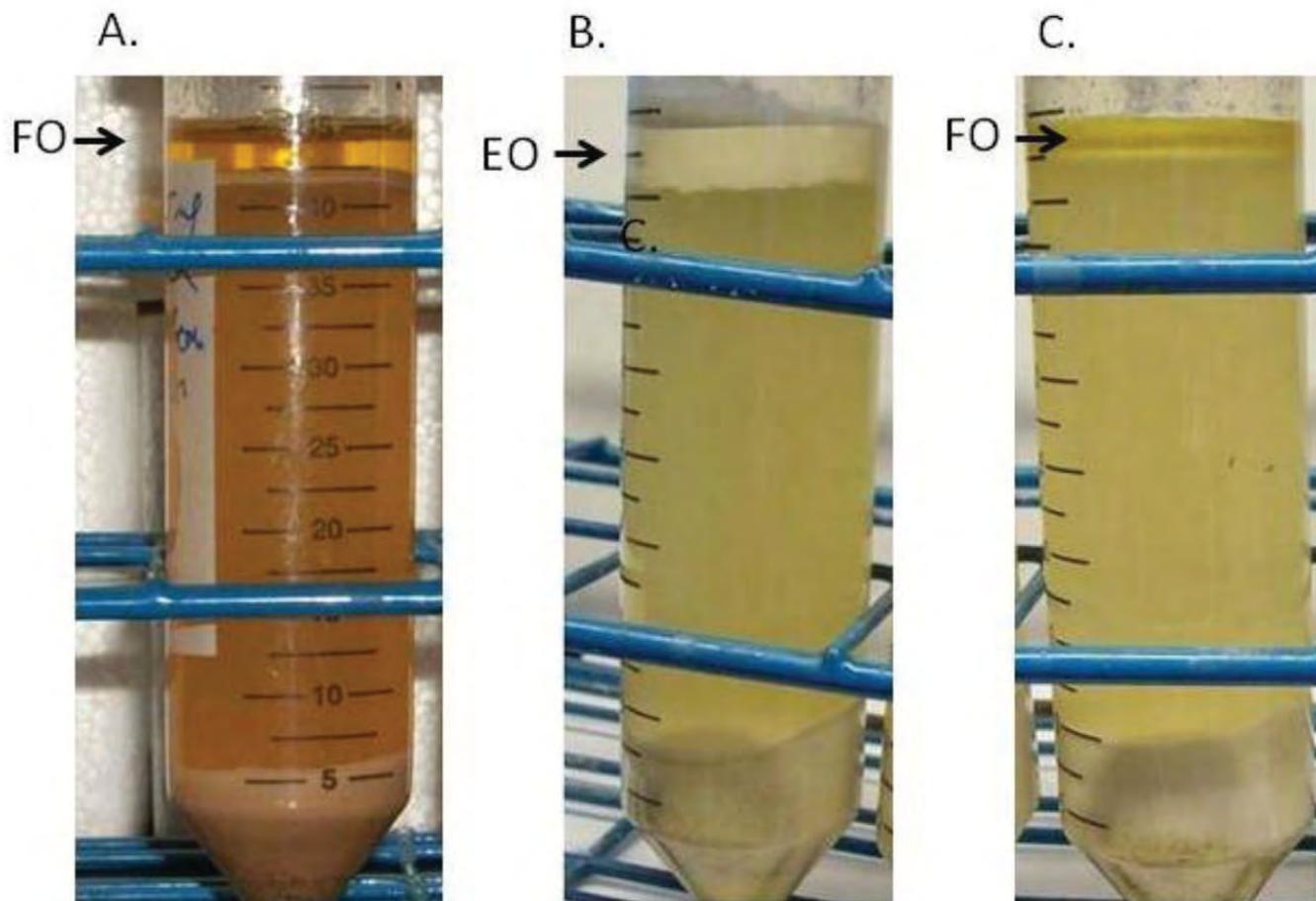


FIG. 1. Aqueous enzymatic extraction of corn oil from wet milled corn germ and dry milled corn germ. (A) Extraction of oil from wet milled corn germ using cellulase, which, after centrifugation results in a floating layer of free oil (FO; described in Moreau et al., 2004). (B) Extraction of oil from dry milled corn germ using cellulase, which, after centrifugation results in a top layer of emulsified oil (EO; described in Moreau et al., 2009). (C) Extraction of oil from dry milled corn germ using cellulase and alkaline protease, which, after centrifugation results in an top layer of free oil (FO; described in Moreau et al., 2009). Photos from R. Moreau.

The development of a green aqueous enzymatic process to extract corn oil from corn germ

**Robert A. Moreau, David B. Johnston,
and Kevin B. Hicks**

Approximately 2.4 million metric tons (MT) of commercial corn oil were produced worldwide in 2012, compared to 2012 world production of palm oil (53.3 MT) and soybean oil (43.1 MT), according to the Foreign Agricul-

ture Service of the US Department of Agriculture (USDA). Most commercial corn oil (~90%) is produced from corn germ that is expeller pressed and/or hexane extracted from wet milled corn germ, a by-product of the corn wet mill industry (Moreau, 2011a). Because of

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TABLE 1 Aqueous enzymatic extraction of corn oil from various types of corn germ

Type of corn germ	% oil in germ (dry wt)	Cellulase (pH 4–5)	Protease (pH 8)	Yield of only emulsified oil	Yield of free oil (relative %)	Reference
Wet milled corn germ	40–50	Y	N	N	80–90	Moreau <i>et al.</i> , 2004
Dry milled corn germ	20–25	Y	N	Y	0	Moreau <i>et al.</i> , 2009
Dry milled corn germ	20–25	Y	Y	N	64–66	Moreau <i>et al.</i> , 2009
E-germ ^a	41.0	Y	Y	N	80–87	Moreau <i>et al.</i> , 2009

^aEnzymatic corn germ.

the health and safety issues associated with the use of hexane, efforts have been undertaken to develop “green” aqueous or aqueous enzymatic methods for the extraction of corn oil from corn germ.

Our laboratory began research in this area by developing a process to extract corn oil from wet milled corn germ (Table 1; Fig. 1A, page 595). We developed an aqueous enzymatic method that achieved free oil yields of 80–90% (Moreau *et al.*, 2004).

After completing our studies with wet milled corn germ, we shifted to extracting oil from dry milled corn germ, which contains 20–25% oil (Moreau *et al.*, 2009). Using cellulase (at pH 4–5) as the enzyme, at the same concentration and conditions that were successful for wet milled corn germ, we reported that

no free oil was released from dry milled corn germ but there was a large layer of emulsified oil after centrifugation (Fig. 1B, page 595). A free oil yield of about 60% was achieved after we pre-treated the dry milled corn germ by heating it in a microwave oven (25 g of germ, heated for 40 seconds at 1250 Watts) or heating it in a boiling water bath (6 g of germ and 40 ml water heated for 30 minutes). A free oil yield of about 65% was achieved by adding a second enzymatic step (treating 4 hours with cellulase followed by changing the pH to 8.0 and treating with alkaline protease for 2–12 hours) (Fig. 1C, page 595).

The higher oil yields that we achieved with wet milled corn germ may be attributable to the fact that wet milled corn germ contains higher levels of oil than dry milled corn germ (and perhaps less barriers to impede oil extraction) and may be due to the fact that most wet milled corn germ is heated in a commercial drier to remove most of the moisture. When dry milled corn germ is used as a feedstock for aqueous enzymatic oil extraction, cellulase alone is not sufficient to break the barriers and release the oil; and oil can be liberated either by heat pretreatment of dry milled corn germ or by adding a second enzymatic step with alkaline protease (Moreau *et al.*, 2009).

We also evaluated enzymatic corn germ, “E-germ” (Johnston *et al.*, 2009), as an experimental noncommercial corn germ feedstock and achieved oil yields of 80–87% when using the combination of cellulase and alkaline protease (Table 1).

Unlike some of the other oilseeds that have been used as a feedstock for aqueous and enzymatic oil extraction, an advantage of corn germ is that free oil can be released by an enzyme treatment step and then separated by centrifugation, without the need for a second demulsification and centrifugation step. By using our processes, wet milled corn germ and E-germ resulted in high oil yields (80–90%). Research in our laboratory is continuing with the goal of increasing the oil yields of dry milled corn germ from the current 60–65% to 90% (for more details see Moreau *et al.*, 2011b).

