THE VORTEX

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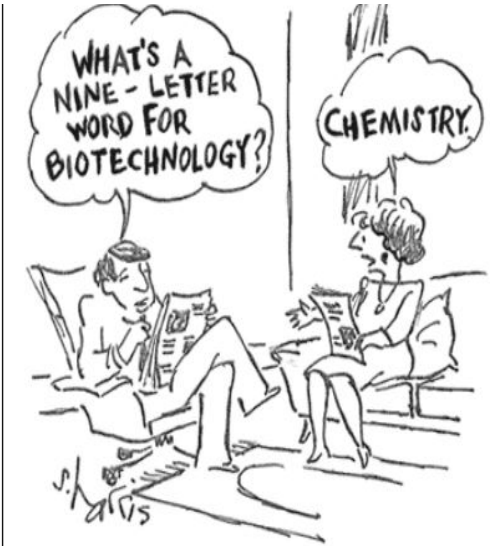

There is a healthy debate among scientists about which discipline can provide the best tools for solving key societal problems. This cartoon pokes fun at the sometimes—contentious relationship between chemists and biologists. Chemists are often the inventors and early developers of materials and techniques that drive biotechnology. Is this area (arguably biology's most useful side) then really just chemistry in disguise? Can chemists lay claim to the most impactful discoveries within it? Whether you'd argue yes or no to these questions, it could be the biology community that gets the last laugh. Another clue in the man's crossword puzzle: "A biologist who has made seminal discoveries in his or her field, perhaps (4 words)." The answer: N-O-B-E-L-L-A-U-R-E-A-T-E-I-N-C-H-E-M-I-S-T-R-Y.

---Chad Mirkin, Northwestern

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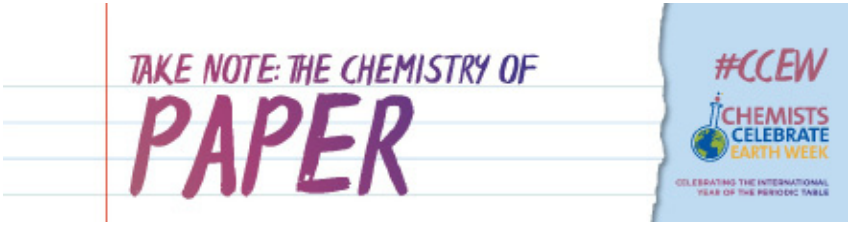
CHEMISTS CELEBRATE EARTH WEEK



The California Section, ACS will again join with dozens of other community groups to celebrate the 48th Anniversary of Earth Day and John Muir's 181st Birthday on April 20th, 2019 on the beautiful grounds of the John Muir National Historic Site in Martinez, CA. (10 AM - 4 PM). Our volunteers will interact with hundreds of families, young and old, at the Cal ACS canopy location. The hands-on demonstrations and activities will feature the ACS Earth Week theme, "Take Note: The Chemistry of Paper." The event will also celebrate the United Nations-declared International Year of the Periodic Table of the Chemical Elements (IYPT2019).

Your Earth Week Coordinator, Sheila Kanodia, requests your help as volunteers at the beautiful John Muir National Historic Site in Martinez on April 20th. Preferred shifts are from 10 AM -1 PM and 1 PM - 4 PM but we are flexible! Help us present enjoyable hands-on activities for young children, students and adults.

Each year, as part of Chemists Celebrate Earth Week (CCEW), ACS sponsors an illustrated poem contest for K-12 students. The California Section invites K-12 students to compete. Please visit the Earth Week page at calacs.org site for poetry contest rules and forms to participate.

For more information or to volunteer on April 20th, please contact our Section office at <office@calacs.or> or Sushila Kanodia at <sushila.kanodia@gmail.com>

Thank you!

JOHN MUIR NATIONAL HISTORIC SITE
4202 Alhambra Ave.
Martinez, CA 94553

Directions: https://www.nps.gov/jomu/planyourvisit/directions.htm

THE VORTEX

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Chair's Message

Dear members of the CalACS section,

Spring is right around the corner and so is the National ACS meeting in Orlando, FL. If you are attending the meeting and would like to share with the local section pictures from the national meeting, please add @CaliforniaACS to your tweets. We'll retweet them and they'll be displayed on our webpage.

The deadline for applications for the Lloyd Ryland Outstanding High School Chemistry Teacher Award is March 31st so please submit your applications to office@calacs.org. You can find the application under "Grants and Awards" tab on our website https://calacs.org. The local competition for the chemistry olympiad is also this month. You can read more about this event on our website, or at www.acs.org.

