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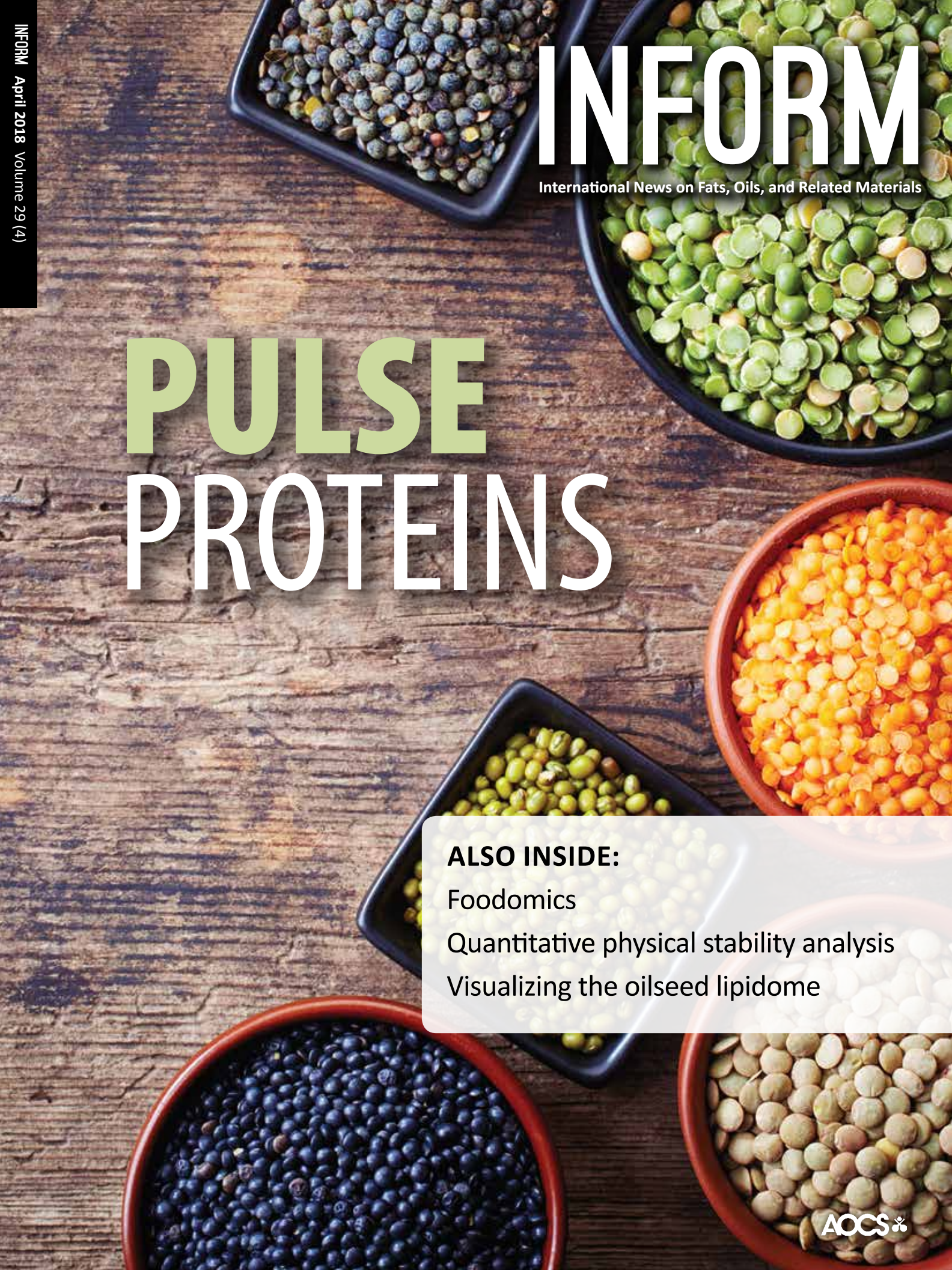

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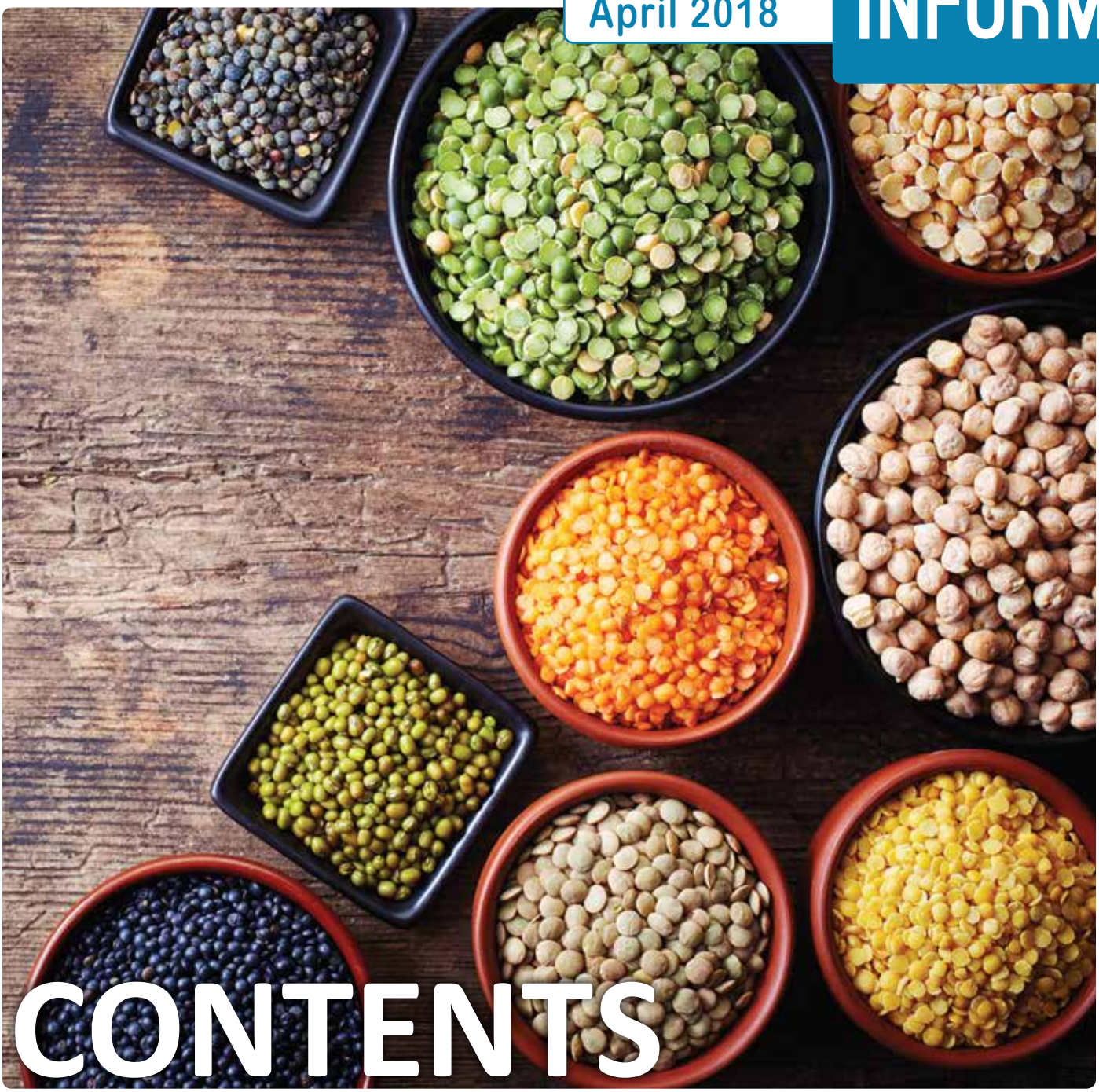

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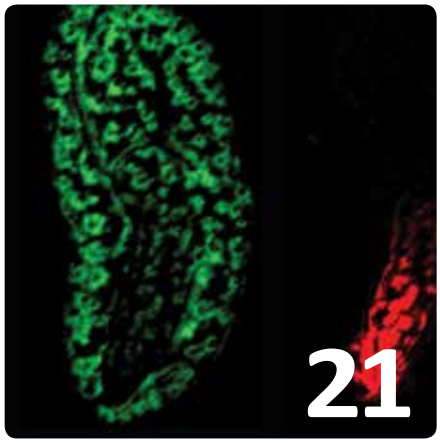

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Pulses rising

Laura Cassiday

Pulses such as chickpeas, lentils, peas, and beans are culinary staples in many parts of the world. Stews, hummus, falafel, chilies, and curries all feature the flavorful and nutritious dried legumes. However, these days pulses are showing up in unexpected places—in pastas, breads, snack foods, beverages, and meat and dairy substitutes. In addition to being rich in protein and fiber, pulses are widely considered more sustainable than animal-based proteins. To heighten awareness of the benefits of pulses, the United Nations Food and Agriculture Organization (FAO) declared 2016 the "International Year of Pulses."

- **Pulses are legumes harvested solely for their dry seeds, such as chickpeas, lentils, peas, and beans.**
- **Manufacturers are incorporating pulses, a good source of plant-based protein and other nutrients, into a wide array of foods.**
- **Challenges facing the pulse industry include improving protein quality, reducing antinutritive factors, enhancing sensory attributes, and accelerating pulse breeding and agricultural innovation.**

Members of the legume family, pulse seeds grow in pods and vary widely in shape, size, and color. The FAO defines pulses as crops that are harvested solely for their dry seeds. This definition excludes oilseeds, such as soybean and peanut, as well as seeds that are eaten in their immature form as vegetables, such as green peas and green beans. The FAO has recognized 11 primary pulses, including dry beans (e.g., kidney, lima, pinto, navy), dry broad beans (fava, horse), dry peas (garden), chickpeas, dry cowpeas (black-eyed pea), pigeon peas, lentils, bambara beans (groundnuts), vetches (used mainly for animal feed), lupines (used mainly for animal feed and as ornamental flowers), and minor pulses (winged bean, guar bean) (https://tinyurl.com/FAO-pulses).

CHANGING DIETS

Archeological sites in India, Egypt, Mesopotamia, the Mediterranean, and Switzerland indicate that humans have been cultivating pulses for thousands of years. Currently, India leads the world in the production and consumption of pulses. Other major producers, in order of total production, include Canada, Myanmar, China, Brazil, and Australia (FAO, https://tinyurl.com/FAO-pulsebook, 2016). From 1990 to 2014, pulse production grew at the fastest annual rate (7.7%) in North America, largely due to a greater than 10-fold increase in Canadian pulse production.

Although worldwide pulse production is increasing, per capita consumption has witnessed a long-term decline. From 1961 to 2001, India's per capita pulse consumption decreased from 24 kg to 12 kg per year (Canadian Special Crops Association, https://tinyurl.com/CSPA-2014, 2014). During the same period, China's pulse consumption fell from 10 kg to 1 kg. Meanwhile, North America's per capita pulse consumption remained modest but steady at about 3.5 kg per year. In developing economies such as India and China, increased urbanization and rising incomes have contributed to reductions in pulse consumption, with concomitant increases in the consumption of meat, dairy, and processed foods.

However, as the world population soars toward an estimated 9.6 billion in 2050, experts predict that meat will become scarce and expensive, and the demand for plant-based proteins like pulses will rise dramatically. Projections indicate a 23% increase in global pulse consumption by 2030, with a more rapid increase (about 50%) in Africa (Global Pulse Confederation, https://tinyurl.com/GPC-strategy, 2016).

INVESTING IN PULSES

Many governments, universities, food companies, and investors have recognized the potential of pulses. Canada is emerging as a pulse powerhouse. In the early 1900s, research conducted by the Canadian Department of Agriculture (now known as Agriculture and Agri-Foods Canada; AAFC) identified two varieties of field peas that are suitable for growth in Canada. "Further research carried out since the late 1970s by AAFC and Canadian universities has developed suitable varieties of beans, lentil, chickpea, faba bean, and other pulses for prairie climate and growing seasons, while providing high yield and resistance to pests and pathogens," says Janitha Wanasundara, research scientist at AAFC, in Saskatoon, Saskatchewan. The fruits of these labors are being realized: Canada is now the world's largest pulse exporter, with most exports going to India (https://tinyurl.com/FAO-pulsebook).

Canadian pulse production, which includes beans, chickpeas, lentils, and peas, takes place primarily in the provinces of Saskatchewan, Alberta, and Manitoba. Canada leads the world in the production of dry peas, which are attracting increasing interest from food companies as a source of pea flour and protein. In September 2017, French food company Roquette announced that it would build the world's largest pea-processing plant in Portage la Prairie, Manitoba. Also in 2017, Verdient Foods launched a "massive" organic pea-processing plant (funded in part by movie director James Cameron) in Vanscoy, Saskatchewan (Hui, A., https://tinyurl.com/pulses-future, 2017).

However, the pulse boom in Canada is showing signs of a slowdown in 2018 (Skerritt, J., https://tinyurl.com/pulse-bust, 2018). A surge in pea and lentil production since 2014 led to a global oversupply, which caused prices to plummet. In December 2017, India imposed a 30% duty tax on chickpea and lentil imports. Now farmers have swapped some of their pulse acreage for canola and wheat. According to the AAFC, pea plantings in Canada will likely decline to a seven-year low in the spring of 2018, whereas lentil acreage will drop 27%.

US-based company Cargill (Minnetonka, Minnesota) is investing in pea-based ingredients. In January 2018, the company signed a joint venture agreement with PURIS (Minneapolis, Minnesota), North America's largest producer of pea protein (Wyers, R., https://tinyurl.com/pea-protein-surge). PURIS produces pea protein, starch, fiber, and flour from

organic, non-genetically modified (non-GM) peas grown in the Midwestern United States. Cargill's financial backing will allow PURIS to expand its operations and build a second plant in the United States.

PULSE APPEAL

Consumers are increasingly attracted to pulses because they satisfy several trends: plant-based protein, non-GMO, gluten-free, and clean label. Increased globalization has made people more aware of pulses, says Wanasundara. "The food items on our plate have been changing," she says. "Migration of different cultural groups across the globe, increasing awareness of different cuisines, and the search for new ingredients have contributed to an increased use of pulses."

"In Europe, people are becoming more aware of the problems caused by the massive production of animal-based foods, so there's an interest in plant proteins," says Raffael Osen, deputy head of the Department of Process Development at the Fraunhofer Institute for Process Engineering and Packaging in Freising, Germany. "Many consumers have problems with soy due to GMO issues, as well as the image that soy is grown in former rainforest areas in South America." In contrast, consumers are familiar with peas, beans, and lentils, which are "locally grown" in European countries.

Because ingredients like "pea protein" and "lentil flour" are easily understood by most consumers, manufacturers can use them to replace synthetic or less familiar ingredients for clean-label claims. And since pulses are gluten-free, they appeal to consumers with celiac disease, as well as the increasing number of people who consider themselves sensitive to gluten. Some food manufacturers who incorporate pulse-based ingredients advertise their products as "allergen-free" or "allergen-friendly" to differentiate them from soy protein, which must be labeled as an allergen in North America, the European Union, and other areas.

"I think this form of product positioning is a bit premature," says Phil Kerr, president and founder of SERIO Nutrition Solutions, LLC, in St. Louis, Missouri, USA. "In many instances, the reason why foods are on the major allergen list is because there's large exposure to the global population. As the exposure to pulses gets larger, they may very well be recognized as allergens like other legumes, such as peanuts and soybeans." Indeed, allergies to lentils and lupines, which often overlap with peanut allergies, have already been reported. In addition, about 400 million people worldwide have a genetic disorder called glucose-6-phosphate dehydrogenase deficiency, which makes them susceptible to "favism"—a hemolytic response to fava bean consumption.

A

Nutrition Facts

Serving size	1/2 Cup (125 mL) Cooked
Amount per serving	
Calories	**140**
	% Daily Value*
Total Fat 0.5g	1%
Saturated Fat 0g	0%
Trans Fat 0g	
Cholesterol 0mg	0%
Sodium 5mg	0%
Total Carbohydrate 23g	8%
Dietary Fiber 9g	32%
Total Sugars 0g	
Includes 0g Added Sugars	0%
Protein 12g	
Vitamin D 0mcg	0%
Calcium 25mg	2%
Iron 2mg	10%
Potassium 252mg	6%
Folate 39mcg DFE	10%

*The % Daily Value tells you how much a nutrient in a serving of food contributes to a daily diet. 2,000 calories a day is used for general nutrition advice.

B

Nutrition Facts

Serving size	1/2 Cup (125 mL) Cooked
Amount per serving	
Calories	**150**
	% Daily Value*
Total Fat 0.5g	1%
Saturated Fat 0g	0%
Trans Fat 0g	
Cholesterol 0mg	0%
Sodium 5mg	0%
Total Carbohydrate 25g	9%
Dietary Fiber 4g	14%
Total Sugars 0g	
Includes 0g Added Sugars	0%
Protein 12g	
Vitamin D 0mcg	0%
Calcium 12mg	0%
Iron 3mg	15%
Potassium 273mg	6%
Folate 55mcg DFE	15%

*The % Daily Value tells you how much a nutrient in a serving of food contributes to a daily diet. 2,000 calories a day is used for general nutrition advice.

FIG. 1. Nutrition facts for A) whole green lentil (boiled) and B) split red lentil (boiled). Credit: Lentils.org

PROTEIN QUANTITY AND QUALITY

With a crude protein content of approximately 21–26% by weight, pulses are a much better source of protein than cereal grains such as wheat, barley, and quinoa (Nosworthy, M.G., and House, J.D., http://dx.doi.org/10.1094/CCHEM-04-16-0104-FI, 2017). The protein content of pulses can be influenced by both genetic and environmental factors. For example, the protein content in 59 different pea lines ranged from 13.7–30.7% by weight, with an average of 22.3% (Tzitzikas, E.,N. *et al.*, http://dx.doi.org/10.1021/jf0519008, 2006). Pulses are also rich in complex carbohydrates and dietary fiber, low in fat, and a good source of several vitamins and minerals such as calcium, folate, potassium, and iron (Fig. 1).

To increase the protein content of pulse ingredients, manufacturers make pulse protein concentrates or isolates (Nosworthy, M.G., *et al.*, http://dx.doi.org/10.1094/CFW-62-4-0139, 2017). Pulse protein concentrates are produced by a "dry" method called air classification. Seeds are milled to produce a fine flour. Then, the flour is passed through a spiral air stream to separate fine particles (protein) from coarse particles (starch). Air classification does not completely remove starch granules from the protein fraction, but it does increase the overall protein content. For example, in one study, air classification increased the protein content of peas from 21.5 to 56.3%, of lentils from 19.5% to 49.3%, and of fava beans from 31.6 to 75.1% (Elkowicz, K, and Sosulski, F.W., https://doi.org/10.1111/j.1365-2621.1982.tb07673.x, 1982).