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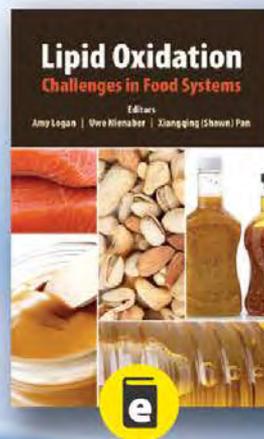


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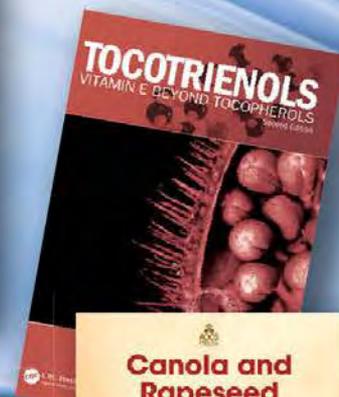
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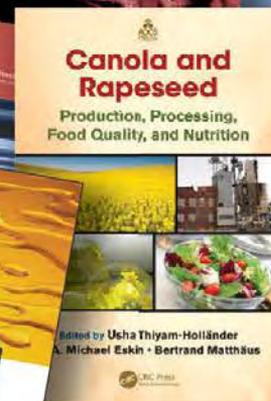
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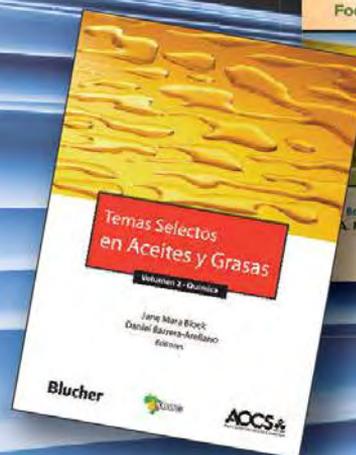
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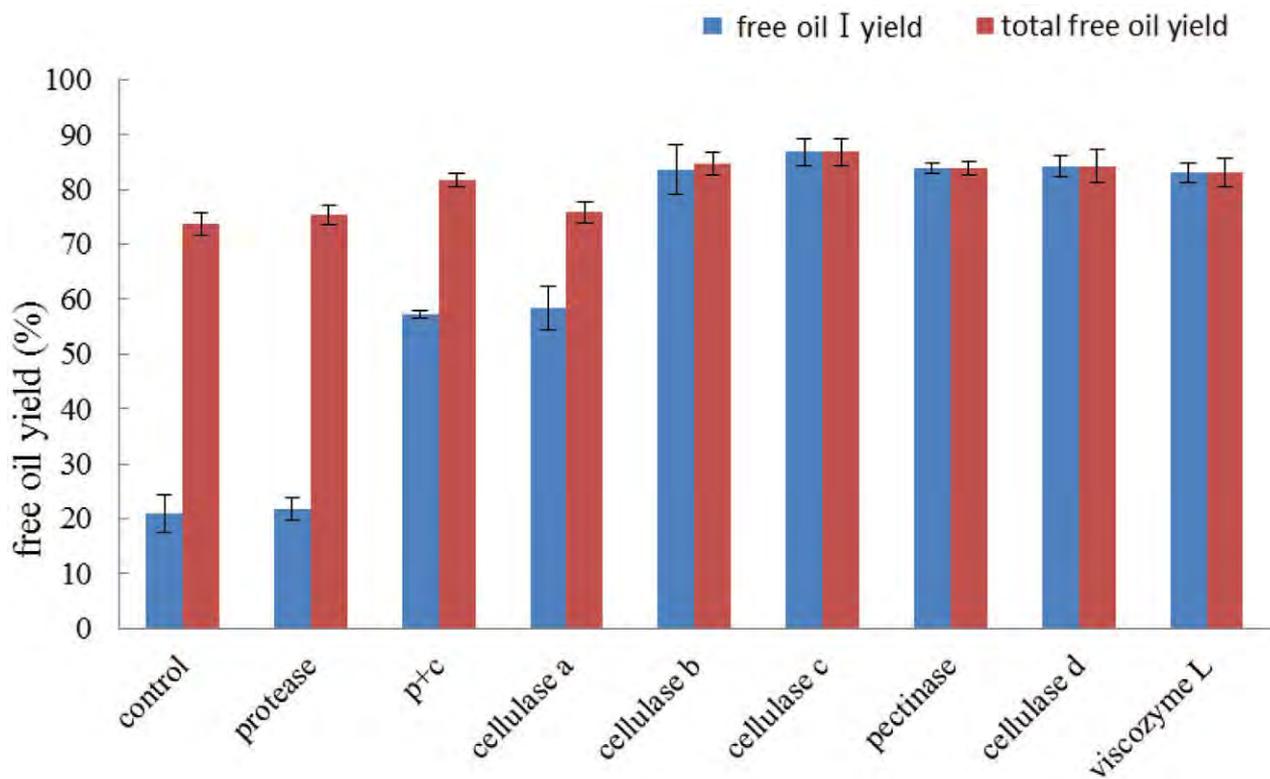


FIG. 1. Effect of different enzymes on free oil yields by aqueous enzymatic extraction process. p+c: a commercial enzyme containing pectinase and cellulase. A slurry made of 200 g *Camellia oleifera* kernels in 800 mL distilled water with 2 mL enzyme (1%, vol/wt) at each enzyme's optimal pH and temperature was incubated for 4 h with constant shaking. A control was run without adding enzyme. Total free oil yield was calculated as free oil a.yield obtained directly by centrifugation plus free oil yield b. obtained from the emulsion after demulsification.

Aqueous enzymatic extraction of camellia, or “tea,” oil

Wang Yingyao

Camellia oleifera is classified as a perennial woody plant and is one of the four commercially valuable edible oils derived from woody plants in the world. Its de-hulled seeds contain 45%–52% oil, 5%–10% theasaponin, about 10% protein, and 20% carbohydrate on a dry weight basis. The oil is obtained from the mature seeds within the kernel. Camellia oil's fatty acid composition is similar to olive oil, and it is even known around the world as “ori-

ental olive oil.” Theasaponin, which belongs to the saponin class of triterpene glycosides, has superior natural properties and is a very efficient emulsifier (Li *et al.*, 2011).

We adopted the aqueous enzymatic extraction (AEE) process to simultaneously obtain the oil and value-added by-product theasaponin from the *Camellia oleifera* kernel under mild physical-chemical conditions. During the development of the extraction process, we had to avoid the formation of stable oil-in-water emulsions, increase oil extraction yields, and reduce the enzyme cost as much as possible.

The typical cotyledon cells of the de-hulled seed are about 59 μm in diameter and 66 μm long, and the oil bodies

CONTINUED ON NEXT PAGE

Camellia INFORMATION

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range from 0.8 to 1.1 μm in diameter. Physical comminution with dry grinding was used to disrupt the cell walls and reduce the size of the resulting particles so that there would be more surface area available for enzyme reaction. After one and two grindings, the average diameter of the particle size was 468.9 μm and 40.92 μm , respectively. The corresponding free oil yield was 63.32 and 84.71%, respectively.

The effects that can be achieved by enzymatic hydrolysis are closely related to the composition of oilseed cells. The effect of various commercial enzymes (including protease, cellulase, and pectinase, and a multi-activity enzyme preparation) on a slurry of ground-up kernels in which the seed-to-water ratio was 1:4 was further studied. The results are shown in Figure 1, page 599. Cellulase b, cellulose c, pectinase, cellulose d, and viscozyme L yielded the highest oil extraction, and the free oil yield was close to the total free oil yield. This means that there was almost no emulsion formation when these enzymes were used. So, it is completely feasible to develop an effective AEE process in a *Camellia oleifera* kernels system without having to contend with stable emulsion formation.

We investigated and evaluated the variables (namely, enzyme loading, reaction time, temperature, and pH) on the objective

of maximizing total free oil yield and theasaponin recovery by response surface methodology. The optimal conditions for cellulase c were an enzyme concentration of 1%, an initial pH of 4.0, a temperature at 43°C, and an incubation time of 2.7 h with constant shaking. These optimal conditions yielded an 89% free oil yield and an 81% crude theasaponin recovery.

Preliminary *in vitro* studies showed that the crude theasaponin had inhibiting effects on *Aspergillus flavus* and *Escherichia coli* in 1.4 mg theasaponin/ml water. The composition of the crude theasaponins was in complete accord with the Chinese National Standard Feed Additive of Sacchariterpenin (a feed additive isolated from *Camellia oleifera*), which means that the recovered by-products could be used as a feed additive.

Some of the needs that trigger technological innovation in the oil extraction sector—such as cost savings, environmental and safety concerns, and nutrition issues—seem to be satisfied by successful development of aqueous enzyme-based processes.

Enzymatic extraction is likely to be a viable technology for extraction of some vegetable oils in the future, but further evaluations of the processes with respect to water recycling, free oil yield improvements, systematic process engineering, and economics should be conducted before considering scaleup.

Wang Yingyao is engaged in lipids processing research and now acting as the principal investigator of the lipids group in Academy of State Administration of Grain in China. He can be contacted at wyy@chinagrain.org.

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Current status of enzymatic aqueous extraction processing in China

Xiaonan Sui, Yang Li, Baokun Qi, and Lianzhou Jiang

In China, many studies have been conducted on the extraction of oil from a variety of seeds, such as soybeans, peanuts, sunflower, perilla, hemp, tea, and hazelnuts, using enzymatic aqueous extraction processing (EAEP) methods. Initially, the EAEP method was evaluated for extracting oil from traditional oil crops, such as soybean, peanut, and rapeseed.

The Heilongjiang province, located in the northeast part of China, is the major production area for soybeans for the country; and most of the studies involving the EAEP of soy oil were conducted by researchers in this province. The maximum oil yield for the EAEP of soybean oil was 93.1%

after demulsification of the cream (Li *et al.*, 2010). In southern China, most of the EAEP oil studies were conducted on rapeseed, since rapeseed oil is one of the main edible oils there. The highest rapeseed oil yield achieved was 73–76% when carbohydrase hydrolysis was used as a pretreatment method (Zhang *et al.*, 2007). EAEP has also been also conducted on nontraditional oil crops in China, such as perilla seeds (Li *et al.* 2013), microalgae (Liang *et al.*, 2012), and tea seeds (Zhang *et al.*, 2012).

In addition to studying EAEP of oil from traditional and nontraditional oil crops, researchers in China are now considering ways in which EAEP can be used to achieve a balanced fatty acid profile, since the concept that oils with balanced fatty acids composition promote good health has been widely recognized by nutritionists and medical scientists. The Chinese Nutrition Society's dietary reference intake (DRI) recommendation is a fatty acid ratio of 0.27:1:1 (saturated fatty acids/monounsaturated fatty acids/polyunsaturated fatty acids) (Chinese Nutrition Society, 2000)—an ideal that is impossible to achieve from

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a single variety of oilseed with EAEP methods because no single variety of oilseed has a completely balanced ratio of fatty acids. A balance can be achieved only by blending different types of oil. Yet, studies have been conducted to identify reliable extraction methods. A study carried out by Li *et al.* (2013) reported that oil with the ideal fatty acid composition (0.27:1:1) was successfully extracted from a mixture of seeds (soybean, peanut, linseed, and tea seed) using EAEP methods. However, no other study about extracting oil with balanced fatty acid composition from seeds has been published to date.