Calling all volunteers! If you are interested in helping out various events sponsored by California ACS, please e-mail office@calacs.org. We have plenty of events that can benefit from our local section members' help.

Patrick S. Lee Ph.D.

Gifts & Donations

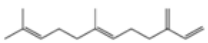
A gift of $25 to our High School Chemistry Teachers programs helps support the teacher and school with Chemistry supplies and equipment. Call or email and find out how your valued contribution can be used. Donations to the California Section are tax deductible.

Lou Rigali, LR101898@aol.com

WCC Meeting Report, Sugar-derived Lubricants: A High-performance, Ecofriendly Alternative to Petroleum-derived Lubricants Saturday, February 16, 2019 at Chevron's Richmond Technology Center, Richmond, CA

We welcomed Dr. Lynn Rice from Novvi in Emeryville with a large engaged audience for our first event of 2019. Lynn discussed her company's achievements in developing renewable and highly biodegradable lubricants with excellent performance characteristics.

Lynn started the presentation with an overview of lubricants, indispensable fluids that prevent wear and enable smooth operation of, as Lynn shared, "everything that moves". She explained that lubricants are composed of a base oil with chemical additives. The working environment in which the lubricant will need to perform determines the chemical and physical properties that the lubricant will need to have; examples include the thermal, oxidative, and hydrolytic stability, its viscosity and its low temperature properties. Formulations chemists, like Lynn, come up with the recipes of base oils and specific additives to design lubricants for specific applications.



In Lynn's work, the base oils are biologically derived, rather than from fossil fuels. In addition to being renewable, biolubricants have some superior characteristics relative to petroleum-derived ones, including generally better biodegradability and lower toxicity, and can also have significantly superior performance. The specific base oils Novvi makes are derived from renewable β-farnesene, a hydrocarbon produced by genetically modified yeast from sustainable sources of sugar. This foundational technology comes from Amyris, one of the joint venture partners of Novvi. Currently, the Novvi oil feedstock is produced from sugar cane in Brazil, but the fermentation is not feed-specific, and the hydrocarbon could be produced from a wide variety of plant materials.

Lynn briefly described the processes used to make base oils from the renewable hydrocarbon, highlighting the first step, a partial hydrogenation of β-farnesene into mono-olefin with high yield (~90%), a Novvi proprietary technology. In the following step, the mono-olefin is oligomerized with one or more linear alpha olefins (which can also be renewably sourced). To improve the chemical stability of the crude oligomer, remaining C-C double bonds are then saturated by hydrogenation. The final processing step is distillation leading to production of base oils in several viscosity grades. Lynn showed results of a proprietary test of base oil oxidative stability, and Novvi oils show the same outstanding performance as petroleum-derived Poly Alpha Olefin (PAO) synthetic oils.

However, the bio-derived Novvi oils have significant advantages over the fossil-fuel derived synthetic oils when one considers their environmental performance. First, the Novvi oils are 100% renewable hydrocarbons. Lynn discussed a life-cycle analysis of these oils that showed a 70% greenhouse gas reduction compared to the equivalent petroleum hydrocarbons. Further, the Novvi oils have better biodegradability, with typical PAOs showing 5 to 40% biodegradability, while Novvi products range from about 3 to greater than 70% biodegradable.

Novvi products are already in use, and Lynn shared some examples with us. One large US university requires biodegradable hydraulic fluids to be used in its elevators. The university originally used a vegetable oil based fluid, but had low-temperature performance problems, as well as other maintenance concerns. The university changed to a Novvi hydraulic fluid and now enjoys good performance from a biodegradable product. In particular, the

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P*FAS: PFOA, and PFOS (Part 4)*

Bill Motzer

In this series, I've discussed perfluoroalkylated substances (PFAS), a large group of fluorinated compounds, including oligomers and polymers. PFAS consist of neutral and anionic surface-active compounds having high thermal, chemical, and biological inertness that are generally hydrophobic but are also lipophobic. Therefore, they do not accumulate in fatty tissues but can accumulate in human blood and breast milk. An important chemical subset are perfluorinated organic surfactants such as perfluorooctanoic acid (PFOA) and perfluorooctane sulfonate (PFOS). These were discussed in more detail in Parts 2 and 3 (January and February 2019 Vortex).