In contrast, pulse protein isolates are produced by the "wet" method of alkaline extraction followed by protein precipitation (Nosworthy, M.G., *et al.*, http://dx.doi.org/10.1094/CFW-62-4-0139, 2017). Milled flour is incubated in an alkaline solution (pH 8–11), which solubilizes most of the protein but not other flour components, such as starch. Insoluble material is removed by filtration or centrifugation. Then, the pH of the solution is decreased to an appropriate isoelectric point, and the protein precipitates. The protein precipitate is collected

by centrifugation and washed, neutralized, and dried. Protein contents of 90.1–90.8%, 86.7–89.3%, and 95.3% for pea, lentil, and fava bean isolates, respectively, have been reported (Nosworthy, M.G., *et al.*, http://dx.doi.org/10.1094/CFW-62-4-0139, 2017).

Isolates have a higher protein content than concentrates. In addition, the alkaline extraction reduces the activity of antinutritive factors that affect protein digestibility, such as trypsin inhibitors and hemagglutinins. As a result, pulse protein isolates are generally more digestible than concentrates.

In the European Union, food manufacturers can make protein claims on packages based solely on protein content, and not quality (Marinangeli, C.P.F., and House, J.D., https://doi.org/10.1093/nutrit/nux025, 2017). If a food derives 12% of its energy from protein, it can be labeled as a "source of protein." Greater than 20% energy from protein qualifies for a "high source or protein" claim. However, in North America, claims of "good" or "excellent" sources of protein require evidence of protein quality (Nosworthy, M.G., and House, J.D., http://dx.doi.org/10.1094/CCHEM-04-16-0104-FI, 2017). The quality of a dietary protein is determined by two factors: 1) the amino acid composition and its correspondence with human amino acid requirements, and 2) the digestibility of the protein. Typically, proteins from animal sources (beef, chicken, pork, eggs, and dairy) are the best suited to human requirements, while also being highly digestible. In contrast, plant-based proteins often lack one or more indispensable amino acids, and they are less digestible than animal proteins.

There are three major methods to determine protein quality, each with its own advantages and disadvantages (Nosworthy, M.G., and House, J.D., http://dx.doi.org/10.1094/CCHEM-04-16-0104-FI, 2017) (Fig. 2, page 10). The Protein Efficiency Ratio (PER), which is the method accepted by the Canadian government, uses a rodent feeding trial to determine the quality of a test protein compared with a reference protein (the dairy protein casein). Researchers feed an experimental diet or a casein control diet, which each consists of 10% protein, to weanling rats for 28 days. Then, the total weight gain of the rats is divided by the amount of protein consumed. To standardize PER across laboratories, the raw PER value is adjusted to the average PER of casein (2.5). The protein rating of a food is calculated by multiplying the adjusted PER by the grams of protein in a reasonable daily intake. For example, white bread has 12.6 g of protein in a reasonable daily intake of 150 g (8.4% protein), and a PER of 1.0, so the protein rating is 12.6. In Canada, foods must have a protein rating of at least 20 to be labeled a "good" source of protein, and of at least 40 to be considered an "excellent" source of protein.

PER is a simple calculation that requires only the measurements of weight gain and protein intake. Another advantage is that PER reveals the actual impact of a protein source on growth. However, the method assumes that all of the protein the rat consumes is used for growth, and neglects contributions to maintenance and other metabolic processes. Also, rats require much higher levels of the sulfur-containing amino acids (cysteine and methionine) than humans do, which could underestimate the quality of many pulse proteins that are limiting in

A. Protein Efficiency Ratio (PER)

$$PER = \frac{Weight\ gain\ (g)_{Test\ Protein} \div Mass\ of\ protein\ consumed\ (g)_{Test\ Protein}}{Weight\ gain\ (g)_{Casein} \div Mass\ of\ protein\ consumed (g)_{Casein}}$$

$$PER_{Adjusted} = PER \times 2.5 \qquad \text{(Average PER of Casein = 2.5)}$$

$$Protein\ Rating\ = Protein\ in\ a\ Reasonable\ Daily\ Intake\ of\ a\ Food\ (g) \times PER_{Adjusted}$$

Protein Rating ≥ 20 → "Good" source of protein
Protein Rating ≥ 40 → "Excellent" source of protein

B. Protein Digestibility Corrected Amino Acid Score (PDCAAS)

$$Amino\ acid\ score\ for\ each\ IAA = \frac{IAA\ (mg)\ per\ g\ total\ protein_{Food}}{Reference\ pattern\ IAA\ (mg)\ per\ g\ protein}$$

$$PDCAAS_{Food} = Lowest\ amino\ acid\ score\ for\ IAA \times True\ fecal\ nitrogen\ digestibility$$

(PDCAAS values > 1.0 are truncated to 1.0)

$$Corrected\ protein\ level\ in\ a\ food\ = PDCAAS_{Food} \times Protein\ per\ serving\ (g)$$

$$\%\ DV = \frac{Corrected\ protein\ level\ (g)}{50\ g\ DV\ protein}$$

% DV ≥ 10% → "Good" source of protein
% DV ≥ 20% → "Excellent" source of protein

C. Digestible Indispensable Amino Acid Score (DIAAS)

$$Level\ of\ each\ digestible\ IAA\ in\ the\ food = IAA\ (mg) \times Ileal\ IAA\ digestibility\ coefficient$$

$$Amino\ acid\ score\ for\ each\ IAA\ = \frac{Digestible\ IAA\ (mg)\ per\ g\ protein_{Food}}{Reference\ pattern\ IAA\ (mg)\ per\ g\ protein} \times 100$$

$$DIAAS_{Food} = Lowest\ amino\ acid\ score\ for\ IAA$$

Crude protein 5–9.9 g *and* DIAAS ≥ 75 → "Good" source of protein
Crude protein ≥10 g *and* DIAAS ≥ 100 → "Excellent" source of protein

FIG. 2. Three methods for calculating protein quality for protein quality claims: PER (A), PDCAAS (B), and DIAAS (C). PER, used by Canada, measures weight gain in rats over 28 days as a function of their protein intake. PDCAAS, used by the United States, takes into account the amino acid score of a protein and its true fecal nitrogen digestibility. DIAAS, recommended by the Food and Agriculture Organization (FAO) and the World Health Organization (WHO) but not yet implemented by any country, considers the amino acid score of a protein and the ileal digestibility of each indispensable amino acid (IAA). Credit: Adapted from Marinangeli, C.P.F., and House, J.D., http://dx.doi.org/10.10.93/nutrit/nux025, 2017.

these amino acids (Nosworthy, M.G., and House, J.D., http://dx.doi.org/10.1094/CCHEM-04-16-0104-FI, 2017).

In 1989, the FAO and the World Health Organization (WHO) established the Protein Digestibility Corrected Amino Acid Score (PDCAAS) as the preferred method to measure protein quality (Nosworthy, M.G., and House, J.D., http://dx.doi.org/10.1094/CCHEM-04-16-0104-FI, 2017). PDCAAS is a more quantitative measure of protein quality than PER: The method measures the amount of each indispensable amino acid in a protein sample, as well as the protein sample's digestibility. The amino acid score reflects the amount of each indispensable amino acid (lysine, methionine plus cysteine, threonine, and tryptophan) in a protein sample compared with the amount of the same amino acid in a reference provided by the FAO (typically, the essential amino acid requirements of a child aged 6 months to 3 years). An amino acid score of 1.0 or greater indicates no deficiency in that amino acid. The amino acid with the lowest score is con-

sidered to be the first limiting amino acid in that protein, and its amino acid score is used in the calculation of PDCAAS. In the United States, PDCAAS is used to regulate most protein claims.

The other portion of the PDCAAS calculation, true fecal nitrogen digestibility, is determined by a rodent assay (Nosworthy, M.G., and House, J.D., http://dx.doi.org/10.1094/CCHEM-04-16-0104-FI, 2017). Rats are fed a controlled diet, and the amount of nitrogen in their feces is subtracted from the amount of nitrogen they ingested, with a correction for the contribution of endogenous protein to the total fecal nitrogen. To calculate PDCAAS, the lowest amino acid score is multiplied by the true fecal nitrogen digestibility. PDCAAS values are truncated at 1.0 (the value set for casein). Then, the corrected protein level can be determined by multiplying the PDCAAS by the amount of total protein in a serving. Finally, the percent daily value is calculated by dividing the corrected protein level by 50 g, the daily reference value for protein recommended by the US Food and Drug Administration. For a product to make a "good" source of protein claim, it must have a percent daily value of at least 10%. An "excellent" source of protein contains at least 20% of the daily reference value.

In 2011, an Expert Consultation of the FAO and WHO recommended replacing the PDCAAS with a different measure of protein quality, the Digestible Indispensable Amino Acid Score (DIAAS) (Nosworthy, M.G., and House, J.D., http://dx.doi.org/10.1094/CCHEM-04-16-0104-FI, 2017). The DIAAS is thought to be more accurate than the PDCAAS because it measures ileal digestibility, rather than fecal digestibility. Feces typically contain a significant amount of bacterial protein contaminants, with a final amino acid composition that is quite different from that in the ileum (the final section of the small intestine). For example, in growing pigs, fecal digestibility is about 22% greater than ileal digestibility, which could cause protein quality to be overestimated by PDCAAS. Another advantage is that DIAAS considers the ileal digestibility of each amino acid in a protein sample, rather than the digestibility of the total protein, as in PDCAAS.

To calculate DIAAS, researchers compare the amount of each digestible indispensable amino acid in a protein sample with that of a reference pattern, and multiply this ratio by 100. The DIAAS corresponds to the lowest amino acid score for the indispensable amino acids. Unlike PDCAAS, DIAAS values are not truncated, making it easier to assess the quality of protein blends. For DIAAS, protein claims are based on both quantity and quality. Foods with a crude protein content of 5–9.9 g and DIAAS values of at least 75 qualify for a "good source of protein" claim, whereas foods with a crude protein content of at least 10 g and a DIAAS of at least 100 are "excellent" sources of protein.

Although DIAAS likely provides a more accurate assessment of protein quality than PER or PDCAAS, ileal sampling is quite invasive. Therefore, most of the data on ileal digestibility comes from pig or rodent models, rather than humans (Nosworthy, M.G., and House, J.D., http://dx.doi.org/10.1094/CCHEM-04-16-0104-FI, 2017). Because of this and other scientific and economic hurdles, a 2014 FAO/WHO Expert

Consultation recommended that additional research be conducted before DIAAS replaces PDCAAS as the standard method to validate protein claims (Lee, W.T.K., *et al.*, http://doi.org/10.3945/jn.115.222109, 2016). As of yet, no jurisdiction uses the DIAAS to verify protein claims.

PDCAAS values are usually higher than DIAAS values for the same protein, but the differences are generally small (1.8–9%) for plant-based proteins (Marinangeli, C.P.F., and House, J.D., https://doi.org/10.1093/nutrit/nux025, 2017). However, because the cutoff for DIAAS protein claims is set relatively higher than for PDCAAS claims, some pulse proteins that would qualify as "good" sources of protein under the PDCAAS system would be ineligible for a protein claim under the DIAAS method.

Pulses vary in their limiting indispensable amino acid. Cooked Canadian red kidney beans, whole green lentils, split red lentils, split green peas, and black beans are limited by the sulfur-containing amino acids (methionine and cysteine) (Nosworthy, M.G., *et al.*, http://dx.doi.org/10.1002/fsn3.473, 2017). In contrast, cooked Canadian navy beans, split yellow peas, chickpeas, and pinto beans are limited by tryptophan.

Table 1 shows the amino acid score, total protein digestibility, and PDCAAS of several pulses, as well as wheat flour, rice flour, soy, and casein. Pulses generally have a higher PDCAAS than cereal grains, but a lower PDCAAS than soy or animal proteins (e.g., casein). Although pea protein isolates are more digestible than concentrates, they have a lower amino acid score because the alkaline extraction process alters the amino acid composition of the isolated protein. Therefore, the PDCAAS values of pea protein isolate and pea protein concentrate are roughly equivalent (Table 1).

Many pulse proteins do not have sufficiently high PDCAAS values to make protein claims at the amount of protein present in a typical serving. However, combining pulse proteins with other plant protein sources, such as cereal grains, can improve the overall amino acid score, and thus the weighted-average PDCAAS. Pulses tend to be higher in lysine and lower in sulfur-containing amino acids than cereal grains such as wheat and rice, whereas cereals are lower in lysine and higher in the sulfur-containing amino acids. "We can partner pulses with other commodities that offset their amino acid deficiencies," says James House, professor and head of the Department of Food and Human Nutritional Sciences at the University of Manitoba, in Winnipeg, Canada. "The classic example is the South American staple of beans and rice. By creating pulse-cereal blends, we can obtain higher-quality proteins."

POSSIBLE HEALTH BENEFITS OF PULSES

In addition to being a good source of protein, pulses may benefit cardiovascular health, weight management, and gastrointestinal function (Dahl, W.J., *et al.*, https://doi.org/10.1017/S0007114512000852, 2012). Some studies also suggest that pulses could act as prebiotics by improving intestinal microbial homeostasis. However, evidence for these benefits is currently inconsistent. Long-term studies of large populations are required to investigate possible roles of pulses in disease prevention.

TABLE 1. PDCAAS values for various protein sources

Protein	Amino Acid Score	True Protein Digestibility (%)*	PDCAAS
Pea (yellow, split)	0.73	87.9	0.64
Pea (green, split)	0.59	85.2	0.50
Lentil (green, whole)	0.71	87.9	0.63
Lentil (red, split)	0.59	90.6	0.54
Chickpeas (Kabuli)	0.61	85.0	0.52
Pinto Beans	0.77	76.2	0.59
Kidney Beans	0.70	78.6	0.55
Black Beans	0.76	70.0	0.53
Navy Beans	0.83	80.0	0.67
Wheat Flour	0.47	92.3	0.43
Rice Flour	0.54	92.0	0.50
Soy Flour (50% protein)	0.92	83.5	0.77
Pea protein Isolate (82% protein)	0.54	97.1	0.53
Pea Protein Concentrate (50% protein)	0.58	92.6	0.54
Soy Protein Isolate (93% protein)	0.87**	96.0	0.84
Casein	1.04	96.6	1.00

*True fecal nitrogen digestibility

**Other sources (e.g., Hughes, G.J., *et al.*, http://dx.doi.org/10.1021/jf203220v, 2011) have calculated a PDCAAS for soy protein isolate of 1.00.

Credit: "Protein quality of cooked pulses," Pulse Canada (https://tinyurl.com/pulsecanada-cooked)

Pulses are high in resistant starch, which is less easily broken down into glucose by digestive enzymes in the small intestine and then absorbed into the blood than other forms of starch. Bacteria in the large intestine transform resistant starch into short-chain fatty acids, which contribute to gastrointestinal health. The high contents of resistant starch and fiber in pulses make them low-glycemic foods, which could be beneficial in the prevention and management of type 2 diabetes (Dahl, W.J., *et al.*, https://doi.org/10.1017/S0007114512000852, 2012).

With a glycemic index (GI) of 39–55, pulses are less likely to affect blood sugar levels than white rice (GI 80), white bread (GI 100), and potatoes (GI 121). Studies suggest that including pulse flours in cereal-based food products can help reduce the glycemic profile of these foods. For example, an extruded breakfast cereal containing 56.5% pea flour or pea semolina, 31.5% corn meal, 6.5% pea fiber, 0.5% salt, and 5% sugar

released about 20% less glucose *in vitro* than the same cereal lacking the pea ingredients (Pulse Canada, https://tinyurl.com/pulsecanada-pea).

In a crossover clinical trial involving 23 overweight, hypercholesterolemic men and women, researchers compared the effect of muffins made with white-wheat flour, whole pea flour, or fractionated pea flour (pea hulls only) on insulin resistance over a 28-day period (Marinangeli, C.P, and Jones, P.J., https://doi.org/10.1017/S0007114510003156, 2011). They found that eating two muffins a day containing either whole pea flour or fractionated pea flour reduced fasting insulin levels by about 20% compared with muffins containing wheat flour. Also, the muffins containing pea ingredients lowered estimates of insulin resistance by about 25% compared with the muffins made from wheat flour.

A recent meta-analysis of 21 clinical trials involving 940 adults found that eating one serving per day of beans, peas, chickpeas, or lentils could contribute to modest weight loss (Kim, S. J., *et al.*, https://doi.org/10.3945/ajcn.115.124677, 2016). The study participants lost an average of 0.34 kg (0.75 pounds) over 6 weeks by incorporating one serving a day of pulses, without making an effort to reduce other foods. The weight loss may have been due to the low GI of pulses and/or the increased feeling of satiety that pulses appear to induce. "Though the weight loss was small, our findings suggest that simply including pulses in your diet may help you lose weight, and we think more importantly, prevent you from gaining it back after you lose it," says Russell de Souza, assistant professor at McMaster University, in Hamilton, Canada.

ANTINUTRITIVE FACTORS

Although rich in several nutrients, pulses also contain antinutritive factors that limit digestibility and nutrient absorption, including phytic acid, protease inhibitors, phenolic compounds (tannins, phenolic acids), lectins (hemagglutinins), saponins, and oxalates. Phytic acid is a storage form of phosphorous that binds minerals and prevents their absorption by the small intestine. A good source of some minerals, such as potassium, phosphorus, magnesium, and calcium, pulses could help correct micronutrient deficiencies. However, phytic acid limits the bioavailability of the minerals in pulses, particularly calcium, zinc, and iron. Trypsin and other proteases are enzymes involved in the digestion of dietary proteins. Protease inhibitors interfere with their actions, thus reducing the digestibility of pulse proteins. Similarly, tannins form complexes with proteins and digestive enzymes, inhibiting their activities.

Cooking pulses can alter the amount of antinutritive factors (Nosworthy, M.G., and House, J.D., http://dx.doi.org/10.1094/CCHEM-04-16-0104-FI, 2017). The method of cooking appears to affect antinutrient composition. For example, traditional cooking methods (roasting, boiling) are more effective at destroying lectins, whereas microwaving is better at reducing trypsin inhibitor activity. Food processing techniques such as soaking, autoclaving, micronization, extrusion, germination, and fermentation also alter the antinutritive content and, therefore, protein digestibility and nutrient availability.

PULSE SUSTAINABILITY

Many consumers view pulses as sustainable, environmentally friendly alternatives to animal protein sources. Pulses require less water, energy, and nitrogen fertilizer to produce than most animal and plant protein sources (Global Pulse Confederation, https://tinyurl.com/GPC-strategy, 2016). Many pulse crops are well adapted to arid and semi-arid climates, and they can tolerate drought and frost stress better than most crops. Pulse cultivation occurs over a wide geographical area. "In contrast to soy, where three countries—the Unites States, Brazil, and Argentina—really dominate the global supply, pulse crops are grown all over the world," says Kerr.

For many years, farmers have recognized the key role that pulses and other legumes play in crop rotation. Legumes harbor symbiotic, nitrogen-fixing bacteria in structures called root nodules. These bacteria, known as rhizobia, fix nitrogen gas (N_2) from the atmosphere into ammonia (NH_3), which is eventually released from the plant's roots into the soil. In this way, pulses are a more environmentally friendly alternative to nitrogen fertilizers.