Despite the growing body of research on EAEP methods, the lack of pilot-scale studies limits their commercial applicability. Such studies are urgently needed to develop the widespread application of EAEP in China, as the edible oil industry is reluctant to adopt such methods without evidence that they will work on a commercial scale.

Xiaonan Sui, Yang Li, Baoku Qi, and Lianzhou Jiang are researchers in the College of Food Science, Northeast Agricultural University, National Research Center of Soybean Engineering and Technology, Harbin, P.R. China. Professor Lianzhou can be contacted at jlzname@163.com.

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Surfactant-assisted aqueous extraction process (SAEP)

Linh D. Do and David A. Sabatini

Surfactants are surface-active agents, which can lower the interfacial tension (IFT) between two immiscible phases. While IFT values between vegetable oils and water generally range from 15 to 50 mN/m, extended surfactants can lower the IFT to <0.1 mN/m, thereby liberating the oil from mechanically ruptured cell walls. These low IFT values are not possible using conventional surfactants, illustrating the

importance of extended surfactants. To sufficiently reduce the IFT values, the hydrophobicity-hydrophilicity between the surfactant solution and the oil must be balanced. The surfactant solution can be made more hydrophobic by increasing the alkyl chain of the surfactant molecule. However, due to the highly hydrophobic nature of triglycerides, the main component in vegetable oils, aqueous conventional surfactant solutions cannot achieve

ultralow IFT values with vegetable oils because the hydrophobic-hydrophilic balance was not achieved within the limiting solubility of the surfactants.

Extended surfactants have propylene oxide (PO) and/or ethylene oxide (EO) groups inserted between the alkyl tail and sulfate head group of the surfactant molecule (chemical structure shown in Fig. 1). The PO groups increase the surfactant interaction with the oil phase and the EO groups increase the surfactant interaction with the water phase, thereby increasing surface activity and reducing IFT. The surfactant interaction at the oil-water interface is often tuned by varying the electrolyte concentration (i.e., sodium chloride).

The electrolyte concentration at which the lowest IFT is obtained is defined as the optimum IFT* at the optimum salinity concentration (S^*). Table 1 shows the IFT* for 0.15 wt% C_{10} -18PO-2EONaSO₄ solution with various vegetable oils. Ultralow IFT values were reached within 10 minutes, which is highly desirable for design of vegetable oil extraction systems. In batch studies, extended-surfactant solutions extracted vegetable oils effectively (80–95% oil extraction efficiency) from a variety of oilseeds, including corn germ, peanuts, canola seeds and palm kernels, at ambient temperature within 30–45

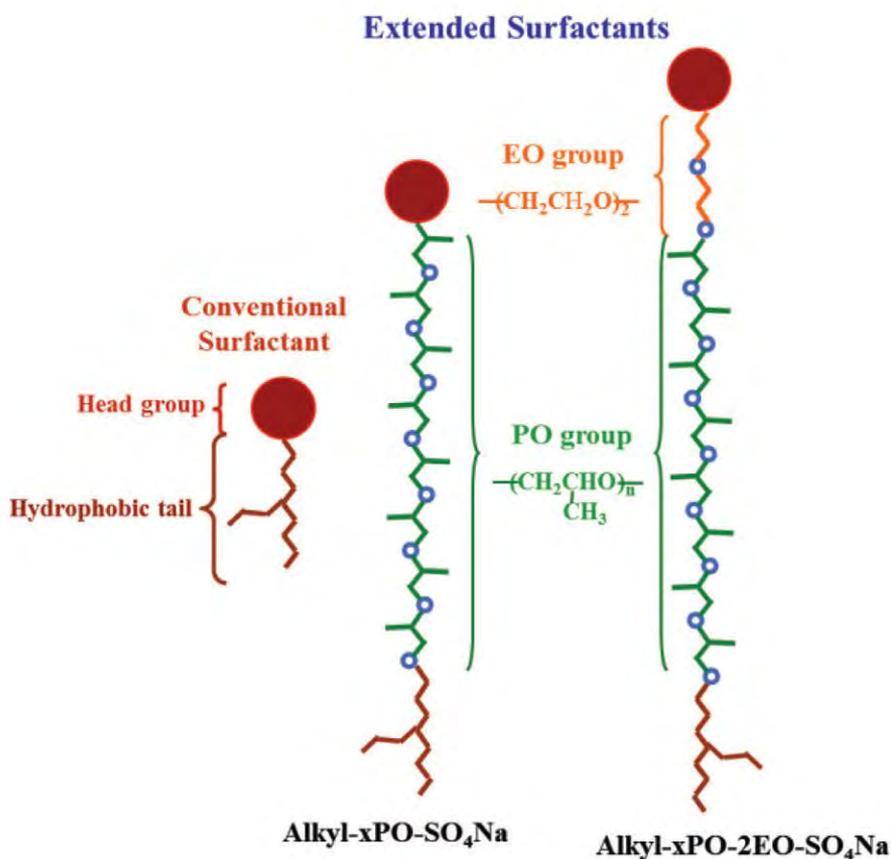


FIG. 1. Chemical structures of extended surfactants vs. conventional surfactants. Abbreviations: EO, ethylene oxide; PO, propylene oxide.

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minutes, which is highly competitive with other extraction processes. Further, the surfactant solution still produced ultralow IFT even after the initial extraction process, allowing the surfactant to be recycled with marginal makeup. Since it is an aqueous process, SAEP is especially attractive for extracting oil-bearing materials with high moisture content, such as algae.

SAEP produces a free oil phase, an emulsion phase, an aqueous surfactant phase, and a solid meal phase. When testing the SAEP on peanut and canola seeds in a semicontinuous pilot-scale study, the total oil extraction efficiency was similar to that of the batch studies (>90%), with the free oil phase being 85% and another 10% of the oil being in an emulsion phase. Even though the emulsion phase is much smaller than for other processes, recovery of oil in the emulsion phase could still be desirable.

Ongoing research is evaluating use of lower PO number (<4) extended surfactants, which are petroleum based, in mixture with other biorenewable surfactants to improve the environmental profile of this process while still maintaining ultralow IFT values with vegetable oils. Finally, further scale-up studies should be carried out to study the commercial feasibility of the SAEP technology.

TABLE 1. Optimum IFTs* between 0.15% aqueous solution of $C_{10-18}PO-2EONaSO_4$ with various vegetable oils at optimum S^* at 25°C^a

Oil	S^* (g/v%)	IFT* (mN/m)	EACN
Jojoba	3.0	5.18 E-4	10.2
Macadamia	3.5	3.45 E-5	11.7
Algae	5.5	6.33 E-3	16.1
Canola	6	5.70 E-3	16.9
Corn germ	6	2.00 E-4	16.9
Soybean	6.5	6.40 E-3	17.7
Peanut	7.5	8.40 E-3	19.0

^aAbbreviations: IFT, interfacial tension; PO, propylene oxide; EO, ethylene oxide; S, salinity concentration; *, optimum; v, volume; EACN, equivalent alkane carbon number (e.g., decane = 10, octadecane = 18).

Linh D. Do is a postdoctoral researcher in the School of Civil Engineering and Environmental Science and research associate at the Institute for Applied Surfactant Research at the University of Oklahoma (Norman, USA). She can be contacted at dieulinh@ou.edu. David A. Sabatini is a David Ross Boyd Professor and holds the Sun Oil Company Endowed Chair in the School of Civil Engineering and Environmental Science at the University of Oklahoma, where he is Director of the Water Technologies for Emerging Regions (WaTER) Center and Associate Director of the Institute for Applied Surfactant Research. Sabatini is also editor-in-chief for Journal of Contaminant Hydrology and an associate editor for AOC'S Journal of Surfactants and Detergents. He can be contacted at sabatini@ou.edu.

[FAST FACT: Cottonseed fake meat]

According to a recent *Time* magazine article, scientists turned animal tissue grown from stem cells into food (<http://tinyurl.com/stemcell-meat>), giving a possible insight into what we will eat in the future. Finding and perfecting meat alternatives is not a new science. Soy protein is a popular meat substitute in many vegetarian and vegan dishes. But did you know AOCS was one of the first to experiment with cottonseed protein as a meat alternative?

During the AOCS Spring Meeting of 1926, former AOCS President David Wesson served members "Croquettes à la Creole," a beefsteak substitute made with shortening, water, cade oil (juniper oil, now considered an essential oil), and cottonseed protein. Wesson was a

great innovator during the early years of the cottonseed industry, discovering methods to deodorize cottonseed oil and developing the first quality control process for the systematic examination of cottonseed products and the first physical auditing methods for cotton mill operators. Wesson continued to refine his cottonseed protein inventions until his death in 1934.

Cottonseed protein is still explored as a possible meat substitute (*International Journal of Food Sciences and Nutrition*; doi: 10.1080/09637480701288488). However, Wesson's recipe, which received a "thumbs down" from attendees, has not been served at another AOCS meeting since.