PFOS was also used by 3M in the manufacture of aqueous film-forming foams (AFFF) used in suppressing and quelling fuel-fed fires, particularly after aircraft crashes and in extinguishing fires at oil refineries, chemical plants, and fuel storage-tank facilities. Scientists at 3M originally believed that PFOS was relatively inert and would not accumulate in the environment. However, by 2000 the company admitted that PFOS-based surfactants, which were used in AFFF, were accumulating in the environment, particularly in soil, surface water and groundwater and subsequently emerging in humans and animal tissues at concentrations that raised health questions and concerns. Similar PFOA-based surfactants were also soon linked to potential human health problems.

A 2016 Harvard T.H. Chan School of Public Health investigation, using U.S. Environmental Protection Agency (U.S. EPA) data, discovered PFAS in drinking water at 664 military training facilities and 533 civilian airports. In 2017, a Department of Defense (DOD) report to Congress admitted that there were 393 active and closed military installations with known or suspected PFOA or PFOS drinking water contaminants. Because of these studies, in 2017, Washington State passed legislation banning PFAS-containing AFFF beginning in 2020. Fire trucks will no longer be allowed to spray AFFF on fuel spills and car fires, although AFFF use will continue at airports, military bases, petroleum refineries, and chemical plants. In October 2018, Congress passed, and the President signed the Federal Aviation Administration (FAA) Reauthorization Act of 2018 allowing, by 2021, civilian airports usage of fluorine-free foams. However, current FAA rules still require U.S. airports to use military-grade foams containing PFAS. Public-interest groups such as the Environmental Working Group and the International Persistent Organic Pollutants Elimination Network (IPEN) want immediate cessation of PFAS AFFF. (A more detailed report of PFAS in AFFF can be found in Chemical and Engineering News, January 14, 2018, pp. 16-19.)

The current analytical detection method for PFOS and PFOA is by liquid chromatography-mass spectrometry/tandem mass spectrometry (LC-MS/MS). However, both LC-MS/MS and gas chromatography-mass spectrometry (GC-MS) can also be used to determine PFOS and PFOA precursors. Current methodologies for PFAS analysis are designed to measure a discrete list of about 20 compounds. PFOA and PFOS are generally in nanogram per liter (ng/L) amounts in the reporting limit range of 2-15 ng/L.

To date, no fully validated or approved EPA method for PFAS detection in non-drinking water (i.e., surface water, groundwater, wastewater, and recycled water) or solids at State or Federal levels exists. For drinking water, the U.S. EPA has authorized use of EPA Method 537 for selected perfluorinated alkyl acids (PFAAs). For non-drinking water matrices, some analytical laboratories are using modified 537 methods using direct injection-isotope dilution. However, these

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modified methods have no consistent sample collection guidelines and have not been validated or systematically assessed for data quality. Additionally, there are only a few reports of analysis of foods using this method and, because of the lack of suitable analytical data, many assumptions have been made to determine exposure estimates from food products.

Many additional PFAS are not determined as discrete compounds by existing analytical methods, including EPA Method 537. Therefore, current environmental PFAS risk potential may be grossly underestimated. Public entities and state environmental agencies are pressuring the U.S. EPA to develop more suitable methods that more closely measure the full extent of PFAS environmental contamination. Thus, the U.S. EPA has developed a test method under the Resource Conservation and Recovery Act (RCRA) publication SW-846 – Test Methods for Evaluating Solid Waste: Physical/Chemical Methods. The SW-846 direct injection method allows for quantifying 24 PFAS analytes in matrices other than drinking water. It also includes development of an analytical method for determining short-chained PFAS in drinking water with planned external laboratory validation by ten labs with public review by February 2019 and validation of a solids method using isotope dilution.

In future articles I'll review current and proposed U.S. EPA and California PFAS regulatory requirements, PFAS sample collection procedures, and some possible forensic techniques for source identification.

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Novvi product eliminated the need for oil heating systems that the vegetable oil formulations had required. In a second example, Novvi has formulated an automotive engine oil (0W-20) that has been in use at the Sonoma Raceway for nearly three years. Based on the good performance seen there in its racing fleet, use of Novvi products has expanded throughout the Raceway's operations, including more conventional fleet vehicles and shop equipment. Test results on other challenging applications including oil for a high-temperature air compressor, and an excavation field test of a hydraulic fluid, showed notably superior performance for Novvi's products.

Lynn and her colleagues at Novvi have demonstrated that top-tier lubricant performance can be achieved by fluids with top-tier environmental friendliness.

After her presentation, Lynn answered many questions from the interested audience. In this part of the presentation, we learned that Novvi oils are similar in cost to PAO products. While the β-farnesene is produced in Brazil in close proximity to where the sugar cane is produced, the lube oils are manufactured in a plant in Deer Park, TX. The products are made on a metric ton scale presently.

