

THE VORTEX



Which mushroom is toxic?
Help is on the way, see page 2

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CalACS meeting Report: New Test for Detecting the Most Poisonous Mushroom toxin

On Wednesday May 20, 2020 The Section held an Online Zoom Event featuring Dr. Candace Bever, Microbiologist in the Foodborne Toxin Detection and Prevention Research Unit (FTDP) of USDA in Albany CA.

Dr Bever discussed a simple, 10 minute-portable test that can detect Amanitin the deadly toxin that is the class of mushroom toxins that cause the most serious issues.

In 1997 Sam Sebastiani Jr. the adult son of Sonoma Valley vintner Sam Sebastiani Sr. died after eating a death Cap mushroom from a park in San Francisco. The new test is an antibody-based assay, similar to a pregnancy test. This test specifically binds deadly amanitins, toxins produced by some wild mushrooms. The test is completed in 10 minutes and can detect amanitins either from mushroom samples or from urine of an intoxicated person or dog. Dr. Candace Bever describe how the test was made, how it works, and showed examples of using it with mushrooms and urine samples.

This test only identifies the presence or absence of this specific class of toxin; it does not detect other compounds such as hallucinogens or toxins that cause other gastrointestinal or neurological symptoms.

So, it cannot determine if a mushroom is edible

The new test can identify the presence of as little as 10 parts per billion of amanitin in about 10 minutes from a rice grain size sample of a mushroom or in the urine of someone who has eaten a poisonous amanitin-containing mushroom. The test also works with dog urine, as dogs are known to indiscriminately eat mushrooms.

No definitive point-of-care clinical diagnostic test currently exists for amatoxin poisoning. Early detection of amanitin in a patient's urine would help doctors trying to make a diagnosis. This work may help develop such a test. There appears to be an effort to commercialize the test. Meanwhile I will continue to purchase mushrooms for my *marinra* pasta and *frittata* sauce at my supermarket.

Dr Bever received her B.S. in biological sciences from Carnegie Mellon University and earned her Ph.D. in marine science from the Virginia Institute of Marine Science at the College of William & Mary. She completed a post-doc and served as a Project Scientist at UC Davis before joining the USDA.



Olympiad

Our high school students have had a banner year in the Olympiad! Two students from Dougherty Valley High, Anugraph Chemparathy and Michael Han, scored High Honors were again among the top 20 students who have qualified to attend Study Camp this year which will be held virtually. Venkat Raman and Kenneth Moon of Dougherty Valley and Ian Chen of California High scored High Honors and are among the top 50 students who took the exam. Of the twenty student who attend Study Camp, four will be selected to be the American Team along with two alternates to compete in the International Olympiad if it is held.

Zachery Deng (Lowell), Vivian Hir (Quarry Lane), Jeffery Liu (American), Eric Ma (Mission San Jose), Jay Sarva (Stanford Online), and Christina Yu (Mission San Jose) scored Honors and are among the top 146 student who took the exam.

Eileen Nottoli

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Chair's Message

Jim Postma

I thought it might be time again to plan your summer reading. Given the quarantine, you might be looking for something productive to occupy your time or you might be looking for gift ideas for a science-oriented relative or colleague. Here are my suggestions:

What's Cooking in Chemistry?: How Leading Chemists Succeed in the Kitchen by Hubertus P. Bell et al, Editors. You'll get more than 50 personal recipes and anecdotes from leading organic chemists, such as *Lonely soup*, *Wild boar - Tuscan way*, and *Dulce de Leche*. You will also get biographies and sketches of their work. This is the one if you like cooking, eating, and chemistry.

The Poison Squad: One Chemist's Single-Minded Crusade for Food Safety at the Turn of the Twentieth Century by Deborah Blum. The New York Times Book Review description: "The Poison Squad offers a powerful reminder that truth can defeat lies, that government can protect consum-

ers and that an honest public servant can overcome the greed of private interests." African American Women Chemists by Jeannette E. Brown (and if you like that, there's a sequel: *African American Women Chemists in the Modern Era.*) From an ACS Fellow.