Patents (cont. from page 577)

Process for the enrichment of methyl ricinoleate from castor oil methyl esters by liquid-liquid extraction

Rao, K.V.S.A., *et al.*, Council of Scientific & Industrial Research, US8383847, February 26, 2013

The present invention provides a process for the enrichment of methyl ricinoleate from castor oil methyl esters by liquid-liquid extraction (LLE) in presence of refined vegetable oils using an aqueous polar solvent. The invention provides an extraction of methyl ricinoleate by a nondestructive extraction method with good yields (75–90%) and purity (95–99%). The method consists of mixing castor oil methyl esters in a refined normal vegetable oil (feed) in a suitable proportion and selective extraction of methyl ricinoleate in a nondestructive manner by LLE using a polar aqueous solvent, followed by desolventization and drying of the solute to get an enriched methyl ricinoleate fraction with good yields. The nonhydroxy fatty acid methyl esters of castor oil methyl esters are retained in the vegetable oil and can be used for the preparation of biodiesel or oleochemicals or reused in the process for the enrichment of methyl ricinoleate after removing the hydroxyl fatty acid either by distillation under reduced pressure or further by LLE using pure solvent.

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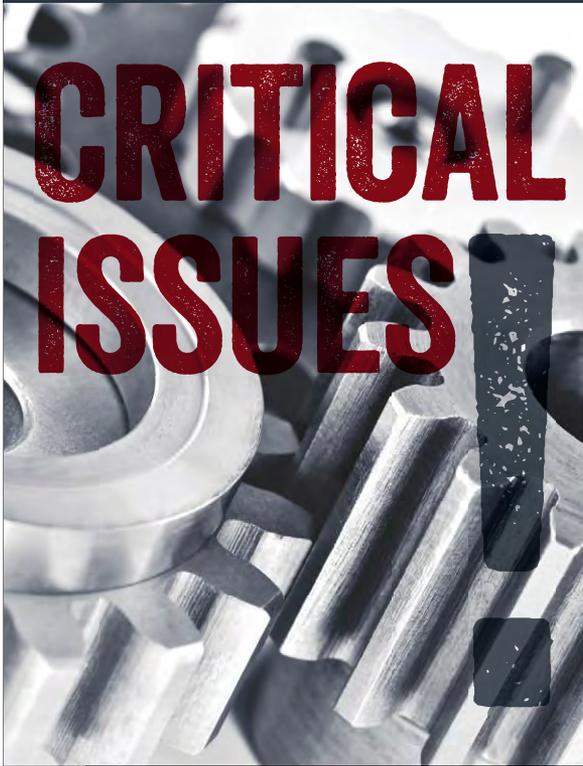
Buthe, A., *et al.*, Toyota Motor Engineering & Manufacturing North America, Inc., US8394618, March 12, 2013

A substrate or coating is provided that includes a lipase with enzymatic activity toward a component of a fingerprint. Also a process for facilitating the removal of fingerprints is provided wherein an inventive substrate or coating including a lipase is capable of enzymatically degrading of one or more components of the fingerprint to facilitate fingerprint removal from the substrate or said coating. Applying heat to the substrate or coating increases the rate of fingerprint removal.

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.



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October 2013

S1

BUILDING A POSITIVE

Michael Logli

“Safety culture” is defined as how safety is handled in the workplace and the overall attitude of employees toward safety. Developing a positive safety culture may seem to be an expensive endeavor. Despite the upfront cost, which fluctuates based on number of employees and the type of plant, a positive safety culture can save lives and save money over time.

While the specifics will vary, building a safety culture involves five steps. Safety managers must:

- Earn the support of upper management
- Get employees invested
- Identify hazards
- Train employees
- Track and measure progress

Become a “used car salesman” and sell a positive safety culture

Michael Coleman knows how to build a safety culture from the ground up. He began his safety career in 1999 at the Springdale, Arkansas, USA, headquarters of personal care product manufacturer Rockline Industries, leaving in 2009 to work at Tyson Foods.

The safety culture at Rockline didn’t flourish overnight. It took Coleman six to seven years of consistently achieving the minimum required for Occupational Safety and Health Administration (OSHA) compliance based on OSHA Stan-

SAFETY CULTURE

dard 1910 (29 CFR; <http://tinyurl.com/OSHA1910>)—which includes regulations regarding training, equipment use, and handling hazardous materials—before he had the data he needed to prove to management the benefits of funding a comprehensive safety program. Whereas some data came from Coleman’s own work at Rockline, he also used OSHA-gathered statistics (<http://tinyurl.com/OSHA-stats>) and case studies from other companies with larger plants. Coleman used this information to demonstrate the benefits of a positive safety culture; to provide a detailed plan to improve the safety culture, containing estimations of possible costs and savings; to outline the goals and benchmarks; and to describe the methods to achieve those goals. Coleman presented his plan to his managers and was able to get approval for a comprehensive safety plan focused on both behavior and compliance.

“You have to become a car salesman. You have to believe in your product,” Coleman said. “I taught [managers] how to sell the product, too.”

As part of any plan to improve safety culture, managers must sell safety with statistics and a no-tolerance approach. Chuck Coffey, corporate director of safety for Ag Processing Inc. (AGP; Omaha, Nebraska, USA) discussed this point in his presentation at the 2013 AOCS Annual Meeting & Expo in Montréal, Québec, Canada. Management must be fervent in its desire to achieve safety goals, setting an example for others, Coffey said. By simply wearing personal protective equipment (PPE) on the job, managers can lead by example. Enforcement must be universal as well, or employees will not believe in the safety culture.

“If you have a plant manager or superintendent who doesn’t care about safety, then it will be impossible to develop a [positive] safety culture,” Coffey said. “It’s like telling a kid

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SAFETY RESOURCES

For more general advice and information about safety culture and safety programs, go to:

- “Stop trying to create a safety culture”—<http://tinyurl.com/OHS-Safety>
- DuPont STOP program—www.training.dupont.com/dupont-stop
- OSHA safety blog—www.allaboutosha.com/safety-programs
- American Society of Safety Engineers—www.asse.org
- OSHA-sponsored training courses—www.osha.com/courses/general.html
- NIOSH (National Institute of Occupational Safety and Health) Safety and Prevention tips—www.cdc.gov/niosh/topics/safety.html

to wear their seat belt. If you don't wear yours, they won't wear theirs, either.”

With management on board, Coleman's next task was to get employees invested in developing a positive safety culture. The best way to do this is to give employees a stake in safety procedures and rules, letting their opinions matter in continued safety discussions. When employees care about safety, they improve the culture and are more willing to discuss safety issues.

To accomplish this, many plants, Coleman's included, establish employee-led safety committees with regular meetings. John Jefferson, senior vice president at Planters Cotton Oil Mill (Pine Bluff, Arkansas, USA), said his company's committees create safety policies, comment on processing issues, and bring ideas to management. They also develop goals and benchmarks and discuss previous incidents. Members rotate off committees at regular intervals, with terms varying from a month to six months to a year. Committee minutes should be recorded and be made easily accessible for employees.

“We try to make it a part of their job, just like quality and production,” Jefferson said. “The more you stress [safety] the better [the culture] gets.”

Encouraging safety with incentives

Using incentives in different ways can also keep employees involved. Coleman posted safety statistics in break rooms, and as his teams passed benchmarks and met goals, Coleman threw small parties to celebrate achievements and awarded company jackets. The jackets, when worn outside of work, helped spread the word of the company's safety culture, which can lead to other positive developments, Coleman said.

“I have had people apply because they heard about our safety record and they wanted to work somewhere that had [a positive safety culture],” Coleman said.

While incentive programs with more extravagant awards exist, sometimes less is more. Incentives should not overshadow safety. Employees and managers may not report incidents in order to meet safety goals and earn incentives, which harms the safety culture. Although Bunge North America, the North American operating arm of Bunge Ltd. in White Plains, New York, USA, does not have a corporatwide incentive program for its 125 plants and 4,000 employees, individual facilities do reward other activities, said Mike Snow, vice president, industrial management, at the company's North American headquarters in St. Louis, Missouri, USA.

“Some of our locations have incentives, but all incentives are focused on proactive activities such as conducting job hazard analyses before beginning a task rather than on injury rates or other reactive measures,” Snow said. “We prefer to use incentives when implementing new programs or procedures rather than providing incentives for ongoing safety activities.”

Find the hazards; record the hazards

It is important to be thorough and try to consider all possible hazards, Coleman said, no matter how unlikely. Employees on the floor may recognize issues that arise through production, but they may not predict uncommon hazards. Coleman chose to perform a full hazard analysis and critical control points inspection to identify plant hazards and inform employees of them. Similarly, Coffey schedules annual internal inspections of plant operations safety. To perform these checks, he sends employees to unfamiliar areas of the plant. Their fresh eyes give a unique perspective and identify hazards other employees may not notice. AGP also performs a job hazard analysis for each employee's position, Coffey said. The hazard analysis outlines the hazards and dangers involved in each job for the purpose of making employees more careful.

With the hazards identified, safety training can now be developed to address the specific challenges that facility will face. Effective safety training builds a strong foundation for a positive safety culture. New employees receive training from the moment they are hired, while older employees are retrained each year to reinforce safety ideals, Coleman said. Training can involve online work, classroom-style learning, books, testing, videos, workshops, and other methods. Coleman also referred to DuPont (Wilmington, Delaware, USA) and its Safety Training Observation Program (STOP). This behavior-based training program (<http://tinyurl.com/dupont-stop>) is a for-profit branch of the company's safety division, and Coleman said he referred to STOP training materials for advice on improving the safety culture. OSHA does require that some training programs—such as bloodborne pathogens training, lockout/tagout training, and PPE training—take place annually (<http://www.osha.gov/Publications/osha2254.pdf>), but individual programs should be focused on a particular company's needs and goals.