Approximately 60 people attended Lynn's presentation which was followed by an informal pizza lunch. We were particularly pleased by the significant number of college students in attendance, with a particularly large group from Los Medanos Community College.

Letter to the Editor

If you have questions about how to get attendance at a local section meeting ask the WCC. (see report on page 4) Great Topic: Sugar-Derived High Perfmance Lubricants; Dynamic Speaker: Lynn Rice; There were ~40 STUDENTS from Berkeley and Los Medanos and ~ 15 others. We made contacts with the ACS Student Affiliates at UCB.

Al Verstuyft

New Findings Regarding the Antiadhesive Activity of Cranberry Phenolic Compounds and Their Microbial-Derived Metabolites against Uropathogenic Bacteria

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Abstract

Findings concerning the antiadhesive activity of cranberry phenolic compounds and their microbial-derived metabolites against Gram-negative (Escherichia coli ATCC 53503 and DSM 10791) and Gram-positive (Enterococcus faecalis 04-1) bacteria in T24 cells are reported. A-Type procyanidins (A2 and cinnamtannin B-1) exhibited antiadhesive activity (at concentrations ≥250 μM), a feature that was not observed for B-type procyanidins (B2). The metabolites hippuric acid and α-hydroxyhippuric acid also showed effective results at concentrations ≥250 μM. With regard to conjugated metabolites, sulfation seemed to increase the antiadhesive activity of cranberry-derived metabolites as 3-(3,4-dihydroxyphenyl)propionic acid 3-O-sulfate presented active results, unlike its corresponding nonsulfated form. In contrast, methylation decreased antiadhesive activity as 3,4-dihydroxyphenylacetic acid was found to be active but not its corresponding methylated form (4-hydroxy-3-methoxyphenylacetic acid). As a whole, this work sustains the antiadhesive activity of cranberry-derived metabolites as one of the mechanisms involved in the beneficial effects of cranberries against urinary tract infections.

Keywords: bacteria adherence; bladder cells; cranberry; microbial-derived phenolic metabolites; urinary tract infections (UTI); uropathogens

Introduction

Urinary tract infections (UTI) are the most pervasive of bacterial infections and represent a large economic and medical burden worldwide. Antibiotics are generally prescribed as treatment against UTI, although they can have adverse side effects, including the development of antimicrobial resistance and reduction of beneficial intestinal bacteria.(1) As a natural alternative, cranberry (Vaccinium macrocarpum) has been widely recommended in the prevention and treatment of UTI prophylaxis.(2,3) Among other components, cranberry is rich in polyphenols, particularly proanthocyanidins, anthocyanidins, and flavonols, together with phenolic acids and benzoates. (4) Although many epidemiological and intervention studies have proved the efficacy of cranberry products in UTI prophylaxis,(2,3) others have shown mixed results.(5,6) One of the last meta-analyses concerning this topic(7) reported a large interindividual variability in cranberry efficiency against UTI; it also concluded that patients at some risk of these infections were more susceptible to the beneficial effects of cranberry consumption.(7)

Among other possible mechanisms behind the protective effects of cranberry against UTI is the capacity of cranberry polyphenols to act as antiadhesive agents in preventing/inhibiting the adherence of pathogens to uroepithelial cell receptors,(8) which appears to be a major step in the pathogenesis of these infections.(9) One of the very first studies about this topic reported the *in vitro* inhibition of the adherence of uropathogenic P-fimbriated Escherichia coli by procyanidin A2 and other cranberry A-type procyanidins.(10) Numerous studies have further proven the antiadhesive activity of different cranberry phenolic compounds/fractions/extracts against strains of uropathogenic E. coli (UPEC) using cell culture methodologies.(11−13) Experiments on cell cultures have also evidenced antiadhesive activity against UPEC of urine collected after