FOOD APPLICATIONS

Pulse flours, concentrates, isolates, and extracts are being incorporated into a wide range of foods beyond the traditional uses, including breads, pastas, snacks, breakfast cereals, meats, desserts, and beverages. Pulse flour has been used to wholly or partially replace wheat flour in baked goods and pasta. Chickpea brine can be whipped into a foam and used as an egg white substitute. Pulse purees have been used in desserts to reduce the fat and butter content. Pea protein is being used to make dairy-free cheeses. Pea starch can replace cornstarch in coatings for French fries, mozzarella sticks, and onion rings. Pulse ingredients are being explored as emulsifiers and foaming, gelling, and thickening agents in sauces and beverages. Beyond Meat, a company based in Los Angeles, California, USA, sells plant-based burgers made from pea protein isolates and chicken substitutes made from a mixture of pea and soy proteins.

In one study, yellow pea flour was incorporated into a spaghetti formulation at 30% inclusion with durum semolina (Pulse Canada, https://tinyurl.com/Pulsecanada-pasta). The resulting spaghetti had similar quality attributes to 100% durum spaghetti, including texture, cooking time, and cooking loss. The spaghetti containing pea flour was also higher in protein and fiber than the 100% durum spaghetti. Color was the major difference between the two pastas. The spaghetti made with pea flour was less bright, more red, and firmer than the regular spaghetti (Fig. 3A, page 14). However, by adjusting the drying conditions of the pea-flour-containing spaghetti, researchers could increase the brightness of the pasta by 20%, reduce the redness by more than 50%, and decrease the firmness by 30% (Fig. 3B, page 14).

Pulses flours have also been tested as binders, fillers, and extenders in ground meat products. For example, Wanasundara, Phyllis Shand at the University of Saskatchewan, and colleagues tested chickpea flour as an extender (a non-

meat substance with substantial protein) for low-fat pork bologna (Sanjeewa, W.G.T., *et al.*, https://doi.org/10.1016/j.foodres.2009.07.024, 2010). To reduce the fat content of processed meat, formulators can substitute water for fat, but this typically changes the texture and water-holding ability of the meat. However, adding 5% chickpea flour to low-fat bologna increased the firmness, decreased water release, and increased cook yield, with flavor properties similar to the control bologna.

In another study, Wanasundara and colleagues with InfraReady Products (Saskatoon, Canada) found that adding micronized lentil flour to red meat improves color stability and slows lipid oxidation in fresh and frozen meat (Cross, B., https://tinyurl.com/lentils-beef, 2013). Micronization is a process in which a substance is treated with infrared radiation, which causes the substance to reach very high temperatures (750–930°C) in a short time. Micronization of grains and pulses causes the seeds to swell and rupture, reducing their cooking time and altering other physical and chemical properties. For example, micronization can improve pulses' PER, indispensable amino acid score, and shelf stability, while decreasing undesirable attributes such as tannins, phytic acid, trypsin inhibitor and lipoxygenase activities, and oligosaccharides.

The researchers added micronized or non-micronized green lentil flour to ground meat at levels of 5–12% total volume. Wheat flour, which is a common binding agent, is typically added at 5%. The team found that the micronized lentil flour was better at maintaining meat redness and delaying the oxidation of pigments and lipids than the non-micronized flour. The lentil flour also provided protein and contributed to increased meat firmness and juiciness. "The micronized lentils brought a nice nutty flavor to the product," adds Wanasundara.

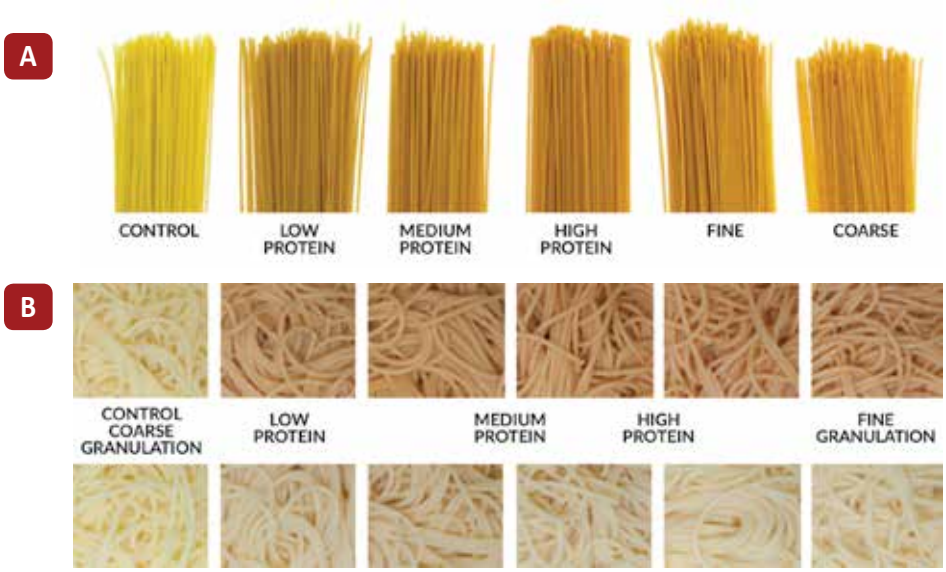

FIG. 3. (A) Spaghetti made with 100% durum semolina (control) or 30% yellow pea flour/70% durum semolina with different protein contents and fine or coarse flour milling.(B) Cooked spaghetti made with 30/70% yellow pea flour/durum semolina, prepared under high temperature-short time drying conditions (top row) or low temperature-long time drying conditions (bottom row).
Credit: Pulse Canada, https://tinyurl.com/Pulsecanada-pasta

Osen and colleagues used sweet lupine to formulate a refreshing, protein-rich beverage (Fraunhofer Institute, https://tinyurl.com/lupine-drink, 2017) (Fig. 4). Although lupine is perhaps best known as an ornamental flower, the plant's seeds are a traditional pickled snack food in some parts of the world, such as the Mediterranean basin and Latin America. Osen chose lupine for the protein-rich beverage because this pulse contains protein that dissolves in the acidic pH range, whereas most other proteins are not soluble at low pH. A slightly acidic drink tastes refreshing. However, like other pulses, lupine con-

FIG. 4. (A) Cultivated sweet lupine. (B) A refreshing, protein-rich drink made from the extract of sweet lupine. Credit: Fraunhofer IVV

tains phytic acid, which binds minerals and inhibits enzymes, limiting the drink's digestibility. To reduce the phytic acid content, Osen and colleagues used a two-stage mashing and fermentation process, during which microorganisms degraded the phytic acid by hydrolysis. The result was a lupine protein isolate in the form of paste or powder, with a relatively neutral taste. Although the drink is non-alcoholic, the process is similar to beer brewing and can be conducted at any brewery.

FUTURE CHALLENGES

Although pulses are poised to become a major food trend, there are still some issues that hinder their widespread use. Sensory attributes are one such challenge. Pulses generally impart an off-flavor to foods, ranging from "fruity" to "beany" to "earthy" to "bitter." In some cases, these flavors may add to the appeal of the food, but in other cases, they are undesirable. Deflavoring methods are being investigated, and some have been patented by food companies. The details of such methods are usually proprietary but involve the removal or modification of volatile and nonvolatile compounds that are responsible for the undesirable sensory characteristics (Nosworthy, M.G., *et al.*, http://dx.doi.org/10.1094/CFW-62-4-0139, 2017).

If pulse crops are to meet predicted future demands, agricultural innovation, which lags behind cereal and oilseed crops, must accelerate. Over the past few years, global pulse crop production has remained relatively stagnant in terms of yield per hectare, hectares planted, and total volume produced (Global Pulse Confederation, https://tinyurl.com/GPC-strategy, 2016). Pulse crops must be further optimized for yield and resistance to disease, pests, weeds, and drought. There are also opportunities to improve protein content and quality, minimize antinutrients, shorten cooking time, and improve sensory attributes.

"Protein quality is not a typical breeding target for pulses, but we're trying to encourage that," says House. "However, it's very expensive and time-consuming to analyze amino acids, and when you're a breeder, you've got thousands of samples to screen. We're trying to give new tools to breeders so that they can more quickly assess amino acid composition in germplasm."

Thus far, pulse producers have embraced classical breeding over genetic modification, perhaps to avoid the controversy that GMOs often generate in pulses' target market (people seeking plant-based proteins and soy alternatives). Yet transgenic approaches and gene editing can rapidly accelerate the rate of trait discovery and development. "We'll probably see some efforts using new technology, not necessarily transgenics, but perhaps gene editing approaches such as CRISPR technology, to make more rapid advancements in pulse breeding," says House. However, he notes that the distinction between genetic modification and gene editing can be challenging to communicate to the average consumer.

According to Kerr, the infrastructure and manufacturing expertise for pulses are currently at the stage soy was at about 40 years ago. "Pulse crops are at a very exciting point in their

development. There's tremendous motivation for companies around the world to look at pulse crops as another protein-rich raw material that can be consumed in whole form or refined into various ingredients and then used in new food products," says Kerr. "The pulse industry will need organizations like AOCS that have a long history of expertise in not only the biotechnology of the raw material, but also in the processing technology, nutrition science, and food chemistry needed to make and use these ingredients."

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Quantitative physical stability analysis of concentrated, fragrance-infused liquid fabric care dispersions

Matt Vanden Eynden, Christelle Tisserand, Yoann Lefeuvre, Pascal Bru, and Gerard Meunier

- Multiple light scattering technology allows for passive, quantitative stability analysis of concentrated liquid dispersions.
- Individual destabilization phenomena, such as creaming and clarification, can be plotted to compare specific kinetics of each sample.
- Fragrance-infused detergents and fabric softener formulations are distinguishable in only seven hours, days ahead of what is seen visually with the naked eye.

Kinetically stable mixtures, such as certain liquid dispersions, typically do not show signs of phase separation during the shelf life of the product. Specifically, liquid laundry detergents, liquid fabric softeners, and laundry gels contain many ingredients but can still be shelf-stable for long periods of time. In contrast, the addition of fragrance compounds or microcapsules can rapidly cause instability within such mixtures and can result in cloudiness, flocculation, and eventual phase separation.

Analyzing the stability of such dispersions and emulsions can help maintain product integrity by making it possible to predict shelf life and optimize the formulation, but relying on the human eye to detect phase separation is inefficient, time-consuming, and subject to guesswork. As a result, stability testing at higher temperatures and with non-objective techniques, such as the eye test or shelf test, can require weeks or even months of an operator's time. Fortunately, since the instability and eventual phase separation is caused by particle migration and particle size change events, and the rate at which these inevitable events occur ultimately correlate to the shelf life of the product, much faster and quantitative methods can be used to explore the integrity of these dispersions.

In this article, we describe how a Multiple Light Scattering (MLS) technique employed by the Turbiscan™ device can be used to quickly and accurately determine destabilization phenomena such as sedimentation, creaming, flocculation, coalescence, as well as complete phase separation. The Turbiscan™ can detect both transmission (T) and backscattering (BS) intensity of light as a mobile reading head moves ver-

tically about a sample in a glass cell. At every 40 µm, light scattering data is collected to provide information about the size and concentration of the scattered particles in the medium as stated by Mie scattering theory. This technique allows for the tracking of all scattering particles in concentrated media, as high as 95% w/v, as they move throughout the entire sample height during the natural ageing process of a sample.

Quantifying such kinetics will provide insight into long-term stability and allow the operator to quickly modify any defect in the formulation rather than wait days or weeks for shelf- and bottle-tests to complete.

In a recent study, we used MLS to test the stability of a stable liquid detergent formulation once it was modified with an original and then an optimized perfume component. In addition, a similar test was performed with a liquid fabric softener to analyze the effect of encapsulated perfumes and fragrance-free oils. Individual backscattering profiles provided detailed insight into the potential phase separation before any phase separation was visible to the naked eye. Overall stability results showed how stability of varying fragrances affected the short- and long-term stability of these formulations, providing not only a resource for fast formulation optimization but for advanced long-term shelf life studies.

To test the stability performance of a heavy-duty liquid detergent, three emulsion formulations were studied. All emulsions were analyzed by the Turbiscan™ for only 7 hours at a temperature of 45°C with a scanning frequency of every 10 minutes. Figure 1 depicts the type of graph that is generated.

The color profile of the graph displays early scans in blue and later scans in red. Consequently, it is easy to see that the left side of the graph, representing the bottom of the vial, displays a growing negative change in the BS signal as oil particles migrate away from this region toward the top of the vial. Conversely, the top of the sample (right side of the graph) shows a local increase in the BS signal as oil particles move toward this region of the vial, producing a creaming event. In addition, no global change in the BS signal is observed as the middle of the sample remains relatively static, indicating that a flocculation or global particle size increase is not observed.

While individual kinetics can easily be plotted to investigate the stability of particular regions in the sample, it is useful to quickly analyze the global stability of the system by utilizing a one-click function in the device software called the Turbiscan Stability Index (TSI). This is an algorithm-based calculation that detects the magnitude of the BS signal variance that is seen from scan to scan throughout the experiment. Essentially, more variance in the scans equates to a larger amount of particle migration and size change, and an overall lower stability. A higher TSI number indicates a more unstable sample.

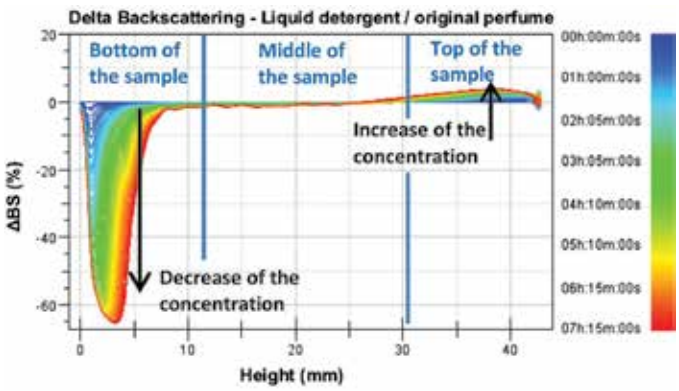
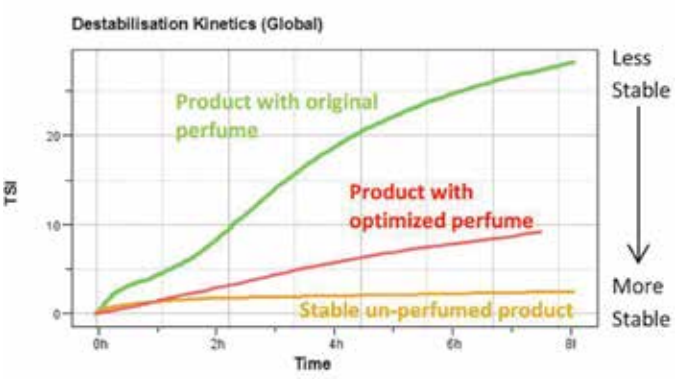
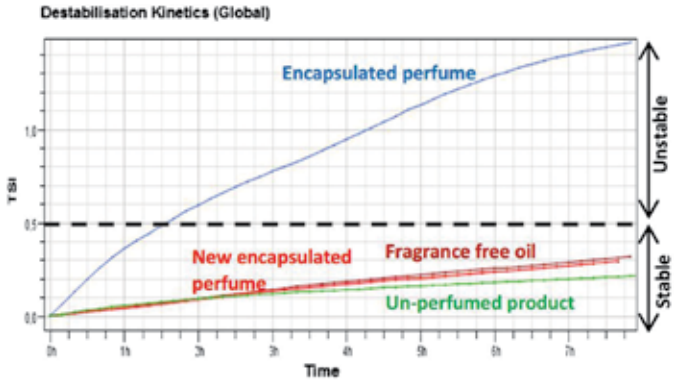


FIG. 1. A typical graph in which the backscattering profile versus the sample height can be seen as the sample ages



Sample	TSI (7 hours)
Original Perfume	26
Optimized perfume	8.4
Un-perfumed	2.3

FIG. 2. Turbiscan Stability Index (TSI) for a stable liquid detergent with no added perfume, modified with an original perfume, and modified with an optimized perfume component



Sample	TSI (7 hours)	TSI (1 hour)
Encapsulated perfume	1.4	0.38
New Encapsulated perf.	0.3	0.07
Fragrance free oil	0.3	0.07
Un perfumed product	0.2	0.07

FIG. 3. Turbiscan Stability Index (TSI) for an unscented liquid fabric softener and the same fabric softener with an encapsulated perfume, an alternate perfume, and a fragrance-free oil

In Figure 2, the TSI plots of the three samples are vastly different after the 7-hour experiment. While the stable, un-perfumed emulsion shows a TSI of 2.3, the original perfume is much higher at 26. At this point, the formulator can go back and change the perfume component to achieve a greater stability, all at the cost of only spending 7 more hours of analysis. It can be seen that the optimized perfume greatly enhances the stability of the system (TSI = 8.4).

In the second experiment, a liquid fabric softener is analyzed in a similar manner in which 20 mL of the generated emulsions are scanned for 7 hours at only 37°C. After 7 hours, the TSI plots shown below in Figure 3 are analyzed to show how the formulation with an encapsulated perfume greatly underperforms that of an alternate (new) perfume and of a system with a fragrance-free oil. Compared to the un-perfumed product, the addition of new perfume and addition of the fragrance-free oil show very similar stabilities. It should be noted that the "failure" formulation is observed to have fat droplets that can visually be observed creaming to the surface after 3 days of analysis, whereas the Turbiscan™ detects this is less than 7 hours.

Overall, we have shown that Turbiscan™ technology is a superior replacement for visual and manual stability tests. The quantitative, high-resolution profiles that are obtained allow for identification of individual destabilization kinetics of the product and also provide the ability to quantify such kinetics whether it be sedimentation of solid particles, aggregation of proteins, or migration and coalescence of fat and oil droplets. Coupled with an extremely accelerated testing timeframe, the technology has been shown to be a powerful tool in the stability analysis of emulsions, dispersions, and foams. Higher quality products can arise from the insight provided by the Turbiscan™ into the specific failures that a formulation may be experiencing and allowing the recipe to be fine-tuned. It remains a useful tool in the R&D lab, where its presence continues to grow.

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Visualizing the oilseed lipidome

Drew Sturtevant, Mina Aziz, and Kent D. Chapman

Lipidomics analyses have become relatively routine in the evaluation of lipid metabolites from oilseeds. While profiles of lipids in seed extracts give detailed measurements of the amounts and types of lipids present, the information about where those lipids were located in the seed is lost with sample preparation and solvent extraction. Now, mass spectrometry imaging allows for the detailed identification and localization of lipids and other metabolites directly on tissue specimens. Application of this technology to oilseeds has revealed that lipid metabolites are localized in a heterogeneous manner in seed tissues (Sturtevant, *et al.*, 2016). In other words, lipid metabolites are distributed unequally in different tissues of the seed, pointing to spatial complexity in lipid metabolism that was not previously known. Although several approaches have been developed to image lipid metabolites by mass spectrometry (MS), one of the most prevalent is by matrix assisted laser desorption/ionization (MALDI) followed by high resolution MS. Lipids are particularly well-suited for this technique because they ionize in a predictable manner, and extensive databases are available for identification.