All of the training and support will mean nothing, though, if the company does not keep records. OSHA requires com-

panies to keep records of safety accidents; these records can be used to demonstrate growth and track progress. In addition, Coffey said it is important to look at near-miss incidents, or close calls where no employees are actually injured. Such occurrences should also be recorded in order to analyze the factors that led to the event. By analyzing these factors, companies can engage in proactive behavior by installing engineering controls or performing specific safety training. The prompt, preemptive response boosts the safety culture.

According to Coffey, a shorter employee at an AGP plant several years ago attempted to reach a valve on an elevated platform 20 feet off the ground. To reach the valve, which required two hands to operate, the employee stood on the guardrail, and he was later disciplined for his unsafe behavior. But after submitting a near-miss report and investigating the incident, Coffey and his staff recommended extending the platform, making it possible for all employees to reach the valve safely. The report prevented future accidents and led to safer behavior.

Getting employees to report near misses and high-potential exposures, even anonymously, can be challenging in a plant with a poor safety culture. When a safety program becomes more established, employees will report incidents and exposures, and anonymity won't be a factor, according to Bunge's Mike Snow.

"Employees can make reports anonymously, but because we've created a culture where safety should be talked about, most choose to include their names," Snow said.

Assessing return on investment

Over time, incorporating these guidelines into a safety program can have a profound effect on the frequency of work incidents and the status of a safety culture. When Coleman started at Rockline in 1999, the company's official OSHA

incident rate—a metric that divides incidents by man-hours worked to create a universal statistic for state and national comparisons—was at 9.4%. When Coleman left the company in 2009, the incident rate had dropped to 0.9%. By comparison, the 2010 average for manufacturing industries was 4.4%.

During that same 10-year period, costs from work-related injuries at Rockline dropped 60–70%. Coleman estimated the safety program itself cost \$120,000–150,000 per year to promote. In addition, improved productivity led to attrition, reducing the Rockline workforce from 650 to 500, he added.

The costs of developing and maintaining a safety program may seem great at first, but the money saved in workers' compensation claims and other costs is greater, Coleman stressed. According to the US-based National Safety Council (NSC), the average cost in 2011 of fatal or nonfatal work injuries (including employers' uninsured costs) was \$48.3 million per incident. This includes lost manhours and the money necessary to cover health care costs. In comparison, Jefferson estimates that Planters spends \$50,000 per year on its safety program. Snow reports that Bunge, with an average OSHA incident rate of around 2%, prefers not to think of its safety program in terms of its cost, viewing it instead as an investment in the company's employees.

For those wanting to improve a safety culture or develop a safety program, the most important point to remember is that doing so is a long, continual process requiring years of effort and compliance. Behavior can be difficult to change, but the results are worth it.

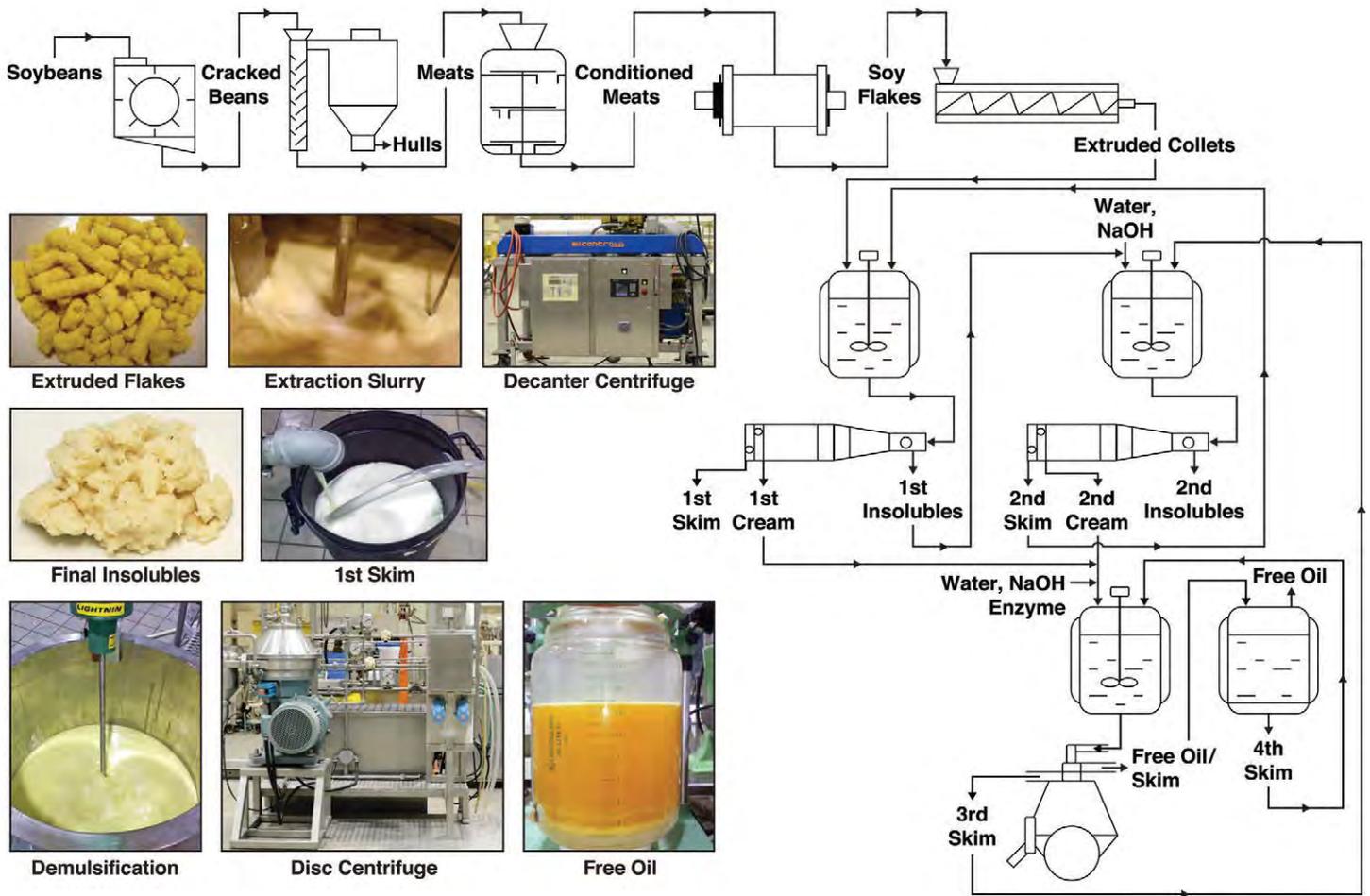
"Fixing a safety culture is like curing a cold. We didn't get sick overnight, and we won't get well overnight," Coleman said. "There is no panacea for a poor safety culture."

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Best practices in enzyme-assisted aqueous extraction of soybeans

figure from page 593





Polyfence Oil from Martech Research LLC prevents rancidity when added to other oils or fat-soluble systems. Photo courtesy of Lugo Nutrition, Inc.

Oils and fats innovation at IFT

Michael Logli

The 2013 Institute of Food Technologist's Annual Meeting & Food Expo in Chicago, Illinois, USA, (July 13–16) showcased products, techniques, and equipment to more than 23,500 registrants and 1,171 exhibitors. Oils and fats-related products at the expo included the following:



Solazyme (San Francisco, California, USA) and Roquette (Lestrem, France) ended their joint venture—Solazyme Roquette Nutritionals—on June 24, 2013, weeks before the IFT Annual Meeting & Food Expo. Both companies were out in full force marketing their respective microalgae powders.

Roquette's microalgae powder, called high lipid algal flour, contains 50% lipids plus a mixture of proteins, soluble and insoluble fibers, and micronutrients. Roquette also has a high-protein blend of the product. Roquette's processing plant, which can produce 5,000 metric tons (MT) per year, should be completed by the end of 2013, and the company plans to begin commercial production of its algae flour by the first quarter of 2014.

Solazyme's microalgae powder was marketed as a proprietary blend of lipids and other ingredients. Like Roquette, Solazyme has a high-protein version of its powder. It also plans to incorporate the non-powdered algae into an oil for the company's novel food oils line of products. The company can produce 100,000 MT per year of products through its joint ventures with Bunge, Archer Daniels Midland, and other companies.

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Bunge North America's Delta Dry powdered vegetable oil acts as a hydrogenated vegetable oil replacement. Photo courtesy of Bunge North America.

the company says. Alexandra Tonelli, representative for Martech's US sales contact, Lugo Nutrition, Inc. (Wilmington, Delaware, USA), said the company plans to market the product as an anti-rancidity agent alternative to rosemary oil or tertiary butylhydroquinone.



AAK, formerly AarhusKarlshamn (Edison, New Jersey, USA) highlighted a new low-saturate coating fat for confectionary applications. The product is a blend of palm kernel oil, palm oil, and canola oil that is *trans*-fat free and contains 60% saturated fat. Cocoa butter, by comparison, is 66% saturated fat. The coating system is suitable for enrobing, said Jessica Blackford, AAK research scientist and AOCS member.



Bunge North America (St. Louis, Missouri, USA) featured several products by using the Mobile Oil Experts (M.O.E.) food truck to cook them into food. One of these products—Delta Dry powdered vegetable oil—is a structured lipid made from a blend of medium-chain triglycerides and canola oil, to which whey or soy protein isolate and hydrocolloids have been added. The product stabilizes sports drinks and enhances protein content, acting as a more healthful replacement for hydrogenated and partially hydrogenated vegetable oils.