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the consumption of cranberry products.(5,14−16) But it is unlikely that the compounds responsible for the antiadhesive activity of urine collected after cranberry consumption would be the original forms present in the original food. It is known that polyphenols, and especially proanthocyanidins, are poorly absorbed in the small intestine, and reach the colon where they are catabolized by the gut microbiota to give rise to a great battery of phenolic metabolites that can be absorbed and further secreted in urine.(17) In fact, in a recent study, Peron and co-workers(18) demonstrated that the antiadhesive properties of urine after cranberry consumption could not be ascribable to the direct effect of A-type proanthocyanidins because their levels in urine were in the low-concentration range (<nM).(18) Therefore, the question that remains to be answered is what is the compound(s) present in urine after cranberry consumption that prevents/inhibits effective adherence of uropathogens to uroepithelial cells. Just recently, Feliciano and co-workers published valuable data regarding the absorption, metabolism, and excretion of cranberry polyphenols(19,20) that could help in the resolution of this cranberry antiadhesive activity puzzle. After cranberry juice consumption by healthy men, a total of 60 phenolic-derived metabolites were quantified in plasma and urine, including hippuric acids, pyrogallol sulfates, valerolactone, benzoic acids, phenylacetic acids, and flavonol glucuronides, as well as cinnamic acid sulfates and glucuronides.(19) Although some of these compounds were tested for antiadhesive activity such as phenolic acids(21) and valerolactones,(22) no data are available for the majority of them, particularly for compounds found in high concentrations in human fluids, such as hippuric and α-hydroxyhippuric acids. On the other hand, the majority of the in vitro studies about antiadhesive activity against uropathogens have focused on P-fimbriated E. coli strains. However, Gram-positive bacteria, including some staphylococcal and enterococcal species, also seem to be implicated in the etiopathogenesis of UTIs,(23) particularly among the elderly population, pregnant women, and people with any other risk factor for UTIs.(24) Therefore, the goal of this paper has been to assess the effect of cranberry phenolic compounds and their metabolites on bacteria adherence to bladder uroepithelial cells, not only for Gram-negative (E. coli ATCC 53503 and DSM 10791) but also for Gram-positive (Enterococcus faecalis 04-1) species. Among the compounds tested, we have included cranberry flavan-3-ols (both A-type and B-type proanthocyanidins) and phenolic metabolites exhibiting a high abundance in urine after cranberry consumption (i.e., hippuric and α-hydroxyhippuric acids) as well as other conjugated metabolites such as sulfate and methyl derivatives [i.e., 3-(3,4-dihydroxyphenyl)propionic acid 3-O-sulfate and 4-hydroxy-3-methoxyphenylacetic acid]. Additionally, and to ensure that the compounds tested were the forms responsible for the observed effects in the cells, their stability after bacteria adherence assays was evaluated through ultra-high-performance liquid chromatography-mass spectrometry (UHPLC-MS) analysis.

Consumption of cranberry (Vaccinium macrocarpon) has been widely recommended for prophylaxis against urinary tract infections (UTI),(2,3) although the mechanisms behind these effects have not been fully revealed.(8) This study points out some new features regarding the antiadhesive activity of cranberry phenolic compounds and their microbial-derived metabolites against uropathogens. First, this paper has demonstrated that the adherence of enterococcal strains (Gram-positive) such as E. faecalis to bladder uroepithelial cells was also inhibited after incubation of the bacteria with certain phenolic compounds. In fact, among the three strains used, sensitivity against phenolic compounds was in the order E. faecalis 04-1> E. coli DSM 10791 > E. coli ATCC 53503. For example, cinnamtannin B-1 and hippuric acid were active against E. faecalis 04-1 at 250 μM, but not against E. coli DSM 10791. Although

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most studies about the antiadhesive activity of cranberry phenolic compounds have subscribed to P-fimbriated E. coli strains, some studies have also proven in vitro antiadhesive activity against other Gram-negative bacteria such as Proteus mirabilis, a species that has been associated with infections in catheterized patients or those having structural anomalies of the urinary tract.(31) Recently, E. faecalis species have received much attention because of their resistance to aminoglycosides and vancomycin.(32) In the same vein, Wojnicz and co-workers(33) explored the effects of a cranberry extract on the growth, virulence factors, and biofilm formation of E. faecalis urine isolates. As far as we know, this paper reports, for the first time, data concerning the activity of cranberry phenolic compounds on the adherence of E. faecalis to uroepithelial cells. Together, the results suggest a wide activity spectrum against uropathogenic bacteria by cranberry phenolic compounds and their microbial-derived metabolites, at least in in vitro conditions.