- **Mass spectrometry imaging results indicate that heterogeneity of lipid metabolites (and metabolic pathways) in seed tissues is a widespread phenomenon.**
- **It is important to consider endogenous heterogeneity to help guide metabolic engineering strategies.**
- **Lipid metabolite localizations suggest that there is much more to learn about the biochemistry, cell biology, and functions of lipid metabolites from a spatial perspective.**

The overall workflow of MALDI-MS imaging for cotton seeds is shown in Figure 1 (page 22). Thin tissue sections are collected on slides from frozen specimens, which are then coated with a chemical matrix, typically either aromatic organic compounds or metal nanoparticles, to promote ionization. A laser is rasterized over the specimen at 10-35 micron steps, and mass spectra are obtained at each spot. Software is then used to construct MS-images based of the *m/z* and ion intensity of each metabolite at each of the sampled locations, and MS databases (e.g., www.lipidmaps.org) enable the identification of specific lipid metabolites. In this way, detailed maps of the chemical organizations of each lipid molecular species are generated and visualized as false color images (Sturtevant *et al.*, 2016). Using this methodology, it is feasible to obtain a spatial view of the oilseed lipidome, especially in the context of the molecular species of triacylglycerols (TAG) and phosphatidylcholines (PC).

The distributions of TAGs in oilseeds have revealed remarkable heterogeneity among different seed tissues. This heterogeneity is widespread and results from differences in molecular species that are located in different seed tissues (cotyledons, hypocotyl, radicle, and endosperm). Although the compositions of seed oils vary in different plant species, heterogeneity has been observed in all oilseeds examined includ-

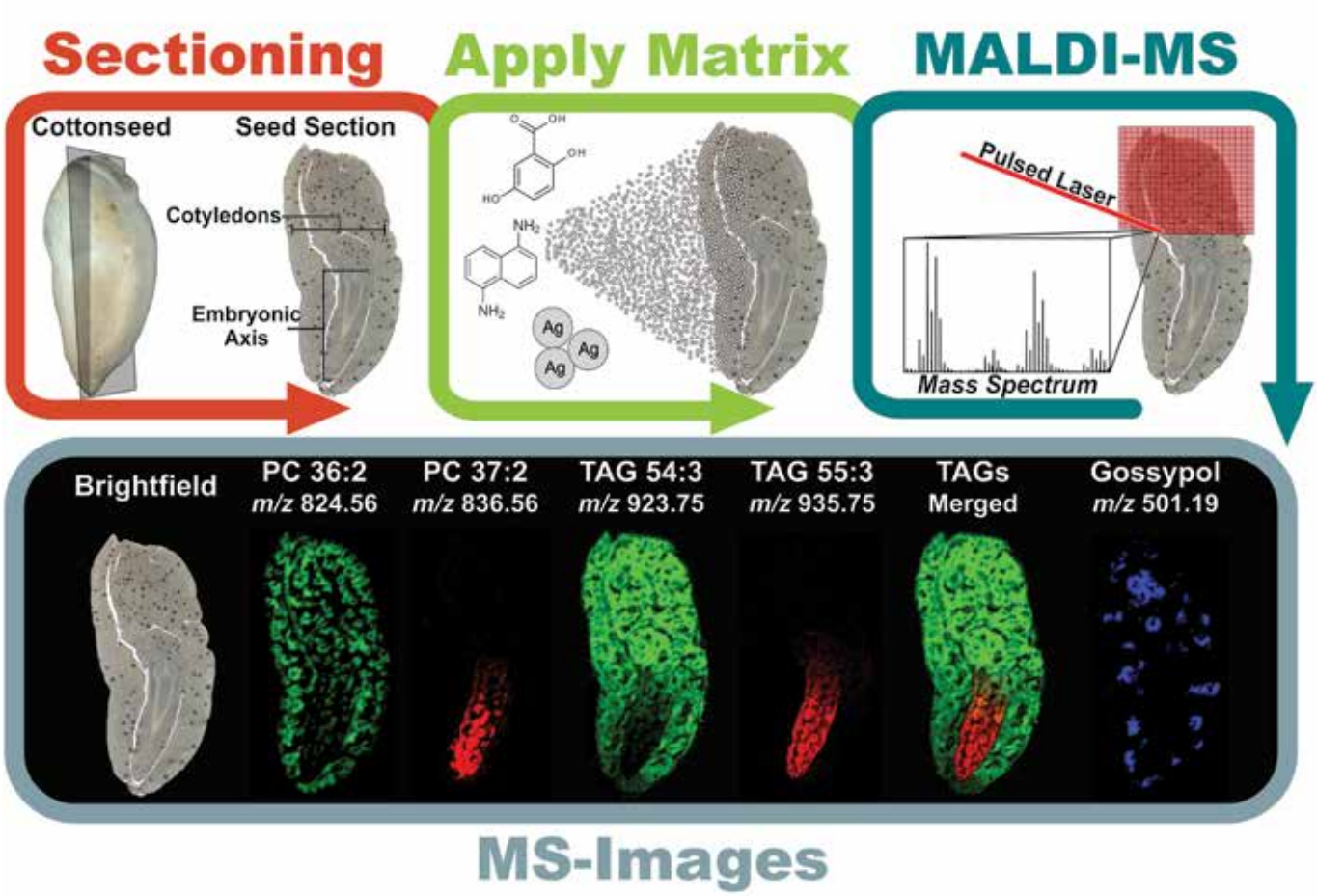

FIG. 1. Work flow for MALDI-MS imaging of oilseeds. Seeds are cryosectioned, coated with a chemical or silver nanoparticle matrix, and a laser is scanned over the tissue at 10 micron steps. Ions from the tissue are ablated from the surface, and a mass spectrum is collected at each location by a high-resolution mass spectrometer. Metabolites are identified in each spectrum using MS databases, and computational programs reconstruct a 2-D image in false color representing the location and relative intensity of each metabolite in the tissue specimen. In this example, two phosphatidylcholine (PC) and two triacylglycerol (TAG) molecular species are shown with different locations in the cotton embryo. Lipid metabolites are shown by their m/z and by their total number of acyl carbons: number of double bonds. Gossypol, a secondary metabolite in cottonseed, is located in discrete gland cells and is also shown as an MS image for comparison. (Sturtevant, *et al.*, 2016.)

ing cotton, castor, camelina, rapeseed, soybean, and the model plant *Arabidopsis*. Similarly, the distributions of phosphatidylcholine (PC) molecular species are also unequally distributed across seed tissues. As the acyl-groups of PC can be incorporated into TAG through various metabolic pathways, a spatial precursor-product metabolic relationship exists between these two lipid classes. Hence, PC precursor molecules of TAG will often localize to the same tissues as their TAG products (Fig. 2). This complex picture of heterogeneous metabolism in seeds was previously under-appreciated, and with the many examples now emerging from MS imaging studies, new questions are arising from both the basic seed science and the biotechnology sectors.

TAG heterogeneity is apparent for common as well as more unusual fatty acids in oilseeds. For one example, in Pima cottonseed, the distributions of two different molecular species of TAGs in cotton embryos are shown in Figure 1. The first, TAG 54:3 contains three oleic acids (C18:1) and is distributed mostly in the cotyledons, and the second TAG 55:3 contains one cyclic fatty acid (sterculic acid) and two oleic acids (Cyc19:1/18:1/18:1) and is distributed almost exclusively in the embryonic axis. The corresponding precursor PC species, 18:1/18:1 PC and 18:1/Cyc19:1 PC, respectively, show a heterogeneous distribution similar to the product TAGs, indicating a metabolic spatial separation for the enzymes involved in the synthesis and assembly of these two different TAGs (Sturtevant, *et al.*, 2017). In another example of heterogeneity (in camelina seeds), the TAGs containing one 20:1 fatty acid were enriched in cotyledonary tissues, whereas those containing two or more 18:2 fatty acids were enriched in the embryonic axis (Horn, *et al.*, 2013). Again, these results suggest that the enzymatic machinery for assembling different molecular species of TAGs is different between the embryonic axis and cotyledon tissues. While the specifics of distribution patterns vary from plant species to plant species, in all cases where it has been examined there is a non-uniform distribution of lipid metabolites across different seed tissues. This kind of spatial information goes entirely unnoticed in conventional analyses of lipid extracts from whole seeds, and suggests that the compartmentalization of oil synthesis in seeds is more com-

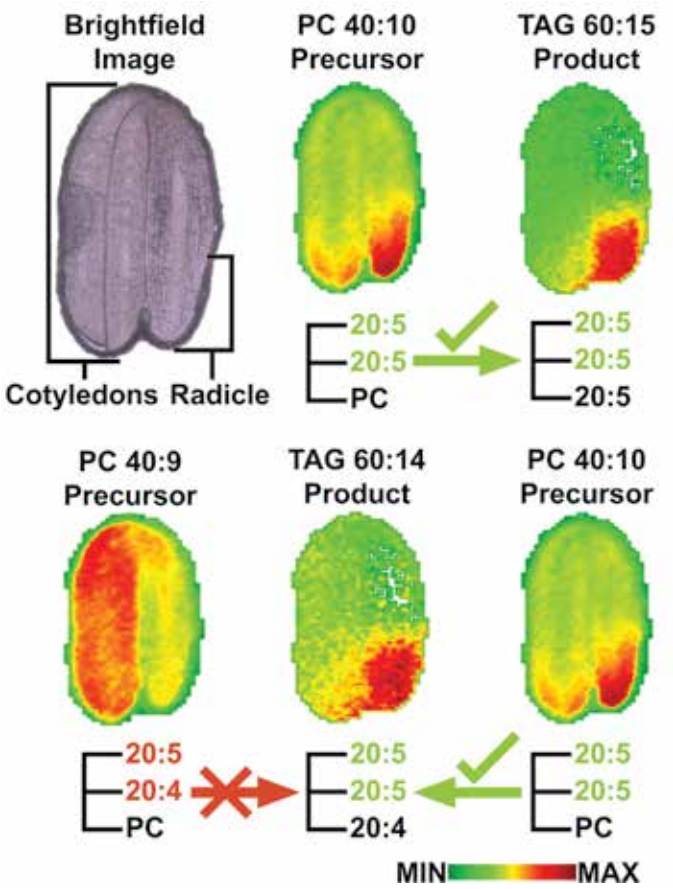

FIG. 2. MS images of Camelina embryos showing the location of engineered oils with eicosapentaenoic acid (EPA, 20:5) incorporated into triacylglycerol (TAG-- 3 EPA, 60:15; 2 EPA, 60:14) and the corresponding distributions of potential phosphatidylcholine (PC) metabolic precursors. This figure illustrates how MS imaging can help determine precursor product relationships for seed oil biosynthesis and also emphasizes that engineered fatty acids may be distributed in an unexpected heterogeneous manner (Usher, *et al.*, 2017).

plex than previously appreciated. There are a few metabolic explanations of how this heterogeneity arises in some seeds (Marmon, *et al.*, 2017), but in many cases the regulation of lipid distribution remains poorly understood, and will be the subject of much future research.

As the spatial heterogeneity of lipid metabolism in oilseeds becomes increasingly apparent, one important question arises, especially from a metabolic engineering standpoint: Can engineering lipid metabolism on top of endogenous heterogeneity have unforeseen consequences on the accumulation of engineered fatty acids? Indeed, recently it was shown that efforts to engineer long-chain polyunsaturated fatty acids (LC-PUFAs) into camelina seeds were impacted by pre-existing endogenous compartmentalization (Usher *et al.*, 2017). In camelina lines engineered to produce eicosapentaenoic acid (EPA, 20:5 fatty acid), TAGs containing two or three EPA acyl chains (e.g. TAG 60:14 (20:5/20:5/20:4) and TAG 60:15 (20:5/20:5/20:5)), and their corresponding PC precursors, were significantly enriched in the radicle tissues of the embryonic axis (Fig. 2). This pattern of distribution indicates that the

endogenous pathways for TAG assembly that incorporated LC-PUFAs may be different and more efficient in the radicle tissues, than in cotyledonary tissues. Visualizing the precursor-product relationship between PC and TAG can also help address potential bottlenecks in metabolic engineering. In the case of the EPA engineered lines, TAG 60:14 has two potential precursor PC species, PC 40:9 and PC 40:10. However only PC 40:10 shares the same localization pattern as the corresponding TAG, making it the likely precursor PC. Uncovering the specific mechanisms that facilitate accumulation of LC-PUFAs in radicle tissues of transgenic embryos might inform next-generation strategies to elevate the LC-PUFA content throughout the embryo and dramatically increase yields of LC-PUFAs in oilseeds. Only by examining the spatial distribution of these LC-PUFAs in engineered seeds did the heterogeneity of TAG assembly become apparent as an important factor for consideration for metabolic engineering strategies.

The concept that lipid metabolites and the pathways leading to their synthesis are different in different tissues of oilseeds indicates that seed oil accumulation is more complex than previously recognized. Not only do breeders and biotechnologists need to contend with multiple pathways that lead to TAG assembly, but also multiple compartments where these pathways predominate. This represents new challenges and interesting biological questions about the significance of metabolic heterogeneity in seeds that will stimulate new avenues for research in the future. This may require the transcriptional profiling of different seed tissues to identify predominant pathways in different seed tissues, or perhaps the identification of additional promoters that can influence the tissue-specific accumulation of desired lipids. In the end, it was the ability to image lipid metabolites *in situ* that raised these considerations, and likely, it will be this mass spectrometry imaging technology that will be important, at least in part, in addressing these exciting opportunities in the future.

Drew Sturtevant is a Ph.D. candidate in biochemistry and molecular biology at the BioDiscovery Institute, University of North Texas. His dissertation work is focused on studying the spatial organization of lipids in oilseeds and the genetic and molecular machinery responsible for these distributions. He can be contacted at drewsturtevant@my.unt.edu.

Further reading

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Mina Aziz is a postdoctoral research associate at the BioDiscovery Institute at the University of North Texas. He is currently studying N-Acylethanolamine (NAE) signaling, with special emphasis on the structural aspects of the key enzyme that regulates NAE function in plants. Another area of research includes the application of MALDI-MS imaging to study the spatial distribution of lipids and secondary metabolites in plant tissues. He can be contacted at mina.aziz@unt.edu.

Kent D. Chapman is Regents Professor of Biochemistry and Director of the BioDiscovery Institute at the University of North Texas. Chapman has worked in the area of plant lipid metabolism for more than 30 years contributing to concepts of storage lipid accumulation and lipid signaling and their evolutionary conservation in plants. He can be contacted at chapman@unt.edu

AOCS MEETING WATCH

May 6–9, 2018. AOCS Annual Meeting & Expo, Minneapolis Convention Center, Minneapolis, Minnesota, USA.

September 6, 2018. JOCS AOCS Joint Symposium, Kobe Gakuin University, Arise Campus, Kobe, Japan.

September 9–11, 2018. Canadian Section of the AOCS (CAOCS) to host 26th CAOCS Canadian Lipid & Bioresource Conference, Saskatoon, Saskatchewan, Canada.

October 28–31, 2018. Fabric and Home Care World Conference, Boca Raton Resort & Club, Boca Raton, Florida, USA.

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Recent developments in foodomics

Alejandro Cifuentes

- **Foodomics studies the food and nutrition domains through the application and integration of advanced omics technologies to improve consumers' well-being, health, and confidence.**
- **Many works have already shown the tremendous possibilities of foodomics to advance food science investigations, including those related to speeding up the resolution of food safety issues, improving food quality and food traceability, and understanding at a molecular level the bioactivity of food and food ingredients in our bodies.**
- **A foodome is the pool of all compounds present in a food sample and/or in a biological system interacting with the investigated food at a given time.**
- **Omics techniques such as genomics, transcriptomics, proteomics, metabolomics, nutrigenetics, nutrigenomics, and microbiomics, among others, are essential tools employed in foodomics.**

As a general consensus, food safety is always the first priority in food science. However, globalization has promoted the fast movement worldwide of food and related raw materials. This has, in turn, pushed researchers and supervisory laboratories to face a new challenge: the threat of global contamination episodes. Consequently, ensuring the safety, quality, and traceability of food has never been more complicated or important than it is today.

Another general trend in modern food science is the connection between food and health. Indeed, food is now considered not only a source of energy but also an affordable way to improve health and/or to prevent future illness (e.g., through the so-called functional foods). However, to understand and demonstrate scientifically the health effects of food and food ingredients is not an easy task. Some of the obstacles include: i) the food complexity; ii) the huge natural variability in foodstuffs, which contain a vast number of different nutrients and bioactive food compounds at a wide range of concentrations; iii) the bioavailability and transformation of these compounds in the human gastrointestinal tract, which multiplies this complexity; and iv) the numerous targets, each with different affinities and specificities within the human body.

The complexity can explain, for instance, the decades-long debate over the relationship between macronutrients and cardiovascular disease and mortality. This debate was stirred up by a cohort study of the dietary intakes of 135,335 individuals with a median follow up of 7.4 years in which the authors concluded that high-carbohydrate intake was associated with a higher risk of total mortality, whereas total fat and individual types of fat were related to lower total mortality. Moreover, total fat and types of fat were not associated with cardiovascular disease, myocardial infarction, or cardiovascular disease mortality, whereas saturated fat had an inverse association with stroke. In summary, the authors concluded that "global dietary guidelines should be reconsidered in light of these findings." Their work, published in the prestigious medical journal, *The Lancet*, in 2017, is a good example of how little we know about many basic aspects related to the long-term effect of foods and ingredients on our health. It also highlights the long way to go before the goal of so-called personalized nutrition can be reached.

On the other hand, in the current postgenomic era, we rely on analytical instruments and methodological developments that were

unimaginable a few decades ago. Consequently, researchers in food science and nutrition are being pushed to move from classical methodologies to more advanced strategies, usually borrowing well-established methods from clinical, pharmacological, and/or biotechnology research. In this context, foodomics—a discipline that studies the food and nutrition domains through the application and integration of advanced omics technologies to improve consumers' well-being, health, and confidence—offers a new way to investigate food and nutrition that would have been considered unapproachable a few years ago.

In doing so, foodomics incorporates multiple concepts and technologies, such as genomics, epigenetics, transcriptomics, proteomics, metabolomics, nutrigenetics, nutrigenomics, and microbiomics. The development and applications of these high-throughput technologies, along with new bioinformatic methods, are opening new possibilities to solve the above-mentioned important challenges. The expected outputs from this field are anticipated to be crucial, as diet is one of the most modifiable factors affecting health. As a result, foodomics is attracting more and more attention (Fig. 1) from research institutions, agencies, food industries, regulatory laboratories, and scientific instrumentation manufacturers due to increasing government food regulations and consumer demand.