Bruce Livingood, technical director of Heartland Flax (Valley City, North Dakota, USA), said customers were interested in using flaxseed oil in culinary applications but disliked its "bitter aftertaste." In response, Livingood developed a proprietary filtration process used before and after microfiltration in the cold press process to remove the oil's cyclic peptides that cause the aftertaste. This "anti-bitter" flaxseed oil has the same shelf life and fatty acid profile as other flaxseed oils, he said.



Glanbia Nutritionals (Carlsbad, California, USA) highlighted an egg replacement system called OptiSol 3000. The product, one of four to win the 2013 IFT Food Expo Innovation Award, is a combination of milled flax seed and whey protein concentrate that replaces eggs in a 1:1 ratio for baking applications. According to Glanbia representatives at their Expo booth, results from the company's self-conducted sensory trials show little to no change in taste and texture compared to foods made with real eggs.



Ankom Technology (Macedon, New York, USA), a developer of analysis equipment, displayed two machines that test materials according to AOCS methods. The XT 15 Extractor adheres to AOCS Method Am 5-04 Rapid Determination of Oil/Fat Utilizing High-Temperature Solvent Extraction to perform solvent extractions and measure fat content. The ANKOM 2000 is a fiber analyzer that follows AOCS Method Ba 6a-05 Crude Fiber Analysis in Feeds by Filter Bag Technique in determining fiber content. The ANKOM 2000 also extracts acid detergent and neutral detergent fibers. Both machines come in smaller versions: the XT 10 and the ANKOM 200.



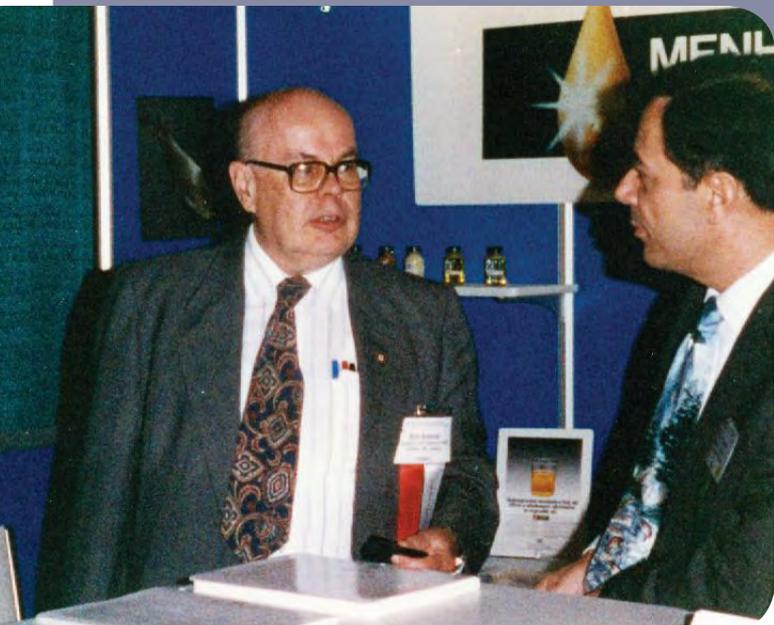
The Ankom XT 15 Extractor performs quality control experiments according to AOCS methods. Photo courtesy of Ankom Technology.



Martech Research LLC (Bishopville, South Carolina, USA) promoted its Polyfence Oil, an oil with a proprietary blend of fruit extracts and antioxidants. When added to oils and fat-soluble systems in concentrations of 0.5% by weight of oil, Polyfence Oil can prevent rancidity and enhance flavor,

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Thoughts upon the passing of Robert G. Ackman



Anthony Bimbo is a consultant in marine oils, marine protein, and omega-3 oils in Kilmarnock, Virginia, USA. He may be contacted at apbimbo@verizon.net.

When a friend or family member passes away you go through a period of sadness, which is a normal part of grieving. You progress to thinking about all the good times and memories you have of the person, lingering over the delights.

These are some of the memories I have of Bob Ackman. The dates and chronology may not be exactly right but it is what I recall.

I first met Bob in the late 1960s when he was still with Fisheries and Oceans Canada (Fisheries Research Board of Canada) in Halifax, Nova Scotia. I was a chemist with one of the fishmeal companies in the USA and was tasked with the responsibility for operating our F&M Model 500 Gas Chromatograph. It was a large, bulky thing with a car battery as the power source and a packed column. The peaks of chromatograms were cut out and weighed to get the percentage of each fatty acid. In our case, since we had a tight budget and no copy machine, we measured the peaks to get the area of the triangle, added them together, and divided by the total.

My boss had met or heard Bob speak at a conference and suggested that I go up to Halifax and meet with him. At that time taking a foreign trip was quite an honor. So I vis-

ited with Bob, toured his lab and office, and was amazed to see he had an integrator that automatically measured the area peaks. No measuring and calculating areas of triangles, no adding columns of numbers. We did not have an adding machine in the lab (there were no calculators), and we used slide rules to do the calculations. Bob's office was another matter; it was cluttered, as most of our offices are today and mine still is. But he knew exactly where everything was and I am sure that if someone moved an item he knew it.

He invited me to dinner at his home and I made a terrible blunder when I passed on the vegetables. His wife Cathy felt that it was a bad influence on their daughters. But that was finally forgiven when they visited Williamsburg, Virginia, and my wife pointed Cathy to the Outlet Mall where she purchased quite a few pairs of shoes.

Our friendship and collaborations continued through the University tenure, the Canadian Institute of Fisheries Technology and finally Dalhousie University, emeritus status and well past his retirement. If I have one regret, it was that I didn't make that one more call to see how he was doing after he finally left the University and worked from his home.

Bob always had a grad student or assistant or colleague who happened to be doing the type of work that you needed and could provide the information or who could do the work. As we were developing the menhaden GRAS petition, we found some gaps in information that we needed filled—and he and his associates plugged that gap. His students and sabbatical visitors came from all over the world to work with him. He had a keen knowledge of analytical techniques and some have become standard methods today.

His students worked on the measurement of antioxidant residues in farmed salmon, the effects of feeds on the fatty acid composition of farmed catfish, and characterization of the wide range of *cis* and *trans* isomers in partially hydrogenated menhaden oil.

I was honored when he asked me to write a chapter for a book he was editing, *Biogenic Lipids, Fats and Oils* (2 volumes), published by CRC Press in 1989. The chapter (my first) was written with a manual typewriter and when it came time to make editorial corrections we literally re-typed a sentence or paragraph and then cut it out and pasted it over the original document, hence cut and paste. I'm not sure what Bob did. I know in later years when most of us moved into computers he was still writing things longhand and his writing was practically indecipherable. He never used email, although he did have an email account that was managed by the department

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secretary. He was an avid user of the copy and fax machines and there was always a cartoon tucked in with the papers. In fact, he was the only person who still sent faxes to me until he finally retired and left the University. I guess I can retire the machine now.

I felt deeply honored when he asked me to co-chair an AOCS Short Course on Modern Applications of Marine Oils held in Toronto, Canada in 1992.

He received the Earl P. McFee Award from the Atlantic Fisheries Technology Conference in 1973. Incidentally, two of his colleagues William Dyer and Graham Bligh also received the award in 1975 and 1983, respectively. They are the authors of the Bligh and Dyer Method for fat extraction, which is used universally today. He received many other awards over his long and illustrious career.

When the Food and Agriculture Organization of the UN (FAO) and the World Health Organization (WHO) called for an expert consultation on the role of dietary fats and oils in human nutrition in 1977, Bob was invited.

The photo published with his obituary [which also appears in this issue of *Inform*] is the way I will always remember him. Jovial, always smiling, full of energy, and always ready to help or answer a question no matter how silly it was. He came into the fats and oils industry in its early days, prior to any mention of omega-3 fatty acids, and when he retired he left a lasting imprint on the field. You do not find a published paper dealing with marine oils that does not have one or more of Bob's publications in the reference list.

Bob was certainly a giant in the fish oil and omega-3 arena and he will be missed by all his friends, family and colleagues.

Tony Bimbo
August 16, 2013

Former AOCS President James Daun retired from the Canadian Grain Commission and is now an independent consultant. He may be contacted at jdaun@mymts.net.

Bob liked to tell the story that his graduate supervisor had heard that a guy named A.T. James had invented this device that separated things in the gas phase and sent Bob over to England to have a look. Of course, the device was the first gas chromatograph. Bob said that he came back to his lab and went to work to build the world's second gas chromatograph. From there he went on to be at the forefront of the use of this technique in lipid analysis, particularly fatty acids.

I first met Bob through a package he sent me while I was an M.Sc. student at the University of Manitoba. I was studying methods for determining the amount of sulfur in rapeseed and canola oils. The package was delivered directly to our laboratory and went directly into a fume hood, as it had been damaged and was leaking a very evil-smelling oil. Apparently Bob had heard that there was a student looking

into the problem of sulfur in rapeseed oil and he had prepared some oils to which he had added dimethylsulfide, the source of the smell, to see if I could really do the analysis. I called him by phone to confirm that his package had arrived damaged but that I would do my best to tell him how much sulfur was in the oil.

When we finally met in person, perhaps at an AOCS meeting, we began a friendship that lasted many, many years.

Bob also liked to clip interesting articles and to send them to people through the post. A few days after they arrived, he would call to see what you thought of the articles. It sure kept me up to date on what was going on in the lipid area.

Jim Daun
August 17 and 20, 2013

Richard Addison is retired from the Bedford Institute of Oceanography. He may be contacted at rfaddison@shaw.ca.