Another new finding reported in this paper concerns the antiadhesive activity of flavan-3-ols, particularly of trimer cinnamtannin B-1 [epicatechin-(4β→8, 2β→O→7)-epicatechin-(4α→8)-epicatechin]. To the best of our knowledge, this is the first time that an A-type procyanidin molecule different from A2 [epicatechin-(4β→8, 2β→O→7)-epicatechin] has been tested for in vitro antiadhesive activity against uropathogens. Both A2 and cinnamtannin B-1 significantly inhibited the adherence of three strains of bacteria tested (E. coli ATCC 53503, E. coli DSM 1079, and E. faecalis 04–1) at concentrations ≥250 μM, a feature that was not observed for procyanidin B2 [(−)-epicatechin-(4β-8)-(−)-epicatechin]] in line with previous reports.(11,21) Therefore, it could be suggested that a certain structure–activity relationship for A-type procyanidins as dimers (i.e., A2) led to higher inhibition percentages (%) than trimers (i.e., cinnamtannin B-1) for the uropathogenic stains tested. The lower number of A-type linkages and/or the greater molecular volume might be the chemical structural features that explained the inverse relationship between the polymerization degree and the antiadherence activity of A-type procyanidins. But in any case, the antiadherence activity of A-type procyanidins is unlikely to be relevant in vivo as their concentration in urine is null or very low as a consequence of their extensive microbial metabolism in the gut.(18,19)

The wide interindividual differences observed in the human urinary/plasmatic phenolic metabolite profile have led to the introduction of the concept of polyphenol "metabotypes" or polyphenol metabolizing phenotypes,(37) which groups subjects with similar capacity to metabolize dietary polyphenols. Some studies have shown subject clusterization according to the capacity to metabolize polyphenols from soy,(38) from pomegranate,(39) or from wine,(40) among other foods. It remains to be proven if the intake of cranberry polyphenols would also cluster subjects according to their phenolic metabolite profile in physiological fluids (urine, plasma, and feces). However, recent studies reporting a preliminary elucidation of urinary metabotypes concerning the metabolism of flavan-3-ols,(41) one of the main classes of polyphenols present in cranberry, make us hypothesize that there could be different phenolic metabotypes related to the production of gut microbial metabolites from cranberry polyphenols, and that these hypothetical metabotypes could be one of the factors putatively associated with the great variability observed in the efficacy of cranberry products.

Finally, in relation to the in vitro conditions used to evaluate the antiadhesive activity against uropathogens, the UHPLC-MS analysis carried out in this study ensured stability of the phenolic metabolites tested under the cell culture conditions and UHPLC-MS parameters used in this study. Moreover, there was no apparent cellular and/or bacterial metabolism of the phenolic compounds tested as no new com-

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pounds were detected. In a previous paper, the possible transformations of phenyl-γ-valerolactones in bladder epithelial cells infected or not with UPEC were evaluated. While 5-(3,4-dihydroxyphenyl)-γ-valerolactone did not undergo any metabolic transformation, its sulfate derivatives underwent a certain metabolism by T24 cells that comprised the opening of the lactone ring and/or desulfation, the latter only to a very small extent (0.5–2%).(22) Compound stability and cell metabolism during cell culture assays is indeed a matter about which great care should be taken in studying bioactivity in vitro.

In summary, the results of this paper demostrate that A-type procyanidins (A2 and cinnamtannin B-1) but not B-type procyanidins (B2) are able to inhibit not only the adherence of uropathogenic E. coli but also of Gram-positive uropathogenic bacteria to bladder cells in in vitro conditions, although it is unlikely that procyanidins were present in urine at relevant physiological concentrations. It is also reported for the first time that cranberry-derived phenolic compounds detected in urine after consumption of cranberries and/or cranberry products such as hippuric acid, α-hydroxyhippuric acid, 3,4-dihydroxyphenylacetic acid, and dihydrocaffeic acid 3-O-sulfate are also able to inhibit the adherence of the uropathogenic bacteria tested. Additive (and even synergistic) effects among all the cranberry-derived phenolic metabolites present in urine are expected in vivo, which could explain, at least partly, the preventive and/or curative effects of the consumption of cranberry against urinary tract infections. Although a lack of standardization in terms of composition and dosage for cranberry products has been considered one of the main reasons for the large differences in the efficacy of cranberry consumption observed among the many epidemiological and intervention studies that have been carried out, interindividual variability in the capacity to metabolize cranberry polyphenols may have a lot to do with the beneficial effects associated with cranberry. New investigations into the gut microbial metabolism of cranberry polyphenols and their further conjugation, mainly in the liver, will give more clues in moving toward a resolution of the cranberry UTI-preventing puzzle.

Part of this work was funded by Ocean Spray Cranberries, Inc. (Lakeville, MA).

Editor's Note

This version of the paper has been significantly edited leaving out figures, tables, references, and some methodology. The original paper can be found at the following link.https://pubs.acs.org/doi/pdf/10.1021/acs.jafc.8b05625

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