Figure 2 shows a schematic illustration of omics tools and applications of foodomics. Namely, transcriptomics, proteomics, and metabolomics approaches have been mainly developed in our laboratory to study how foods (including

FIG. 1. Some representative covers of journals and books devoted to foodomics

functional ingredients) interact with the genome and the subsequent modifications that these interactions can generate at a proteome and metabolome level. Based on this strategy, we have investigated the antiproliferative activity of food ingredients against leukemia and colon cancer (*vide infra*). Also, we have applied omics approaches to investigate non-intended modifications in transgenic foods, and to study issues related to food quality and safety as a whole. Moreover, we have developed new environmentally friendly extraction processes

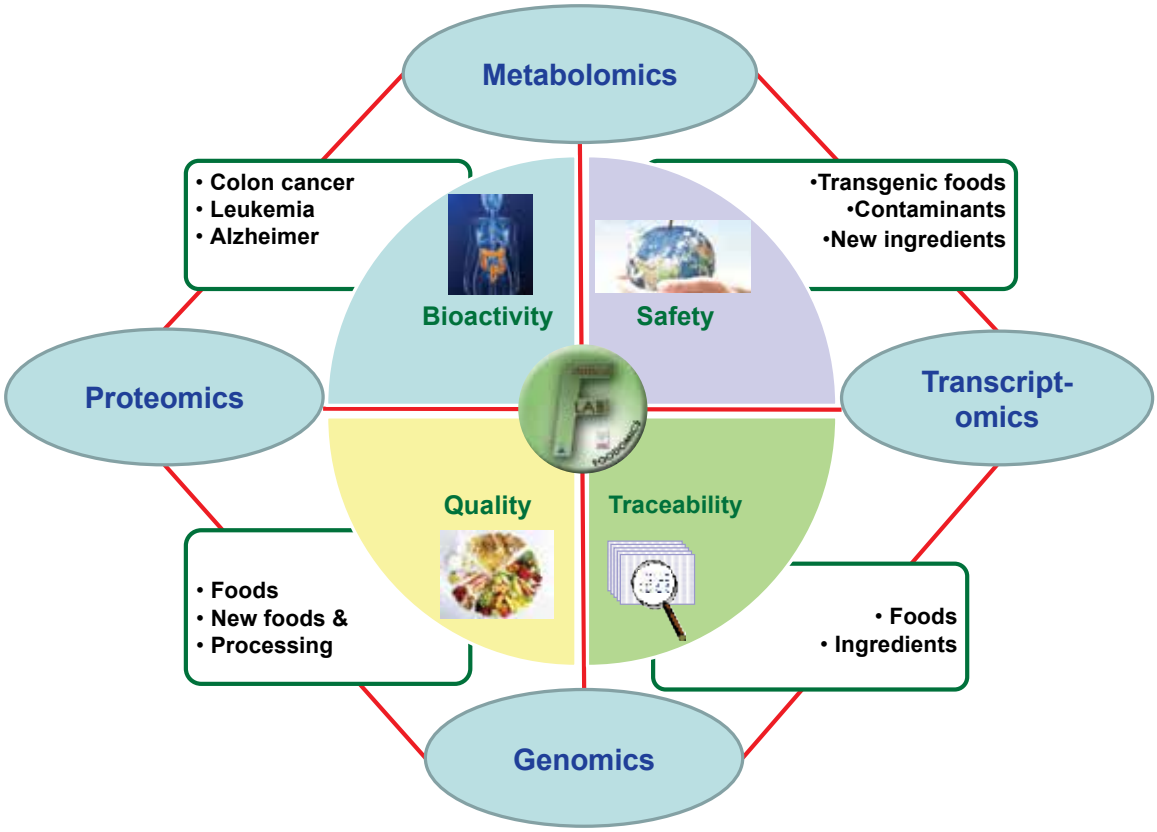

FIG. 2. Schematic of main omic tools and applications of foodomics in our lab

(green processes) to discover and obtain new bioactive compounds from food by-products and natural sources, including algae and microalgae. Last but not least, we have developed advanced analytical methods, including multidimensional methodologies, based on two-dimensional comprehensive liquid chromatography (LCxLC) coupled to tandem mass spectrometry (MS/MS), to chemically characterize the complex composition of the natural extracts. Recently, we have started a new research project to investigate the effect of food ingredients on the development of Alzheimer disease.

Although the outcomes from using a global foodomics strategy are significant, so far there have been very few published papers in which results from the three expression levels (transcriptomics, proteomics, and metabolomics) are simultaneously presented and merged to investigate the bioactivity of a given food or ingredient. Applying data interpretation and integration to such complex systems is not straightforward, and has been identified as one of the main bottlenecks. Consequently, the number of studies on the effect of specific nutrients, or diet, on the transcriptome-proteome-metabolome of organisms, tissues, or cells, is still rather limited due to the number of review papers on this topic being higher than the number of research papers.

Without a common point to begin, you can imagine the huge effort that went into developing the whole analytical approach. Figure 3 shows the typical workflow that is needed to carry out these investigations in our Foodomics Laboratory. Using that work-flow together with two human leukemia cell lines (normal leukemia cell line vs. resistant leukemia cell line), we observed differences in the transcriptional induction of a

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number of genes encoding phase II detoxifying and antioxidant genes, as well as differences in the metabolic profiles induced by a natural extract from rosemary.

In addition, the bioinformatics analysis predicted the inhibition of MYC transcription factor, which could explain the observed antiproliferative effect against the resistant leukemia cells. Some differences in the gene expression patterns could be also observed when human colon cancer HT-29 and SW480 cell lines were treated with the same extract. In addition, a deeper study demonstrated that most of the changes induced by the rosemary extract and one of its main components, carnosic acid, were orchestrated by unfolded protein response and triggered by endoplasmic reticulum stress, while the second most abundant compound in the extract (carnosol), directly inhibits chymotrypsin-like activity of the 20S proteasome. Moreover, this antiproliferative activity against colon cancer cells was corroborated *in vivo* using athymic mice inoculated with colon cancer cells, concluding that the daily administration of the polyphenol-enriched rosemary extract reduces the progression of colorectal cancer *in vivo,* with the subsequent deregulation of 74 proteins.

Although we know these works are only the first steps in a very long race, the mentioned works are a good demon-

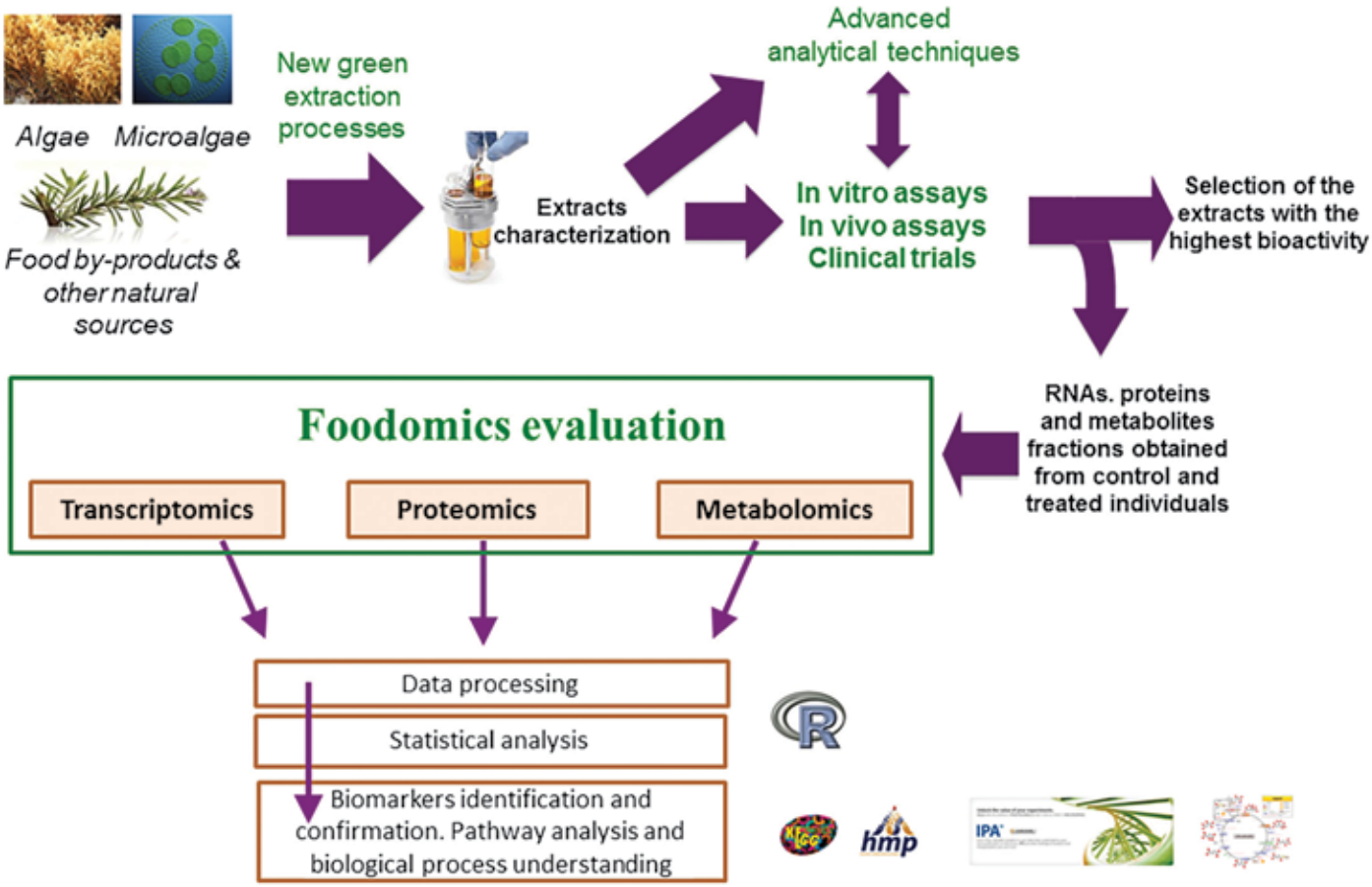

FIG. 3. Typical work-flow used in foodomics to investigate the bioactivity of new functional ingredients and foods

stration of how foodomics can help to investigate and solve crucial topics in food science and nutrition from both a short- and long-term perspective. It is expected that the new omics technologies combined with systems biology, as proposed by foodomics, can lead food science and nutrition into a new era.

Alejandro Cifuentes is a full research professor at the National Research Council of Spain (CSIC) in Madrid, Head of the Laboratory of Foodomics and Director of the Metabolomics Platform (International Excellence Campus CSIC + University Autonoma of Madrid). Alejandro's activity includes advanced analytical methods development for foodomics (including transcriptomics, proteomics, and metabolomics), food quality and safety, as well as isolation and characterization of natural bioactive compounds and their effect on human health. He has received several national and international awards, is member of the editorial board of 15 international journals and Editor of Trends in Analytical Chemistry, Electrophoresis, and American Journal of Advanced Food Science. He has published more than 250 SCI papers, 20 books and book chapters, and 9 patents. His h index is 64 (January 2018), and his works have received more than 13,000 citations. Alejandro defined for the first time in an SCI journal the new discipline of foodomics. He can be contacted at a.cifuentes@csic.es.

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To get more information about foodomics or to get an update on our activity you can visit our web-page: http://www.cial.uam-csic.es/pagperso/foodomics/index_en.html.

High-oleic soybeans get the global green light

Olio is an Inform *column that highlights research, issues, trends, and technologies of interest to the oils and fats community.*

Laura Cassiday

More than seven years after obtaining regulatory approval in the United States, DuPont Pioneer's Plenish high-oleic soybeans have finally achieved European Union approval, paving the way for increased worldwide production and consumption of high-oleic soybean oil. On December 22, 2017, the European Commission authorized the use of the stacked traits found in Plenish for food and feed. The decision surmounts the final regulatory hurdle to global trade of the high-oleic soybeans and soybean oil. Plenish will join Monsanto's Vistive Gold high-oleic soybeans, which achieved final global approval with China's import authorization in June 2017, on the world market in 2018.

"Obtaining full global regulatory approval is key for the entire soybean value chain, as it will boost Plenish and Vistive Gold soybean acreage and allow them to be grown in more diverse geographies," said Richard Galloway, QUALISOY oils expert. "This will allow the industry to reach its goal of planting 16 million high-oleic soybean acres, resulting in 9 billion pounds of high-oleic soybean oil available for global consumption. In the coming decade, high-oleic soybeans will become the fourth-largest grain and oilseed crop in the U.S."

According to the United Soybean Board, in 2017 US farmers grew high-oleic soybeans on 625,000 acres in 13 states. Farmers who signed contracts with Plenish processors—ADM, AGP, Bunge, CHS, and Perdue Agribusiness—received premiums ranging from 40 to 60 cents per bushel above the commodity price (Unglesbee, E., https://tinyurl.com/Agfax-high-oleic, 2017). Farmers must take special precautions to keep high-oleic soybeans segregated from regular soybeans (a process called identity preservation), including separate storage bins and cleaning combines before harvesting.

Global regulatory approval secures a reliable supply of high-oleic soybean oil for the food industry. High-oleic soybean oil is attractive to food manufacturers for several reasons. High-oleic oil contains 0 grams of trans fat per serving and less saturated fat than conventional soybean oil. In addition, high-oleic soybean oil boasts a longer shelf life, a neutral flavor, enhanced functionality, extended fry life, and reduced buildup of polymers on food processing equipment. Non-food applications include synthetic motor oils and automotive lubricants.

High-oleic soybean oils help fill the void created when the US Food and Drug Administration mandated the removal of partially hydrogenated oils from the food supply by June 2018. According to functionality testing by QUALISOY, high-oleic soybean shortening produced baked goods that were the most similar to products made with partially hydrogenated oils (https://tinyurl.com/QUALISOY-brochure, 2017). In addition, the oil stability index (OSI) of high-oleic soybean oil is greater than 20 hours, compared with 8 hours for conventional soybean oil.

The fatty acid profile of Plenish includes 75% oleic acid, 12% saturated fatty acids, 8% linoleic acid, 2% linolenic acid, and trace amounts of other fatty acids (Fig. 1). Vistive Gold contains 72% oleic acid, 6% saturated fatty acids, 16% linoleic acid, and 3% linolenic acid. Both are very similar in fatty acid composition to olive oil, with 75% oleic acid, 15% saturated fatty acids, 9% linoleic acid, and 1% linolenic acid. To generate Plenish soybeans, scientists blocked the *FAD2-1* gene, which encodes a fatty acid desaturase that converts oleic acid (a monounsaturated fatty acid) to linoleic acid (a polyunsaturated fatty acid). The result is a more stable soybean oil, without the need for partial dehydrogenation and the resulting trans fats.

Plenish soybeans are based on Roundup Ready 1 germplasm, the patent for which expired in 2015. In 2009, Monsanto launched the Roundup Ready 2 trait based on next-generation technology that offers the same glyphosate resistance but up to 4 bushels per acre higher yield. Roundup Ready 2 XTend soybeans feature additional resistance to the

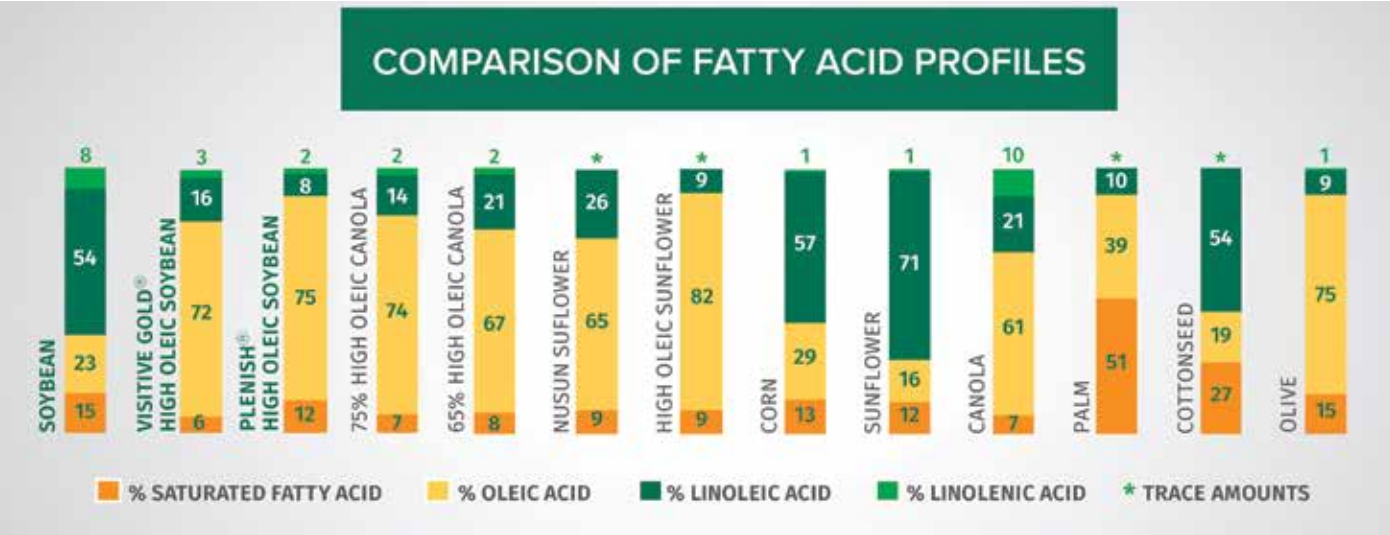

FIG. 1. Comparison of fatty acid profiles in selected vegetable oils. Credit: QUALISOY

herbicide dicamba. DuPont Pioneer is planning to offer Plenish soybeans with Roundup Ready 2 XTend genetics within the next few years (Unglesbee, E., https://tinyurl.com/Agfax-high-oleic, 2017). Vistive Gold soybeans already have the Roundup Ready 2 trait, but Monsanto plans to add the Xtend trait in 2019.

To gain EU approval, the European Commission had to individually study and approve the "stacked" high-oleic and glyphosate resistance traits. Although high-oleic soybean oil must be labeled as genetically modified in the European Union, Galloway does not see this requirement as a barrier to trade, saying that European manufacturers have already shown interest (Michail, N., https://tinyurl.com/FoodNav-Plenish, 2018). Nathalie Lecocq, secretary general of FEDIOL, the EU vegetable oil and protein meal association, says that although Plenish is not targeted at the European market (where soybean oil is not commonly used), the EU authorization will help prevent trade disruptions caused by the presence of trace amounts of high-oleic soybeans or soybean oil in regular soybean or soybean oil supplies.

The health benefits of high-oleic soybean oil compared with regular soybean and other vegetable oils are still being debated. A widely publicized 2017 study compared health effects in mice of a high-fat diet containing either Plenish, conventional soybean oil, olive oil, or coconut oil (Deol, P. *et al.*, http://dx.doi.org/10.1038/s41598-017-12624-9). The researchers found that although Plenish caused less obesity and insulin resistance than conventional soybean oil, it did not lead to a lower incidence of diabetes or fatty liver. All three oils raised cholesterol levels in the blood. Interestingly, coconut oil (which consists of mostly saturated fat) caused fewer negative metabolic effects than any of the other oils tested.

Surprisingly, olive oil and Plenish, but not regular soybean oil or coconut oil, induced hepatomegaly and liver dysfunction in the mice. "[These results] show that the genetic modification of soybean oil may induce detrimental health effects in terms of liver function even though it induces less obesity and insulin resistance than conventional soybean oil," the researchers write (Deol, P. *et al.*, http://dx.doi.org/10.1038/s41598-017-12624-9). However, Plenish had very similar metabolic effects, and a similar fatty acid profile, to olive oil, which is widely touted as being healthful. Rodent studies, especially those involving high-fat diets, can be difficult to translate to humans. With increased high-oleic soybean oil availability worldwide, more randomized controlled trials with human subjects will likely be conducted.