I worked closely with Dr. Ackman between 1967 and 1971. I recall that he was a meticulous analyst: He always wanted at least three independent analyses of a sample before he would accept them. Usually, this involved running a fatty acid methyl ester sample on two columns of quite different polarity (and then trying to reconcile the results), and comparing the inferred fatty acid composition with a measured Wij iodine value (IV) to confirm the extent of unsaturation. After this, the sample would be hydrogenated catalytically, the saturated esters re-run for confirmation of the chain length distribution, and the volume of hydrogen consumed compared to the measured IV. Of course, none of these independent analyses gave absolutely comparable results, and one spent a lot of time trying to explain differences of 0.1% in fatty acid distribution. Then someone further complicated the process by inventing a method to separate classes of fatty acid methyl esters (saturates, monoenes, dienes, etc.) by complexing them with silver ions on thin layer chromatography. . . .

Ackman was not only meticulous; he was imaginative and versatile. He resolved stereoisomeric forms of some branched chain fatty acids by converting them to diastereomers by esterification with (–)-menthol, long before chiral stationary phases were developed. He was interested in "real" fisheries (and other) issues: In 1969, when industrial releases of white (elemental) phosphorus caused fish kills and related marine pollution problems in Newfoundland, he and I developed a method for its analysis that remains the standard method today.

After I left Ackman's lab, we continued to collaborate on such practical issues as the effects of fish oil processing on the removal or degradation of contaminants like DDT and PCBs. When "blackberry odor" affected the quality of some Newfoundland cod, he identified its cause as dimethyl sulfide arising from the breakdown of algal dimethyl-β-propiothetin.

Ackman also developed a quick test to identify ambergris, a secretion of the sperm whale's digestive system that, in the 1960s, was a valuable and expensive natural product used in the perfume industry. I remember beachcombers coming into the laboratory with several pounds of unattractive smelly material hoping to have made their fortune through an ambergris find, only to be disillusioned by Ackman's analysis done on the spot—

Initially, I think people found him a bit intimidating, not just because of his reputation, but because of his acerbic turn of phrase. But when you got to know him, you realized that he could laugh at himself, as well as his targets.

*Richard Addison
August 20, 2013*

Dave Matthews is general manager with Tasa Omega S.A., a subsidiary of TASA, in Peru. The company is gearing up to produce omega oil concentrates. He worked with Ackman for five years. He may be contacted at dmatthews@tasaomega.com.

I started working with Dr. Ackman in August 1982 as a laboratory analyst assistant, right after I completed undergraduate work at Nova Scotia Agricultural College. My first assignment was working on a two-year contract involving GLA [γ -linolenic acid] research for Efamol Ltd. The field was new at the time, and I put the pilot plant together for scale-up of GLA production. This was 15 years before production of omega-3 and omega-6 fatty acids really became commercial, and being in on the ground floor of a new industry with Dr. Ackman was a unique experience for me.

He was the first to report on using wiped film molecular evaporators on fish oil. These are now the industry standard for obtaining omega-3 fatty acids.

One thing about working for Dr. Ackman—"You got some really weird assignments." One of my earliest unusual ones was to collect dead snapping turtles. Conservation officers in southern Nova Scotia were collecting turtles killed by cars on the province's roads. Ackman wanted to do fatty acid analyses on fat depots in the animals, so I had to go out and retrieve the animals from the officers—the fresher, the better.

In another improbable assignment, I worked with Guillermo Napolitano, an Argentinian studying in Ackman's laboratory. His project included analyzing the fat depots of the amphipod *Corophium*, which were to be collected from the sands exposed at low tide in the Bay of Fundy. Dr. Ackman indicated that we would need to collect 2 kilograms of these animals. When we reached the Bay, the scope of the problem was evident—we had to contend with the tremendous tidal reach for which the Bay is famous, and we were collecting very small animals (<11 mm long). With the two of

us picking through the sand by hand and racing the rising tide, we collected about 100 g. We returned to the laboratory—and asked for mercy. Dr. Ackman reluctantly lowered the needed quantity to 1 kilogram—and subsequently we developed a sieving system to get the animals we needed.

Yet another assignment in 1984 involved retrieving porpoises that had become stranded on the seashore and died. The initial report said there were two dead dolphins (subsequently correctly identified as porpoises), each about 2–3 feet long. Although Ackman had no project involving dolphins/porpoises at the time, he told me to go get them "in case we get a project." He thought their brain tissue would be interesting to analyze for fatty acids or pollutants. I went to pick them up in my VW Rabbit, since the departmental truck wasn't available at the time. The animals turned out to be 8 and 9 feet long. The SPCA was on the scene too, and I was able to persuade them to use their truck to help me bring back the larger animal to the laboratory—we just couldn't store two. We managed to wrestle "Flipper" (as he came to be called) onto the elevator, which carried him up to the lab, and put the body in the walk-in freezer. A few days later, Ackman called me after a school tour had come through his laboratory, saying, "You just made a whole bunch of kids cry!" The children had seen the porpoise in the freezer and assumed that the workers in the laboratory were responsible for the animal's death. After that, I built a "coffin" for Flipper so he wouldn't be so obvious. Eventually the freezer died, and we transferred the body to yet another facility "in case we get a project." In 1987, though, I finally disposed of it.

My story of the porpoise reflects that Ackman was quite a saver—but his saving ways paid off many times. He would give me an assignment, and usually I could go to his storeroom and find just about everything I needed to assemble anything.

He also saved reprints and journals and books. In his office, the only wall not covered with shelves overstuffed with paper was the one with the door out to the hallway. His desk was covered at least 2 feet deep in paper—but remarkably, he knew where everything was. And because of that, he did not want anyone to disturb his office. Ackman's students didn't have to go the library to look for a paper. All they had to do was ask him, and he would go straight to it in his office.

The amount of paper in his office, though, meant that available writing space was limited to non-existent. Thus he was well known for composing papers by hand on his lap. Office staff would then transcribe his "horrible handwriting" for his further use. (He never did convert to writing on a computer with a word-processing program.)

He loved his family, and he loved tinkering and writing. "His work was his hobby."

*Transcribed from a telephone
conversation August 20, 2013
Dave Matthews, as told to Marguerite Torrey*

EXTRACTS & DISTILLATES

Solvent fractionation of rice bran oil to produce a spreadable rice bran product

Bakota¹, E.L., *et al.*, *Eur. J. Lipid Sci. Tech.* 115:847–857, 2013.

Rice bran oil is becoming increasingly popular as a functional ingredient, due to its high stability and health benefits. We detail here a new solvent fractionation procedure for the production of a spreadable product derived from rice bran oil. Four different experimental conditions for fractionation yielded four statistically distinct populations. The spreads show distinctive trends in their physical properties, thermal behavior, and rheology, based on incubation time. The rice bran oil-based spreads produced in this work consist primarily of rice bran oil but also contain rice bran wax, tocopherols, tocotrienols, and γ -oryzanol, as well as other sterols. Relative to rice bran oil, they are enriched in rice bran wax, saturated fatty acids, and sterols, all of which contribute to a more rigid rheological profile than that of rice bran oil. This solvent fractionation process may be used to develop rice bran oil-based spreads as functional ingredients or products that may replace nut butters.

Lipidomic fingerprint of almonds (*Prunus dulcis* L. cv Nonpareil) using TiO₂ nanoparticle based matrix solid-phase dispersion and MALDI-TOF/MS and its potential in geographical origin verification

Shen, Q., *et al.*, *J. Agric. Food Chem.* 61:7739–7748, 2013.

A matrix solid-phase dispersion (MSPD) procedure with titanium dioxide (TiO₂) nanoparticles (NP) as sorbent was developed for the selective extraction of phospholipids from almond samples, and matrix-assisted laser desorption ionization–time-of-flight mass spectrometry (MALDI-TOF/MS) was employed for analysis. A remarkable increase in the signals of phospholipid accompanied by a decrease in those of triacylglycerols and diacylglycerols was observed in the relevant mass spectra. The proposed method was applied to five batches of almonds originating from four geographical areas, whereas principal component analysis (PCA) was utilized to normalize the relative amounts of the identified phospholipid species. The results indicated that the lipidomic fingerprint of almonds was successfully established by the negative ion mode spectrum, and the ratio of m/z 833.6 to 835.6 as well as m/z 821.6 could be introduced as potential markers for the differentiation of the

tested almonds with different geographical origins. The whole method is of great promise for selective separation of phospholipids from nonphospholipids, especially the glycerides, and superior in fast screening and characterization of phospholipids in almond samples.

Omega-6 to omega-3 fatty acid ratio and higher-order cognitive functions in 7- to 9-y-olds: a cross-sectional study

Sheppard, K.W., and C.L. Cheatham, *Am. J. Clin. Nutr.* 98:659–667, 2013.

Background: Biochemical and behavioral evidence has suggested that the ratio of n-6 (omega-6) to n-3 (omega-3) could be an important predictor of executive function abilities in children. We determined the relation between the ratio of n-6 to n-3 and cognitive function in children. We hypothesized that children with lower ratios of n-6 to n-3 fatty acids would perform better on tests of planning and working memory. Seventy 7- to 9-y-old children completed three 24-h diet recalls and a subset of the Cambridge Neuropsychological Test Assessment Battery. Parents provided information on their demographics and children's diet histories. Mean n-3 and mean n-6 intakes were related to the mean time spent on each action taken in the planning problem. The ratio of n-6 to n-3 significantly predicted performance on the working memory and planning problems. There was a significant interaction between the ratio and fatty acid intake; when children had high ratios, a higher intake of n-3 fatty acids predicted a better performance on the planning task than when children had lower n-3 intakes. When children had low ratios, a lower intake of n-3 and lower intake of n-6 predicted better performance than when intakes were higher. The relation between cognitive abilities and the ratio of n-6 to n-3 may be mediated by an enzymatic affinity for n-3 fatty acids. The ratio of n-6 to n-3 should be considered an important factor in the study of fatty acids and cognitive development. This trial was registered at clinicaltrials.gov as NCT01823419.