Information

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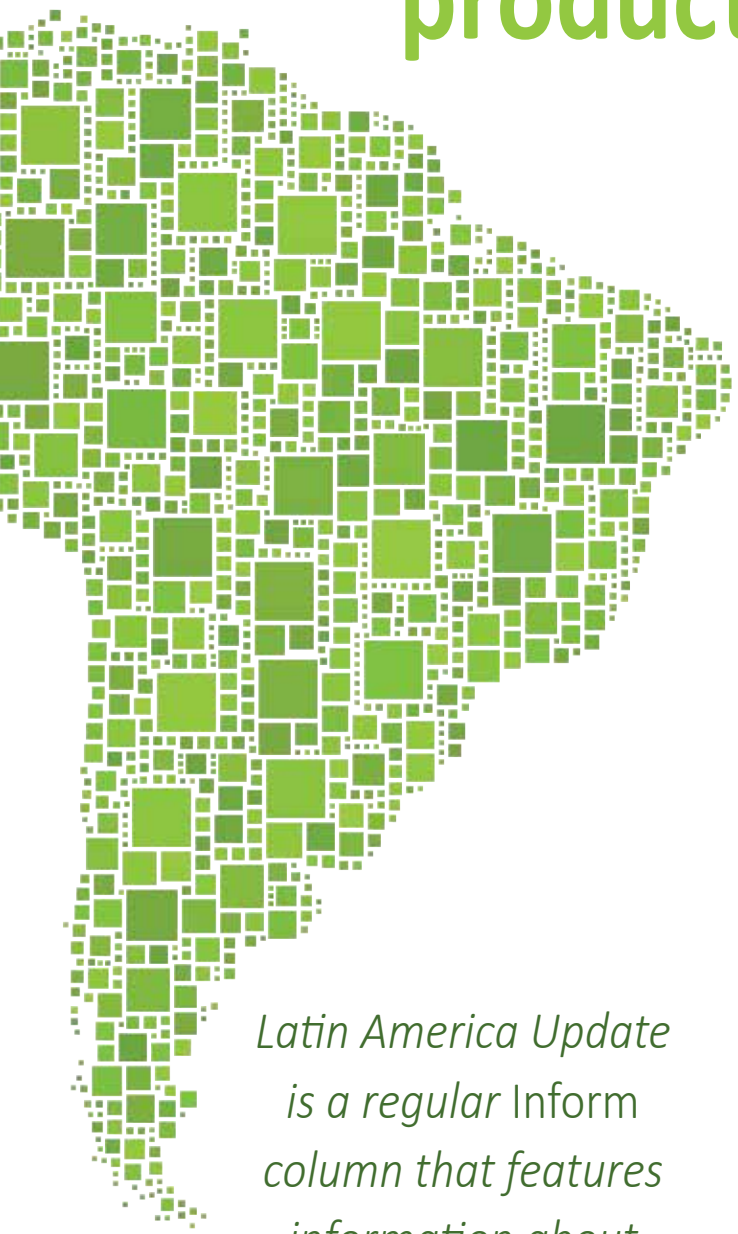
Olio is produced by Inform*'s associate editor, Laura Cassiday. She can be contacted at laura.cassiday@aocs.org.*

2017 Sustainability and its role in food and beverages product launches



Latin America Update is a regular Inform *column that features information about fats, oils, and related materials in that region.*

Leslie Kleiner

On its website, the US Environmental Protection Agency (EPA) defines the pursuit of sustainability as a commitment to present and future generations, which is enacted through the National Environmental Policy Act of 1969. This act declares a national policy "to create and maintain conditions under which humans and nature can exist in productive harmony, that permit fulfilling the social, economic and other requirements of present and future generations" [1]. Since this 1969 enactment, sustainability awareness has largely increased among consumers. To understand sustainability in food in the United States and LATAM, I consulted a recent report from global market intelligence agency Mintel. The report, entitled, "Sustainability," was written by Emma Schofield, Global Food Science Analyst, and published in August 2017. The following Q&A is extracted from this report.

Q: Which of the United Nation's Sustainable Development Goals from the 2030 Agenda for Sustainable Development are being implemented (these apply universally to all countries)?

In 2016, the United Nations brought forward 17 goals to achieve sustainability through the promotion and efforts of each individual country. Most of these goals pertain to ending all forms of poverty, fighting inequality, and addressing climate change (Fig. 1).

FIG. 1. United Nations: Sustainable development goals.
Image extracted from http://www.un.org/sustainabledevelopment/sustainable-development-goals/

Q: Globally, which types of sustainability claims in food and beverages are predominant?

Data from July 2016 to July 2017 shows that 15% of product launches pertaining to food, beverages, pet food, and healthcare, had claims regarding environmentally friendly packaging. For the same time period, 6% of product launches had claims referring to the product as "environmentally friendly," while 3%, 2%, 1%, and 0.2% of product launches had claims regarding ethically humane, ethical treatment of animals, charity contributions, and carbon-neutral efforts, respectively. All of these claims are on the rise, and have experienced an upward trend since 2012.

Q: Which are the main sustainability concerns in the United States? Are they similar to those in Europe? How do they compare for the LATAM region?

In the United States, the iGeneration (aka, Generation Z: those born after 1994, and who have grown up with smartphones [2]) has its own specific concerns. Data collected on consumers aged 18 to 22 (March 2017) shows that this generation is primarily concerned about the economy (67%), climate change (55%), terrorism (54%), and poverty (53%). As these consumers, along with Millennials, enforce their views through purchasing power, sustainability claims (e.g., ethical treatment of animals, fair wages, environmentally friendly) are also expected to grow. US data from August 2011 to July 2017, shows that 20% of food and beverage product launches had environmentally friendly packaging claims. This was followed by environmentally friendly product claims (slightly above 5%). During the same time period in Europe, 14% of product launches in the same category had environmentally friendly packaging. However, countries like the UK, Ireland, Australia, and France had more sustainability claims (41%, 40%, 35%, and 31%, respectively). In LATAM, for the same category and time period, 18% of new product launches had environmentally friendly packaging, followed by 35% environmentally friendly product claims. Brazilian consumers, in particular, showed concerns for the environment. This is illustrated by data (July 2016) on the motivations of Brazilian consumers to purchase sustainable products: 61% of consumers purchase because "it is good for the environment." Other motivations cited by 54%, 51%, and 49% of consumers, respectively, included, "I do not like to waste," "I feel good when I help preserving the environment," and "it Is the responsible thing to do."

Q: Which ethical claims are most popular in various product categories?

A: Globally, from August 2011 to July 2017, hot beverages and chocolate accounted for most claims regarding ethical treatment of humans, as illustrated by Fair Trade claims. More specifically, ethically human claims were conspicuous on hot beverages (14%), chocolate confectionery (13%), ready-to-drink meals (RTDs) (7%), sweeteners and sugar (5%), and desserts and ice creams (3%). Environmentally friendly product claims appeared on hot beverages (16%); chocolate confectionery (12%); sweeteners and sugar (8%); processed fish, meat, and eggs (8%); and breakfast cereals (7%). Finally, the largest incidence of claims was related to packaging: carbonated and soft drinks (39%), water (38%), juice drinks (33%), breakfast cereals (32%), and RTDs (30%). Other categories of claims, such as ethical treatment of animals, charity, and carbon neutral footprint were also made, but were less common among product launches.

Q: What is the response from the food and beverage industry?

Companies such as Nestlé and Pepsico have shared their goals on sustainability. For example, Nestlé shared its goal to strive for zero impact from its operations by the year 2030, while Pepsico's proposed 2025 agenda is to transform its product portfolio to healthier options, work to achieve positive water impact, and lower carbon emissions, as well as to reduce and eliminate waste. The Pepsico agenda also includes advancing respect for human rights, supporting diversity, and advancing women. Other companies use third-party certifications such as "Marine Stewardship Council (MSC), and the Rainforest Alliance, to convey to consumers their support to sustainability.

Q: Where does palm oil stand in terms of sustainability?

The environmental impact of the palm oil industries continues to be a concern to consumers. "Palm- oil-free" claims in France and Italy are emerging, despite efforts by The Roundtable on Sustainable Palm Oil (RSPO), and its claim to certify 21% palm oil as sustainable.

References

[1] https://www.epa.gov/sustainability/learn-about-sustainability#what

[2] https://hbr.org/2015/05/how-to-market-to-the-igeneration

Latin America Update is produced by Leslie Kleiner, R&D Project Coordinator in Confectionery Applications at Roquette America, Geneva, Illinois, USA, and a contributing editor of *Inform*. She can be reached at LESLIE.KLEINER@roquette.com.

What are the latest trends in the **personal care sector?**

Regulatory Review is a regular column featuring updates on regulatory matters concerning oils- and fats-related industries.

Vanessa Zainzinger

Bans, restrictions, ingredient disclosure: These are the key chemical issues for the personal care industries in Europe, the United States, and the rest of the world..

LOOKING FOR SAFER PRESERVATIVES

Preservatives–or the lack thereof–is a prevalent crisis in the cosmetics sector. Last year stakeholders upped their efforts to both defend existing ingredients from impending restrictions or bans, and to encourage investment in developing new preservation systems. Trends to watch are multifunctional additives and formulations based on organic acids. Swiss multinational Lonza, for example, is future–proofing its portfolio by formulating such preservation options. Meanwhile, US business group the Green Chemistry and Commerce Council (GC3) ran a competition with the aim of identifying promising safe preservation systems for personal care and household products, and to help bring them to market. It closed in August 2017 with 48 submissions for potential new systems, which are currently being evaluated.

ARE MINERAL OIL HYDROCARBONS A HEALTH CONCERN?

Another group of substances to watch are mineral oil hydrocarbons. The European Food Safety Authority, the EU's Scientific Committee for Consumer Safety (SCCS) and the Danish Consumer Council have put pressure on the European Commission to consider whether dietary exposure to certain mineral oils in lip care products is a health concern. Beuc, the European Consumer Organisation, recently urged Brussels to impose a safety limit on mineral oil aromatic hydrocarbons (MOAHs) and mineral oil saturated hydrocarbons (MOSHs). Next year will likely see this discussion progress and could force a response from the European Commission.

MICROPLASTICS UNDER SCRUTINY

Meanwhile, more and more EU member states are mulling, or already implementing, bans on microplastics in cosmetics. Belgium has notified a draft plan to voluntarily phase out microplastics in all consumer products by 2019. The ban would initially apply to cosmetics and toothpaste. France will prohibit rinse–off cosmetics containing microplastics from January , and Sweden has proposed to do the same. The UK government had aimed to ban the manufacture of rinse–off cosmetics containing microbeads by the end of 2017. Whether these initiatives will have a significant effect on the cosmetics industry, however, remains to be seen. According to trade bodies, the sector has already phased out solid microbeads from rinse–off cosmetics voluntarily, ahead of any legislation.

INCREASING TRANSPARENCY

While preparing for a shrinking toolbox of available substances, personal care companies are also keeping an eye on ingredient disclosure trends. Under rising demands for transparency, large corporations and retailers are beginning to make information on the chemicals in their products more easily available to consumers; and personal care is fronting this trend. Unilever US has made available fragrance ingredient information for almost 100 of its products available online and through an app. It has plans to provide consumers with information about specific fragrance ingredients above 0.01% (100 parts per million) present in its personal care products by the end of 2018. Procter & Gamble also recently announced an initiative to reveal all the fragrance ingredients in its products above 0.01%, by the end of 2019. If calls from NGOs are heard, fragrance ingredients could soon be added to product labels, too. California's Assembly has already passed a bill that would require ingredient labelling for professional cosmetics.

Vanessa Zainzinger is the biocides editor at Chemical Watch.

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PATENTS

Method of separating oil

Wang, M.M., *et al.*, Croda, Inc., US9738852, August 22, 2017

A method of separating oil from a composition containing an oil and water emulsion, by adding a separation additive which is a fatty ester of alkoxylated glycerol, and performing at least one oil separation step. The method is particularly suitable for separating corn oil from stillage produced in a corn ethanol mill.

Method of killing bedbug eggs

Smith, K.R., *et al.*, Ecolab USA Inc., US9743661, August 29, 2017

The present invention relates to a method of pre-rinsing laundry and treating other surfaces employing a composition that can kill bedbug eggs. The composition includes a fatty alcohol and an alcohol ethoxylate.

Oil and fat composition that can be used as non-tempering type hard butter

Ohara, A., *et al.*, The Nisshin Oillio Group, Ltd., US9743681, August 29, 2017

To provide an oil and fat composition that can be used as a low-trans fatty acid content, non-lauric, non-tempering type hard butter and an oil-based food product comprising the oil and fat composition that exhibits good heat resistance and melting feeling in the mouth. The oil and fat composition of the present invention is characterized by satisfying the following conditions of (a) to (g): (a) an X3 content is 3 to 20% by weight;(b) a weight ratio of P3/X3 is not less than 0.35;(c) an X2O content is 45 to 80% by weight;(d) a weight ratio of XOX/X2O is 0.20 to 0.65;(e) a weight ratio of PStO/X2O is 0.10 to 0.45;(f) a weight ratio of St2O/X2O is 0.05 to 0.35; and(g) a weight ratio of St/P is not more than 0.80;wherein X represents saturated fatty acid having 14 carbon atoms or more; O represents oleic acid; P represents palmitic acid; and St represents stearic acid.

Fuel lubricity additive

Choo, Y.M., *et al.*, Malaysian Palm Oil Board, US9745531, August 29, 2017

A lubricity additive for fuels with sulphur content of not more than 0.05 wt percent is described herein. The lubricity additive comprises a polyol ester or a mixture of polyol esters derived from C8 -C18 saturated and/or unsaturated fatty acids. The polyol esters are produced by: i) esterification of a C8 -C18 saturated or unsaturated fatty acids, or a mixture thereof, with a polyhydric alcohol; ii) transesterification of an oil or a mixture of oils, with fatty acid composition comprising C8 -C18 saturated and/or unsaturated fatty acids, with a polyhydric alcohol. The preferred fatty acids are unsaturated C18 fatty acids, more particularly, oleic acid whereas the preferred polyhydric alcohol is neopolyol, more particularly, trimethylol propane and its isomers. A fuel composition comprising a major amount of fuel with sulphur content of not more than 0.05 wt percent and a minor amount of the lubricity additive is also described herein. The amount of the lubricity additive is not more than 0.1 wt percent.

Conjugated linoleic acid-rich vegetable oil production using heterogeneous catalysis

Proctor, A., *et al.*, The University of Arkansas, US9752099, September 5, 2017

The invention is directed to CLA-rich vegetable oil production from linoleic rich oils by heterogeneous catalysis. The process produces conjugated PUFA in triglyceride form, preferably at least 20% CLA-rich, by isomerization of a non-conjugated PUFA in vegetable oils using a heterogeneous transition metal catalyst promoted by an organic acid and/or thiol-containing compound. The heterogeneous catalysis isomerization process can use steam/vacuum distillation, hydrogenation unit, and/or deodorization to produce CLA-rich soy oil. After processing, any catalyst residue may be removed by filtration, beaching, deodorizing, adsorbents, or centrifugation to obtain high-quality, CLA-rich oils.

Fine particles coated with lipid membrane

Kato, Y., *et al.*, Kyowa Hakko Kirin Co., Ltd., US9757344, September 12, 2017

A coated fine particle comprisisng: (i) a complex of a drug and a cationic lipid, wherein (a) the drug is a nucleic acid and the complex is obtained by mixing the drug and the cationic lipid in water, wherein the complex has a diameter of 10 nm to 1,000 nm, and (ii) a lipid layer formed of lipid(s), wherein the lipid(s) is selected from phospholipid, glyceroglycolipid, sphingoglycolipid, cholesterol, and synthetic lipid, wherein the complex is coated with the lipid layer.

Non-aqueous liquid concentrate for aqueous dispersion

Palaikis, L.V., *et al.*, EarthClean Corp., US9758676, September 12, 2017

A non-aqueous liquid concentrate includes starch, an acrylic acid homopolymer salt, vegetable oil, and clay. The non-aqueous liquid concentrate forms an aqueous dispersion when added to water and is capable of clinging to a surface.

Patent information was compiled by Scott Bloomer, a registered US patent agent and Director, Technical Services at AOCS. Contact him at scott.bloomer@aocs.org.

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Journal authors invited to present at Annual Meeting

The 2018 AOCS Annual Meeting & Expo in Minneapolis, Minnesota, USA, May 6–9, will feature several presentations by authors of high-quality articles recently published in AOCS journals. The authors were invited by the journal editors based on the quality of their article and likelihood that the topic would enhance the annual meeting program. The invited authors will be highlighted in the Annual Meeting Program, and you can search for them using The App. If you are unable to attend, many of the presentations will be available in the inform|connect Premium Content Library after the meeting. Below is the list of chosen journal articles on which the annual meeting presentations will based.

Journal of the American Oil Chemists' Society

Rapid identification and relative quantification of the phospholipid composition in commercial lecithins by 31P-NMR

Yang, Y., *et al.*, *JAOCS 94*: 885–892, 2017, http://onlinelibrary.wiley.com/doi/10.1007/s11746-017-2992-0/abstract.

Author presenting: Ying Yang, *International Flavors & Fragrances Inc., USA*

Twenty-six phosphorus-containing hydrolysis products from six classes of phospholipids (PC, PI, PS, PE, PG, PA) were found and clearly identified and assigned by ^{31}P NMR measurement. The impact of pH on the chemical shift value for these hydrolysis products were observed and reported. The observation of pH-dependence of chemical shift values PA, LPA, GPA, P-choline and H_3PO_4 called for a caution for the analysis of phospholipid composition in complex hydrolysis lecithin samples by ^{31}P NMR. With the full assignment, rapid and routine analysis by using ^{31}P NMR to quantify phospholipid composition (including lyso-phospholipid class) without the need of phospholipid standards has become possible.

Sustainable synthetic approaches for the preparation of plant oil-based thermosets

Llevot, A., *JAOCS 94*: 169–186, 2017, http://onlinelibrary.wiley.com/doi/10.1007/s11746-016-2932-4/abstract.

Author presenting: Audrey Llevot, *LCPO, France*

This paper reviews improvements in the sustainability of chemical modifications and polymerizations of plant oil-based thermoset materials. Such advances include reducing the use of toxic reagents and corrosive acids; preventing waste by promoting biodegradability or self-healing behaviors, or limiting reaction steps and catalyst separation (latent catalysts); and reducing energy consumption by lowering reaction temperatures and employing photo-curing at room temperature.

Application of β-sitosterol + γ-oryzanol-structured organogel as migration barrier in filled chocolate products

Wendt, A., *et al.*, *JAOCS 94*: 1131–1140, 2017, http://onlinelibrary.wiley.com/doi/10.1007/s11746-017-3024-9/abstract.

Author presenting: Eckhard Flöter, *Technical University of Berlin, Germany*

The potential to suppress or prevent oil migration in filled pralines by incorporating sterol/sterolster-structured organogels was evaluated. Samples with different configurations, intermediate gel layer between chocolate and nougat filling, gelled nougat, and gelled chocolate were monitored for 24 weeks at three storage temperatures and assessed by differential scanning calorimetry analysis of a chocolate surface sample. The findings suggest that oil migration can be reduced by the application of sterol/sterolester oleogels, and that the addition of oil via the organogel does not promote additional oil migration.