Rapid quantification of yeast lipid using microwave-assisted total lipid extraction and HPLC-CAD

Khoomrung, S., *et al.*, *Anal. Chem.* 85:4912–4919, 2013.

We here present simple and rapid methods for fast screening of yeast lipids in *Saccharomyces cerevisiae*. First we introduced a microwave-assisted technique for fast lipid extraction that allows the extraction of lipids within 10 min. The new method enhances extraction rate by 27 times, while maintaining product yields comparable to conventional methods ($n = 14$, $P > 0.05$). The recovery ($n = 3$) from spiking of synthetic standards were $92 \pm 6\%$ for cholesterol, $95 \pm 4\%$ for triacylglycerol, and $92 \pm 4\%$ for free fatty acids. Additionally, the new extraction method combines cell disruption and extraction in one step; and the approach, therefore, not only greatly simplifies sample handling but also reduces analysis time and minimizes

sample loss during sample preparation. Second, we developed a chromatographic separation that allowed separation of neutral and polar lipids from the extracted samples within a single run. The separation was performed based on a three-gradient solvent system combined with hydrophilic interaction liquid chromatography-HPLC followed by detection using a charged aerosol detector. The method was shown to be highly reproducible in terms of retention time of the analytes (intraday, 0.002–0.034% RSD, $n = 10$, interday, 0.04–1.35% RSD, $n = 5$) and peak area (intraday, 0.63–6% RSD, $n = 10$, interday, 4–12% RSD, $n = 5$).

A database of chromatographic properties and mass spectra of fatty acid methyl esters from omega-3 products

Wasta, Z., and S.A. Mjøs, *J. Chromatogr. A* 1299: 94–102, 2013.

Fatty acids in products claimed to contain oils with the omega-3 fatty acids eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) were analyzed as fatty acid methyl esters by gas chromatography–mass spectrometry using electron impact ionization. To cover the variation in products on the market, the 20 products that were studied in detail were selected from a larger sample set by statistical methodology. The samples were analyzed on two different stationary phases (polyethylene glycol and cyanopropyl) and the fatty acid methyl esters were identified by methodology that combines the mass spectra and retention indices into a single score value. More than 100 fatty acids had a chromatographic area above 0.1% of the total, in at least one product. Retention indices are reported as equivalent chain lengths, and overlap patterns on the two columns are discussed. Both columns were found suitable for analysis of major and nutritionally important fatty acids, but the large number of minor compounds that may act as interferents will be problematic if low limits of quantification are required in analyses of similar sample types. A database of mass spectral libraries and equivalent chain lengths of the detected compounds has been compiled and is available online.

Determination of *trans* fat in edible oils: current official methods and overview of recent developments

Tyburczy, C., *et al.*, *Anal. Bioanal. Chem.* 405:5759–5772, 2013.

The adverse effects of dietary *trans* fat on biomarkers of chronic disease are well documented. Regulatory authorities in many countries have enacted legislation aimed at reducing *trans* fat content of their food supplies, either by requiring *trans* fat labeling on pre-packaged foods or by limiting the amount of *trans* fat in oils used for food production. Increased use by the food industry of oils with a low *trans* fat content necessitates reevaluation of official methods used by the food industry and regulatory agencies for the determination of total *trans* fat. Attenuated total reflection–Fourier-transform infrared

(ATR–FTIR) spectroscopy and gas chromatography with flame ionization detection (GC–FID) are two techniques used in official methods approved by method-endorsing organizations, for example AOAC International and the American Oil Chemists' Society. Here, we review current official ATR–FTIR and GC–FID methods for determination of *trans* fat, with a focus on factors affecting quantification of low levels of *trans* fat. We include new data on method performance that have only recently become available and provide an overview of notable recent developments in lipid analysis (e.g., infrared spectroscopy procedures, ionic-liquid GC columns, and multidimensional chromatographic techniques) that have the potential to substantially improve the accuracy, sensitivity, and/or speed of *trans* fat determination.

Omega-3 fatty acids and brain resistance to ageing and stress: body of evidence and possible mechanisms

Denis, I., *et al.*, *Ageing Res. Rev.* 12:579–594, 2013.

The increasing life expectancy in the populations of rich countries raises the pressing question of how the elderly can maintain their cognitive function. Cognitive decline is characterized by the loss of short-term memory due to a progressive impairment of the underlying brain cell processes. Age-related brain damage has many causes, some of which may be influenced by diet. An optimal diet may therefore be a practical way of delaying the onset of age-related cognitive decline. Nutritional investigations indicate that the omega-3 polyunsaturated fatty acid (PUFA) content of Western diets is too low to provide the brain with an optimal supply of docosahexaenoic acid (DHA), the main omega-3 PUFA in cell membranes. Insufficient brain DHA has been associated with memory impairment, emotional disturbances, and altered brain processes in rodents. Human studies suggest that an adequate dietary intake of omega-3 PUFA can slow the age-related cognitive decline and may also protect against the risk of senile dementia. However, despite the many studies in this domain, the beneficial impact of omega-3 PUFA on brain function has only recently been linked to specific mechanisms. This review examines the hypothesis that an optimal brain DHA status, conferred by an adequate omega-3 PUFA intake, limits age-related brain damage by optimizing endogenous brain repair mechanisms. Our analysis of the abundant literature indicates that an adequate amount of DHA in the brain may limit the impact of stress, an important age-aggravating factor, and influences the neuronal and astroglial functions that govern and protect synaptic transmission. This transmission, particularly glutamatergic neurotransmission in the hippocampus, underlies memory formation. The brain DHA status also influences neurogenesis, nested in the hippocampus, which helps maintain cognitive function throughout life. Although there are still gaps in our knowledge of the way omega-3 PUFA act, the mechanistic studies reviewed here indicate that omega-3 PUFA may be a promising tool for preventing age-related brain deterioration.

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Engineering fatty acid biosynthesis in microalgae for sustainable biodiesel

Blatti, J.L., *et al.*, *Curr. Opin. Chem. Biol.* 17:496–505, 2013.

Microalgae are a promising feedstock for biodiesel and other liquid fuels due to their fast growth rate, high lipid yields, and ability to grow in a broad range of environments. However, many microalgae achieve maximal lipid yields only under stress conditions hindering growth and providing compositions not ideal for biofuel applications. Metabolic engineering of algal fatty acid biosynthesis promises to create strains capable of economically producing fungible and sustainable biofuels. The algal fatty acid biosynthetic pathway has been deduced by homology to bacterial and plant systems, and much of our understanding is gleaned from basic studies in these systems. However, successful engineering of lipid metabolism in algae will necessitate a thorough characterization of the algal fatty acid synthase including protein–protein interactions and regulation. This review describes recent efforts to engineer fatty acid biosynthesis toward optimizing microalgae as a biodiesel feedstock.

High levels of anti-inflammatory and pro-resolving lipid mediators lipoxins and resolvins and declining docosahexaenoic acid levels in human milk during the first month of lactation

Weiss, G.A., *et al.*, *Lipids Health Dis.* 12: 89, 2013.

The fatty acid mixture of human milk is ideal for the newborn, but little is known about its composition in the first few weeks of lactation. Of special interest are the levels of long-chain polyunsaturated fatty acids (LCPUFA), since these are essential for the newborn's development. Additionally, the LCPUFA arachidonic acid (AA), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA) are precursors for lipid mediators that regulate inflammation. Methods: We determined the

composition of 94 human milk samples from 30 mothers over the first month of lactation for fatty acids using gas chromatography–mass spectrometry and quantified lipid mediators using high-performance liquid chromatography–tandem MS. Results: Over the four-week period, DHA levels decreased, while levels of γ C18:3 and α C18:3 steadily increased. Intriguingly, we found high concentrations of lipid mediators and their hydroxy fatty acid precursors in human milk, including pro-inflammatory leukotriene B4 (LTB4) and anti-inflammatory and pro-resolving lipoxin A4 (LXA4), resolvin D1 (RvD1), and resolvin E1 (RvE1). Lipid mediator levels were stable with the exception of two direct precursors. Conclusions: Elevated levels of DHA right after birth might represent higher requirements of the newborn, and the high content of anti-inflammatory and pro-resolving lipid mediators and their precursors may indicate their role in neonatal immunity and may be one of the reasons for the advantage of human milk over infant formula.

Lipidomic profiling of influenza infection identifies mediators that induce and resolve inflammation

Tam, V.C., *et al.*, *Cell* 154:213–227, 2013.

Bioactive lipid mediators play a crucial role in the induction and resolution of inflammation. To elucidate their involvement during influenza infection, liquid chromatography/mass spectrometry lipidomic profiling of 141 lipid species was performed on a mouse influenza model using two viruses of significantly different pathogenicity. Infection by the low-pathogenicity strain X31/H3N2 induced a proinflammatory response followed by a distinct anti-inflammatory response, infection by the high-pathogenicity strain PR8/H1N1 resulted in overlapping pro- and anti-inflammatory states. Integration of the large-scale lipid measurements with targeted gene expression data demonstrated that 5-lipoxygenase metabolites correlated with the pathogenic phase of the infection, whereas 12/15-lipoxygenase metabolites were associated with the resolution phase. Hydroxylated linoleic acid, specifically, the ratio of 13- to 9-hydroxyoctadecadienoic acid, was identified as a potential biomarker for immune status during an active infection. Importantly, some of the findings from the animal model were recapitulated in studies of human nasopharyngeal lavages obtained during the 2009–2011 influenza seasons. ■