Effect of spraydried flavonoid microparticles on oxidative stability of methyl linoleate as lipid model system

Palma, M., *et al.*, *JAOCS 94*: 99–105, 2017, http://onlinelibrary.wiley.com/doi/10.1007/s11746-016-2920-8/abstract.

Author presenting: Manuel J. Palma, *Universidad de Chile, Chile*

The structural features of encapsulated flavonoids (quercetin and epicatechin) prepared using inulin as the encapsulating agent and capsul as a channelizing agent were observed to

influence the encapsulation efficiency and oxidative stability of methyl linoleate. The addition of the quercetin microparticles showed the highest improvement in oxidative stability, as shown by an increase of the induction period and delay in the formation of oxidation compounds, especially in those microparticles with capsul (Q–IN–C). The results suggest that Q microparticles have a potential application to protect and to extend the shelf-life of lipid matrixes.

Journal of Surfactants and Detergents

Effect of pH on the binding of sodium, lysine, and arginine counterions to L-undecyl leucinate micelles

Corbin, L., *et al.*, *JSD 19*: 1175–1188, http://onlinelibrary.wiley.com/doi/10.1007/s11743-016-1875-y/abstract.

Author Presenting: Kevin F. Morris, *Carthage College*

Micelle formation by the amino acid-based surfactant undecylenyl l-leucine was investigated as a function of solution pH with nuclear magnetic resonance (NMR), dynamic light scattering, and fluorescence spectroscopy. The diameters of micelles in 50 mM undecylenyl l-leucine and 50 mM $NaHCO_3$ solutions were approximately 20 Å in diameter, and the radius and mole fraction associated with micelles changed very little with solution pH. The binding of the amino acids arginine and lysine to the anionic micelles was investigated from pH 7.0 to 11.5. Below pH 9.0, the mole fraction of arginine cations bound to the micelles was approximately 0.4. Above pH 9.0, the arginine counterions became zwitterionic, and the mole fraction of bound arginine molecules decreased steadily to less than 0.1 at pH 11.

Lipids

Identification of oxidized phosphatidylinositols present in OxLDL and human atherosclerotic plaque

Hasanally, D., *et al.*, *Lipids 52*: 11–26, http://onlinelibrary.wiley.com/doi/10.1007/s11745-016-4217-y/abstract.

Author presenting: Amir Ravandi

Oxidized low-density lipoprotein (OxLDL) plays an important role in initiation and progression of atherosclerosis. A major contributing factor influencing the atherogenicity of OxLDL is the composition of its lipid profile. This is the first identification of OxPtdIns molecules in human OxLDL and atherosclerotic plaque, making it possible to investigate their potential role in atherosclerosis.

Trans fatty acids suppress TNF-α-induced inflammatory gene expression in endothelial (HUVEC) and hepatocellular carcinoma (HepG2) cells

Da Silva, M.S., *et al.*, *Lipids 52*: 315–325, http://onlinelibrary.wiley.com/doi/10.1007/s11745-017-4243-4/abstract.

Author presenting: Iwona Rudkowska

Trans fatty acids (TFA) intake has been linked to cardiovascular diseases and liver diseases; yet the effect of TFA on inflammation remains controversial. To determine the *in vitro* effects of TFA on inflammatory gene expression, human umbilical vein endothelial cells (HUVEC) and human hepatocellular carcinoma (HepG2) cells were treated with various types of TFA. All TFA were highly incorporated into cell membranes, as determined by gas chromatography, representing 15–20% of total fatty acids in HUVEC and 3–8% in HepG2 cells. Incorporation of a common industrial TFA increased the ratio of stearoyl-CoA desaturase (SCD-1), a key enzyme involved in fatty acid metabolism. Ruminant TFA significantly reduced the TNF-α-induced gene expression of TNF, VCAM-1 and SOD2 in HUVEC, as well as TNF and IL-8 in HepG2 cells. Overall, physiological and supraphysiological concentrations of TFA prevented inflammatory gene expression *in vitro*. This effect is independent of PPAR-γ activation and may be due to an alteration of fatty acid metabolism in cell membranes caused by the high incorporation of TFA.

EXTRACTS & DISTILLATES

Effect of whole milk compared with skimmed milk on fasting blood lipids in healthy adults: a 3-week randomized crossover study

Engel, S., *et al.*, *Eur. J. Clin. Nutr.* 72: 249–254, 2018, https://doi.org/10.1038/s41430-017-0042-5.

Dietary guidelines have for decades recommended choosing low-fat dairy products due to the high content of saturated fat in dairy known to increase blood concentration of LDL cholesterol. However, meta-analyses including observational studies show no association between overall dairy intake and risk of cardiovascular disease and even point to an inverse association with type 2 diabetes. The objective was to compare the effects of whole milk (3.5% fat) with skimmed milk (0.1% fat) on fasting serum blood lipids, insulin, and plasma glucose in healthy subjects. A randomized, controlled 2×3-week crossover dietary intervention in 18 healthy adults randomly assigned to a sequence of treatments consisting of 0.5 L/d of whole milk and skimmed milk as part of their habitual diet. A total of 17 subjects completed the intervention. Whole milk increased HDL cholesterol concentrations significantly compared to skimmed milk ($P < 0.05$). There were no significant differences between whole milk and skimmed milk in effects on total and LDL cholesterol, triacylglycerol, insulin, and glucose concentrations. Intake of 0.5 L/d of whole milk did not adversely affect fasting blood lipids, glucose, or insulin compared to skimmed milk. Moreover, intake of whole milk increased HDL cholesterol concentration compared to skimmed milk. These findings suggest that if the higher energy content is taken into account, whole milk might be considered a part of a healthy diet among the normocholesterolemic population.

Evidence for the Initial Steps of DHA biohydrogenation by mixed ruminal microorganisms from sheep involves formation of conjugated fatty acids

Aldai, N., *et al.*, *J. Agric. Food Chem.* *66*: 842–855, 2018, https://doi.org/10.1021/acs.jafc.7b04563.

Inubation of DHA with sheep rumen fluid resulted in 80% disappearance in 6 h. The products were analyzed as their fatty acid (FA) methyl esters by GC-FID on SP-2560 and SLB-IL111 columns. The GC-online reduction × GC and GC-MS techniques demonstrated that all DHA metabolites retained the C22 structure (no evidence of chain-shortening). Two new transient DHA products were identified: mono-trans methylene interrupted-DHA and monoconjugated DHA (MC-DHA) isomers. Identification of MC-DHA was confirmed by their predicted elution using equivalent chain length differences from C18 FA, their molecular ions, and the 22:5 products formed which were the most abundant at 6 h. The 22:5 structures were established by fragmentation of their 4,4-dimethyloxazoline derivatives, and all 22:5 products contained an isolated double bond, suggesting formation via MC-DHA. The most abundant c4,c7,c10,t14,c19-22:5 appeared to be formed by unknown isomerases. Results suggest that the initial biohydrogenation of DHA was analogous to that of C18 FA.

Design, synthesis, and antifungal activities of 3-acyl thiotetronic acid derivatives: new fatty acid synthase inhibitors

Lv, P., *et al.*, *J. Agric. Food Chem.* *66*: 1023–1032, 2018, https://doi.org/10.1021/acs.jafc.7b05491.

Emerging fungal phytodiseases are increasingly becoming a food security threat. Twenty-six new 3-acylthiotetronic acid derivatives were designed, synthesized, characterized, and evaluated for activities against *Valsa mali*, *Curvularia lunata, Fusarium graminearum*, and *Fusarium oxysporum* f. sp. *lycopersici*. Among the 26 compounds, 6f was the most effective against *V. mali*, *C. lunata, F. graminearum*, and *F. oxysporum* f. sp. *lycopersici* with median effective concentrations (EC50) of 4.1, 3.1, 3.6, and 4.1 μg/mL, respectively, while the corresponding EC50 were 0.14, 6.7, 22.4, and 4.3 μg/mL of the fungicide azoxystrobin; 4.2, 41.7, 0.42, and 0.12 μg/mL of the fungicide carbendazim; and >50, 0.19, 0.43, and BS > 50 μg/mL of the fungicide fluopyram. The inhibitory potency against *V. mali* fatty acid synthase agreed well with the *in vitro* antifungal activity. The molecular docking suggested that the 3-acylthiotetronic acid derivatives targeted the C171Q KasA complex. The findings help understanding the mode of action and design and synthesis of novel potent fungicides.

Effects of plant sterols or β-cryptoxanthin at physiological serum concentrations on suicidal erythrocyte death

Alvarez-Sala, A., *et al.*, *J. Agric. Food Chem.* *66*: 1157–1166, 2018, https://doi.org/10.1021/acs.jafc.7b05575.

The eryptotic and hemolytic effects of a phytosterol (PS) mixture (β-sitosterol, campesterol, stigmasterol) or β-cryptoxanthin (β-Cx) at physiological serum concentration and their effect against oxidative stress induced by tert-butylhydroperoxide (tBOOH) (75 and 300 μM) were evaluated. β-Cryptoxanthin produced an increase in eryptotic cells, cell volume, hemolysis, and glutathione depletion (GSH) without ROS overproduction and intracellular Ca2+ influx. Co-incubation of both bioactive compounds protected against β-Cx-induced eryptosis. Under tBOOH

stress, PS prevented eryptosis, reducing Ca2+ influx, ROS overproduction and GSH depletion at 75 μM, and hemolysis at both tBOOH concentrations. β-Cryptoxanthin showed no cytoprotective effect. Co-incubation with both bioactive compounds completely prevented hemolysis and partially prevented eryptosis as well as GSH depletion induced by β-Cx plus tBOOH. Phytosterols at physiological serum concentrations help to prevent pro-eryptotic and hemolytic effects and are promising candidate compounds for ameliorating eryptosis-associated diseases.

One-step bioconversion of fatty acids into C8–C9 volatile aroma compounds by a multifunctional lipoxygenase cloned from *Pyropia haitanensis*

Zhu, Z.-J., *et al., J. Agric. Food Chem 66*: 1233–1241, 2018, https://doi.org/10.1021/acs.jafc.7b05341.

The multifunctional lipoxygenase PhLOX cloned from *Pyropia haitanensis* was expressed in *Escherichia coli* with 24.4 mg·L–1 yield. PhLOX could catalyze the one-step bioconversion of C18–C22 fatty acids into C8–C9 volatile organic compounds (VOCs), displaying higher catalytic efficiency for eicosenoic and docosenoic acids than for octadecenoic acids. C20:5 was the most suitable substrate among the tested fatty acids. The C8–C9 VOCs were generated in good yields from fatty acids, e.g., 2E-nonenal from C20:4, and 2E,6Z-nonadienal from C20:5. Hydrolyzed oils were also tested as substrates. The reactions mainly generated 2E,4E-pentadienal, 2E-octenal, and 2E,4E-octadienal from hydrolyzed sunflower seed oil, corn oil, and fish oil, respectively. PhLOX showed good stability after storage at 4°C for 2 weeks and broad tolerance to pH and temperature. These desirable properties of PhLOX make it a promising novel biocatalyst for the industrial production of volatile aroma compounds.

Difference in binding of long- and medium-chain fatty acids with serum albumin: the role of macromolecular crowding effect

Zhu, T.-T, *et al., J. Agric. Food Chem. 66*: 1242–1250, 2018, https://doi.org/10.1021/acs.jafc.7b03548.

Fatty acids (FAs) are transported by serum albumin in plasma. Studies have been undertaken to address the binding of MCFAs or LCFAs to human plasma albumin (HPA) and bovine serum albumin (BSA) by characterizing the binding affinities. Previous research on FA binding to serum albumin was usually performed in dilute solutions that are not sufficiently concentrated for the interpretation of the significance of the results under normal physiological conditions. How macromolecular crowded media affect fatty acids and bovine serum albumin (BSA) binding remains unknown. In this article, we investigated the mechanism of FA-BSA binding in a polyethylene glycol crowding environment by using thermodynamic and spectroscopic methods. Molecular crowding increased the binding constant for saturated medium-chain fatty acids (MCFAs) but significantly decreased the binding constant for unsaturated long-chain FAs. The binding sites tended to increase in all the cases. Further investigation revealed that crowding media might loosen the structure of BSA, facilitating MCFA-BSA binding. This research is useful for understanding the transportation of FAs by BSA under physiological conditions and may also help to control digestion by the eventual incorporation of macromolecular crowding agents into food formulations.

Sterols in infant formulas: a bioaccessibility study

Hamdan, I.J.A., *et al.*, *J. Agric. Food Chem. 66*: 1377–1385, https://doi.org/10.1021/acs.jafc.7b04635.dssss

The design of infant formulas (IFs) seeks to resemble human milk (HM) composition and functionality. The fat sources used usually comprise vegetable oil blends to mimic the fatty acid composition of HM and introduce changes in the animal/plant sterol ratio. In contrast, the use of milk fat globule membrane (MFGM)-rich ingredients could improve this aspect by increasing the ratio. The present study evaluates the bioaccessibility (BA) of sterols (cholesterol, desmosterol, brassicasterol, campesterol, stigmasterol, and β-sitosterol) in three IFs (with or without MFGM) using an in vitro digestion method simulating infant conditions. Analytical parameters confirmed the suitability of the method for all of these sterols. Results showed the presence of MFGM to increase cholesterol content (6–7 vs 2 mg/100 mL), this being the most bioaccessible sterol in the IFs. Although the BA of cholesterol was reduced in MFGM-enriched IF (65.6–80.4% vs 99.7%), the intake of bioaccessible cholesterol from these IFs was higher.

Eicosapentaenoic acid and docosahexaenoic acid containing supplements modulate risk factors for cardiovascular disease: a meta-analysis of randomised placebo-control human clinical trials

AbuMweis, S., *et al.*, *J. Human Nutr. Diet. 31*: 67–84, 2018, https://doi.org/10.1111/jhn.12493.

Over 200 clinical trials have examined the effect of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) supplements on risk factors associated with cardiovascular disease. However, an updated analysis of the evidence is lacking. The aim of the present meta-analysis was to quantify the effect of supplements containing EPA and DHA on risk factors for cardiovascular disease. An analysis was carried on 171 clinical trials with acceptable quality (Jadad score ≥3) that were identified from a comprehensive electronic search strategy of two databases (Pubmed and Cochrane Library). A random effect model was used to obtain an overall estimate on outcomes of interest. Heterogeneity between trial results was tested for using a standard chi-squared test. Results Compared with control, EPA and DHA supplements produced significant reductions of triglycerides of 0.368 mmol L−1 [95% confidence interval (CI) = −0.427 to −0.309], systolic blood pressure of 2.195 mmHg (95% CI = −3.172 to −1.217), diastolic blood pressure of 1.08 mmHg (95% CI = −1.716 to −0.444), heart rate of 1.37 bpm (95% CI = −2.41 to −0.325) and C-reactive protein of 0.343 mg L−1 (95% CI = −0.454 to −0.232). This analysis indicates an increase in both low-density lipoprotein cholesterol (mean difference = 0.150 mmol L−1; 95% CI = 0.058–0.243) and high-density lipoprotein cholesterol (mean difference = 0.039 mmol L−1; 95% CI = 0.024–0.054). The triglyceride-lowering effect was dose-dependent. The lipid-lowering, hypotensive, anti-arrhythmic and anti-inflammatory actions of EPA and DHA supplements were confirmed in this analysis of randomised placebo-control blinded clinical trials.

Impact of added phytosteryl/phytostanyl fatty acid esters on chemical parameters of margarines upon heating and pan-frying

Raczyk, M., *et al.*, *Eur. J. Lipid Sci. Technol. 120*: 1700281, 2018, https://doi.org/10.1002/ejlt.201700281.

The effect of the thermal treatment on chemical parameters of margarines enriched with phytosteryl fatty acid esters is evaluated. Sterols, fatty acids, polar and volatile compounds, dimers, trimers, tocopherols, 3-MCPD and glycidyl esters are analyzed applying GPC-RI, GC-FID, GC-MS, and HPLC. Additionally, a Rancimat test is performed to compare the oxidative stability of margarines. This study shows that margarines with and without added phytosteryl esters are both stable at a mild temperature of 60°C over a period of at least 7 days. On the other side after 15 min of pan-frying at 180°C a degradation of unsaturated fatty acids (14–47%), phytosterols (31–49%), and vitamin-E-active compounds (71–100%) is found. The supplementation of margarines with phytosteryl fatty acid esters have no effect on their oxidative stability during pan-frying characterized by Rancimat test and total polar compounds. Volatile, degradation products of fatty acids as well as phytosterols, are found after pan-frying. The composition of volatiles after pan-frying significantly differed depends on the presence of additional PSE in margarines. For the first time, it is shown that pan-frying with margarines over a period of 15 min results in a remarkable formation of 3-MCPD esters while the increase of glycidyl esters is only very moderate.

Characterization of the polymorphism of milk fat within processed cheese products

Ramel, P.R. and A.G.. Marangoni, *Food Struct. 12*: 15–25, 2017, http://dx.doi.org/10.1016/j.foostr.2017.03.001.

Differential scanning calorimetry (DSC) and X-ray diffraction (XRD) were used to characterize the polymorphism of milk fat within commercial processed cheese products. Using anhydrous milk fat (AMF) as control, we show that the dispersion of milk fat as cream and the embedding of milk fat globules into the casein matrix results in the formation of a higher proportion of the b polymorph of milk fat. Furthermore, the effects of other cheese ingredients on the polymorphic stability of milk fat were determined. By storing and determining the polymorphism of the cheese samples at the final melting temperature of AMF, we found similar amorphous XRD diffraction patterns which confirmed that the characteristic diffraction peaks observed at refrigeration temperatures are from milk fat crystals. Upon fast cooling of molten cheese sample, milk fat crystallized into the b0 polymorph. No correlations were found between the polymorphism of fat and small deformation dynamic shear rheological parameters of processed cheese products. Characterization of the polymorphism of milk fat within processed cheese provides better understanding of the crystallization behavior of fats as affected by the food matrix. Furthermore, the results of the study could provide insights into the functionality of milk fat within food at the nano- or molecular scale.

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Oxidative stability of refined olive and sunflower oils supplemented with lycopene-rich oleoresin from tomato peels industrial by-product, during accelerated shelf-life storage

Kehili, M., *et al.*, *Food Chem. 246*: 295–304, 2018, https://doi.org/10.1016/j.foodchem.2017.11.034.

Tomato peels by-product from a Tunisian industry was used for the extraction of lycopene-rich oleoresin using hexane solvent maceration. Tomato peels oleoresin, TPO, exhibited competitive free radicals scavenging activity with synthetic antioxidants. The efficacy of TPO in stabilizing refined olive (ROO) and sunflower (RSO) oils was investigated for five months, under accelerated shelf-life, compared to the synthetic antioxidant, butylated hydroxytoluene (BHT). TPO was added to ROO and RSO at four different concentrations, namely 250, 500, 1,000, and 2,000 µg/g and BHT standard at 200 µg/g. Lipid oxidation was tracked by measuring the peroxide value, acidity, conjugated dienes and trienes. Results suggested the highest efficiency of 250 µg/g and 2,000 µg/g of TPO, referring to 5 µg/g and 40 µg/g of lycopene, for the oxidative stabilization of ROO and RSO, respectively. The protective effect of TPO against the primary oxidation of these refined oils was significantly correlated to their lycopene contents.

Industrial Applications

Optimization of novel oil extraction technique from canola seeds: lecithin-based microemulsion

Radi, M. and S. Abbasi, *Eur. J. Lipid Sci. Technol.*, http://dx.doi.org/10.1002/ejlt.201700267.

A novel technique using a lecithin-based microemulsion system was developed for canola oil extraction. Pseudoternary phase diagrams of canola oil:lecithin:propanol:water microemulsions were constructed, and the effect of temperature on microemulsion formation was evaluated. (Lecithin:propanol):water microemulsion premixes [(2:1)50:50 and (2:3)80:20] were selected for optimization (temperature, exposure time, premix:canola seeds ratio, and agitation rate) of oil extraction from canola seeds using one-factor-at-a-time design. Based on "one factor at a time" design, the highest extraction yield (82.6% wt) was achieved at 60°C, 60 min, 6:1 premix:canola seeds ratio without agitation; the premix composition was (lecithin:propanol):water (2:1)50:50. The microemulsion-extracted oils had lower peroxide values, higher acidity, and phosphorous and lecithin contents comparable to those extracted with hexane. Simultaneous extraction, recovery, and solubilization capabilities were validated for the technique, which can be used for oil extraction and food formulation.

Congeneration biodiesel, ricinine, and nontoxic meal from castor seed

Zhu, Q., *et al.*, *Renew. Energy* 120: 51–59, 2018, https://doi.org/10.1016/j.renene.2017.12.075

Castor seed, as non-edible energy oilseed, was used to obtain several products, such as biodiesel, ricinine, and non-toxic castor seed meal through ultrasonic-assisted two-phase extraction (UATPE), alkaline transesterification, and recrystallization. The products with higher quality could be obtained by UATPE in shorter extraction time. The optimum conditions for UATPE were: an extraction time of 60 min, a corresponding temperature of 55°C, and a ratio of castor seed meal, petroleum ether, and deionized water of 1:4:6 (mg/mL/mL). Castor oil which was obtained by UATPE was used to produce the biodiesel. The yield of biodiesel could be increased up to 95.7% with the temperature of 60°C, the molar ratio of methanol to oil of 7:1, the amount of the catalyst of 0.7%, and the reaction time of 60 min. Meanwhile, with the trichrolomethanol as re-extraction agent, and ethanol as the recrystallization agent, the purity and recovery of the ricinine could reach 97.7% and 93.8%, respectively. In addition, the structural characterizations of the obtained ricinine and biodiesel were carried out by UV, FITR, ESI-MS, and other analytical methods.

Utilization of microalgae feedstock for concomitant production of bioethanol and biodiesel

Sivaramakrishnan, R. and A. Incharoensakdi, *Fuel 217*: 458–466, 2018, https://doi.org/10.1016/j.fuel.2017.12.119.

The present study focuses on the biorefinery approach of integrated production of bioethanol and biodiesel from microalgae feedstock. Various pretreatment methods were used to determine the maximum recovery of sugars from *Scenedesmus* sp. The total sugar yield of 93% was obtained when the biomass was pretreated by acid hydrolysis. The hydrolysate produced 86% of ethanol (theoretical yield) after the fermentation using *Saccharomyces cerevisiae*. Enzyme catalyzed direct transesterification of the biomass was performed using dimethyl carbonate as a solvent and the maximum yield of 92% methyl ester, 1.86% glycerol carbonate and 4.93% glycerol dicarbonate was achieved. The integrated process of bioethanol and biodiesel production was optimally achieved when direct transesterification was done first followed by ethanol fermentation yielding 92 and 93% of methyl ester and ethanol, respectively.

Assessment of subcritical propane, supercritical CO_2, and Soxhlet extraction of oil from sapucaia (*Lecythis pisonis*) nuts

Teixeira, G.L., *et al.*, J. *Supercrit. Fluids 133*: 122–132, 2018, http://dx.doi.org/10.1016/j.supflu.2017.10.003

The extraction of sapucaia (*Lecythis pisonis*) nut oil (SNO) using subcritical propane (SPE) and supercritical CO_2 (with ethanol as co-solvent; $scCO_2$) as solvent was investigated and compared with the conventional (Soxhlet) extraction. Extraction with $scCO_2$ was performed at 333 K and 20 MPa while the SPE extractions were carried out in different conditions to investigate the effects of temperature (293–333 K) and pressure (2–10 MPa) on the oil yield and the chemical compositions of the products. Results show that SPE allowed a fast extraction with a higher yield (46.22%) obtained at 333 K and 10 MPa, representing 93% efficiency compared to Soxhlet. Only temperature had significant ($p < 0.05$) effect on the extraction yield. SPE yielded the oils with highest values of polyunsaturated fatty acids (~36%). Stability to oxidation ranges from 6.53 to 11.17 h. The major triacylglycerols present in SNO are OOO, SOO, POO, PLO, and POS.

A novel method for extracting steryl glucosides from soy lecithin

Xie, M. and H. Zhao, *Eur. J. Lipid Sci. Technol. 120*: 1700247, 2018, https://doi.org/10.1002/ejlt.201700247.

In this study, a novel method utilizing enzyme and involving simple solvent extraction steps, is developed to yield an extract with high content of steryl glucosides (SG) and acyl steryl glucosides (ASG) from soy lecithin—the by-product of vegetable oil refining. Phospholipase A1 is used to convert phospholipids in soy lecithin into more hydrophilic hydrolysates, from which SG and ASG are separated by solvent extractions. A 3 × 3 full factorial design is employed to investigate the effects of two parameters (enzyme dose and reaction time) on three responses (yield of extract, SG/ASG content of extract, and recovery of SG/ASG). There are significant enzyme dose—reaction time interaction effects on all the responses, except for yield of extract. The highest SG (including ASG) content of more than 90% in the extract is achieved at enzyme dose of 0.03 g and reaction time of 16 h, where the yield of extract obtained is 1.43%.

Utilization of supercritical fluids for the effective extraction of waxes and cannabidiol (CBD) from hemp wastes

Attard, T.M., *et al.*, *Ind. Crops Prod. 112*: 38–46, 2018, https://doi.org/10.1016/j.indcrop.2017.10.045.

Up to 33% of hemp by mass can be lost in the form of dust during processing for fiber production. Heptane Soxhlet extractions and supercritical carbon dioxide extractions ($scCO_2$) of hemp dust samples yielded significant quantities of high value lipophilic molecules including fatty acids, policosanols (fatty alcohols), fatty aldehydes, hydrocarbons, sterols, triterpenoids, and cannabinoids (cannabidiol (CBD)). Dust collected from different stages of the mechanical processing of hemp fibers gave rise to lipophilic extractives with varying compositions, thus making the isolation and purification of these compounds easier. Of particular interest is CBD (5832.5 ± 118.9 μg/g of dust), which has attracted much attention for clinical-level studies due to its therapeutic efficacy in the treatment of a variety of central nervous system (CNS) disorders. Factorial experimental design was carried out to optimize the $scCO_2$ extraction, with 350 bar and 50°C yielding the selective extraction of higher value components.

Medical/Pharmaceutical Applications

Natural dietary products and their effects on appetite control

Suh, J.-H., *et al.*, *J. Agric. Food Chem.*, *66*: 36–39, 2018, https://doi.org/10.1021/acs.jafc.7b05104.

This is a quick reference for anyone wanting to understand the potential mechanism of action and the traditional use of natural products for appetite control.

Natural dietary products have been thoroughly studied for their effects of antiadipogenesis to prevent and treat obesity for decades. Nevertheless, in the past few years appetite control for the treatment of obesity has attracted much attention as a new target. Homeostatic control of energy intake involves a complex system that conveys peripheral signals to the central nervous system where multiple signals are integrated and then provide feedback to regulate satiation. This perspective aims at elucidating the neuronal mechanisms of food intake and energy balance as well as providing an alternative pathway of controlling weight using natural dietary products.

Antiobesity efficacy of quercetin-rich supplement on diet-induced obese rats: effects on body composition, serum lipid profile, and gene expression

Ting, Y., *et al.*, *J. Agric. Food Chem.* *66*: 70–80, 2018, https://doi.org/10.1021/acs.jafc.7b03551.

Quercetin and similar other compounds are naturally expressed in legumes and grains. For example, soy expresses soy isoflavones, which are marketed for various purposes. Quercetin is another isoflavone studied for its antiobesity benefits. From a mechanistic standpoint, it is important to understand how these molecules contribute to overall health, especially weight-related benefits.

The antiobesity effects of quercetin-rich supplement (QRS), which contain quercetin, lycopene, taurine, and litchi flower extract, on a high-fat diet (HFD)-induced obese rats were investigated. The rats that consume HFD with QRS (185 mg/kg rat) have significantly modulated the final body weights [490 ± 11 (HFD) → 441 ± 11 (HFD+QRS) g], total body fat [112.9 ± 4.5 (HFD) → 86.6 ± 5.7 (HFD+QRS) g], liver weights [14.8 ± 0.4 (HFD) → 12.6 ± 0.4 (HFD+QRS) g/rat], and the serum TG [102.5 ± 7.3 (HFD) → 90.7 ± 6.5 (HFD+QRS) mg/dL] to a level that resembled the regular diet-consumed rats ($p < 0.05$). The excretion of lipid in the faeces augmented in QRS groups as compared with the nonsupplemented HFD group [faecal total lipid: 62.43 ± 2.80 (HFD) → 73.15 ± 0.88 (HFD+QRS) mg/g dried faeces, $p < 0.05$]. In the histological analysis, quercetin-rich formulation supplemented groups presented a much less lipid accumulation and smaller size of adipocytes. Moreover, a decreased serum thiobarbituric acid reactive substances [1.55 ± 0.17 (HFD) → 0.78 ± 0.04 (HFD+QRS) nmol MDA eq/mL serum] increased levels of serum Trolox equivalent antioxidant capacity [3.89 ± 0.08 (HFD) → 6.46 ± 0.20 (HFD+QRS) μmol/mL serum], and more active hepatic antioxidant enzymes were observed in the supplemented groups ($p < 0.05$). The result of this work is a good demonstration of how a combination of bioactive compounds could work synergistically and become very effective in disease prevention.

Beneficial properties of phytochemicals on NLRP3 inflammasome-mediated gout and complication

Jhang, J.-J., *et al.*, *J. Agric. Food Chem.* *66*: 765–772, 2018, https://doi.org/10.1021/acs.jafc.7b05113.

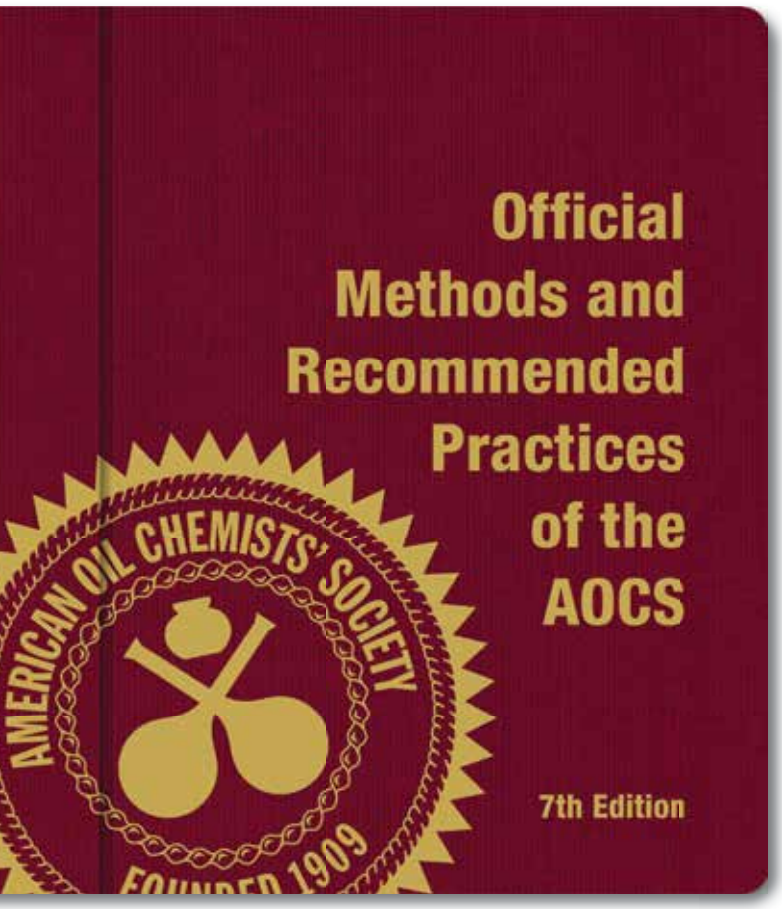
Inflammasomes—immune-system proteins that control inflammatory processes—are potential targets for developing treatments for inflammatory diseases. This review is a quick and short tutorial that highlights the importance of natural products and inflammasomes.

Gouty arthritis is characterized by the precipitation of monosodium urate (MSU) crystals in the joint. Pro-inflammatory cytokine IL-1β is a critical manifestation in response to MSU crystals attack. IL-1β secretion is dependent on the nucleotide-binding oligomerization domain-like receptor pyrin domain containing 3 (NLRP3) inflammasome. Abnormal activation of the NLRP inflammasome is related to cellular oxidative stress. However, recent studies have illustrated that phytochemicals with potent antioxidant activity exert inhibitory effects on NLRP3 inflammasome-mediated diseases. This review focuses on the current findings of studies on the NLRP3 inflammasome and the proposed mechanisms that MSU crystals trigger inflammation via activation of the NLRP3 inflammasome. We also summarized the potential use of phytochemicals on NLRP3 inflammasome-mediated diseases, suggesting that phytochemicals can further prevent acute gout attack.

Food bioactives and their effects on obesity-accelerated inflammatory bowel disease

Chiou, Y.-S., *et al.*, *J. Agric. Food Chem.* *66*: 773–779, 2018, https://doi.org/10.1021/acs.jafc.7b05854.

Current views support the concept that obesity is linked to a worsening of the course of inflammatory bowel diseases (IBDs). Gut microbiota and adipose tissue macrophage (ATM) are considered key mediators or contributors in obesity-associated intestinal inflammation. Dietary components can have direct or indirect effects on "normal" or "healthy" microbial composition and participate in adiposity and metabolic status with gut inflammation. In this perspective, we highlight food-derived bioactives that have a potential application in the prevention of obesity-exacerbated IBD,



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Whole egg consumption exerts a nephroprotective effect in an acute rodent model of type 1 diabetes

Saande, C.S., *et al., J. Agric. Food Chem. 66*: 866–870, 2018, https://doi.org/10.1021/acs.jafc.7b04774.

Food containing whole eggs offers renal protection in rats with type 1 diabetes. This study, if proven correct in humans, suggests that a diet that includes optimal quantities of whole eggs may provide renal protection in people with type 1 diabetes.

Nephropathy is a well-characterized complication of type 1 diabetes (T1D), resulting in proteinuria and urinary loss of micronutrients. We previously found that a whole egg-based diet maintained vitamin D balance in type 2 diabetic rats despite excessive urinary losses due to nephropathy. The goal of this study was to investigate the impact of whole egg consumption in T1D rats. Sprague–Dawley rats were randomly assigned to T1D or nondiabetic control groups and fed a casein or whole egg-based diet for 32 days. On day 26, two-thirds of the rats received a streptozotocin injection to induce T1D. Whole egg consumption attenuated polyuria, proteinuria, and renal hypertrophy in T1D rats. These data suggest that dietary intervention with whole egg may offer renal protection in T1D.

Microbial biosynthesis of silver nanoparticles in different culture media

Luo, K., *et al., J. Agric. Food Chem. 66*: 957–962, 2018, https://doi.org/10.1021/acs.jafc.7b05092.

Metal nanoparticles made by bacteria are a very promising development. Such nanoparticles are expected to have improved solubility and perhaps even better bioavailablity, opening the door wide to new types of medical applications.

Microbial biosynthesis of metal nanoparticles has been extensively studied for applications in biomedical sciences and engineering. However, the mechanism for their synthesis by microorganisms is not completely understood. In this study, several culture media were investigated for their roles in the microbial biosynthesis of silver nanoparticles (AgNPs). The size and morphology of the synthesized AgNPs were analyzed by UV–vis spectroscopy, Fourier-transform-infrared (FT-IR), transmission electron microscopy (TEM), and dynamic light scattering (DLS). The results demonstrated that nutrient broth (NB) and Mueller-Hinton broth (MHB) among tested media effectively reduced silver ions to form AgNPs with different particle size and shape. Although the involved microorganism enhanced the reduction of silver ions, the size and shape of the particles were shown to mainly depend on the culture media. Our findings suggest that the growth media of bacterial culture play an important role in the synthesis of metallic nanoparticles with regard to their size and shape. We believe our findings would provide useful information for further exploration of microbial biosynthesis of AgNPs and their biomedical applications.

Food synergies for improving bioavailability of micronutrients from plant foods

Nair, K.M. and L.F. Augustine, *Food Chem. 238*: 180–185, 2018, https://doi.org/10.1016/j.foodchem.2016.09.115.

The benefits of a balanced diet depend on food synergies that increase the bioavailability of nutrients.

Plant foods are endowed with micronutrients, but an understanding of bioavailability is essential in countries primarily dependent on plant-based foods. Bioavailability depends majorly on food synergies. This review examines the nature of certain food synergies and methods to screen and establish it as a strategy to control micronutrient deficiency in the populations. Strong evidence on the synergistic effect of inclusion of vitamin C rich fruits and non-vegetarian foods in enhancing the bioavailability of iron has been demonstrated. Fat is found to be synergistic for vitamin A absorption. Red wine and protein have been explored for zinc absorption and effect of fat has been studied for vitamin D. Methods for screening of bioavailability, and biomarkers to demonstrate the synergistic effects of foods are required. Translation of food synergy as a strategy requires adaptation to the context and popularization of intelligent food synergies.

Phenolic contents, cellular antioxidant activity, and antiproliferative capacity of different varieties of oats

Chen, C., *Food Chem. 239*: 260–267, 2018, https://doi.org/10.1016/j.foodchem.2017.06.104.

The objectives of this research were to determine the phenolic contents, oxygen radical absorbance capacities (ORAC), cellular antioxidant activities (CAA), and antiproliferative capacities of nine oat varieties and four brans in China. Of all varieties, Longyan 3 and Beiyan 1 exhibited the highest total avenanthramides (146.94 ± 7.31 and 120.95 ± 6.66 μg/g, respectively) and ORAC values (21.03 ± 0.56 and 21.18 ± 1.45 μM Trolox/g, respectively), while Shaotong exhibited the highest total phenolic acids (143.52 ± 9.42 μg/g) and CAA values (33.38 ± 1.74 μM quercetin/100 g). The EC50 of antiproliferative capacities ranged from 167.31 ± 6.42 to 233.42 ± 21.31 mg/mL, with the lowest in Beixiao 8 while the highest in Jinyan 8. ORAC values correlated with avenanthramides while CAA values correlated with phenolic acids. Moreover, phenolic contents, antioxidant properties, and antiproliferative capacities of oat brans was higher than that of corresponding whole oats in most cases.

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