If you are looking for a novel, *The Chemist* by Stephenie Meyer fits the bill, especially if you favor thrills and espionage.

At the other end of the genre spectrum is *A Well-Ordered Thing: Dmitri Mendeleev and the Shadow of the Periodic Table* by Michael D. Gordin. There's a lot more to Dmitri Mendeleev than his Periodic Table (as if that wasn't enough.)

And, if you are interested in the human side of the progress of chemistry, *Cathedrals of Science: The Personalities and Rivalries That Made Modern Chemistry* by Patrick Coffey. You will read about several well-known physical chemists from a perspective beyond just their science.

If you are tempted to write a review of any of these, contact Lou Rigali, the Vortex editor; he would probably be interested in making it a feature in the coming months. You could also contribute to the J. Chem. Ed. column of book reviews.



Cal ACS offers basic Zoom training

ALEX MADONIK

In this age of COVID-19, our virtual world has expanded as our physical world has contracted. With so much of our professional and personal lives moving to online platforms, we're all trying to learn more about online meeting tools.

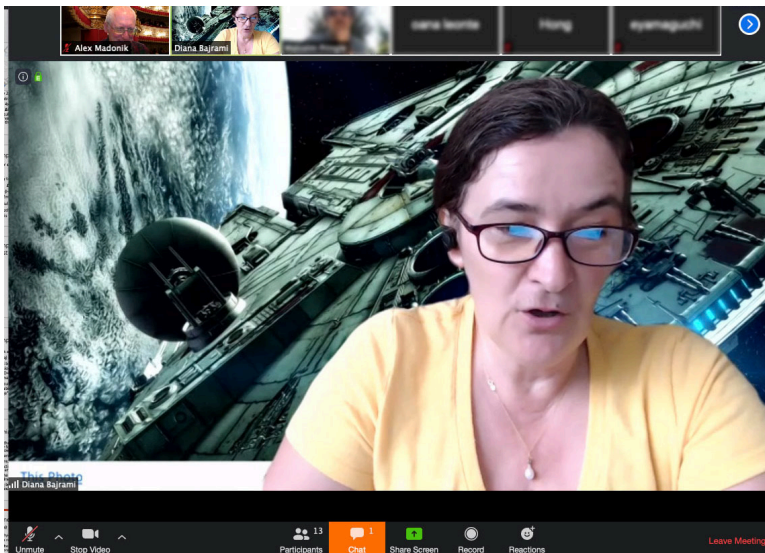
In California, all instructors in the state's public university and community college system have access to Zoom accounts, but many of us had never used them before March of this year. By now, probably every reader of the Vortex has participated in a Zoom meeting, and California Section Chair Jim Postma suggested that we organize some training sessions, to be sponsored by the Senior Chemists Committee. At the College of Alameda, I've been fortunate to work with some talented distance education specialists, and Professor Diana Bajrami agreed to offer two introductory training sessions, which took place on May

11th and 13th. There were over a dozen participants in each session, an ideal size for interaction and asking questions.

Professor Bajrami's presentation covered Zoom basics, such as setting up a free account (meetings are limited to 40 minutes, but can include 100 participants), inviting participants, managing meetings, and meeting security. She demonstrated features such as Chat and Breakout Rooms, which allow participants to exchange text messages or hold separate group discussions. Everyone came away better prepared to host their own meetings.

CalACS is using a professional-level account to host unlimited meetings for larger groups (up to 300 participants). The Executive Committee has already met twice on Zoom (much more convenient for some of our distant members) and almost 60 people joined us for the May 20th Section Meeting on the Detection of Amanitin Mushroom Toxins. Even bigger Zoom meetings are clearly in our future!

You can review Professor Bajrami's presentation on the Cal ACS web site, which also provides links to other resources.



Screen shot of Dr Professor Bajrami's Zoom presentation with a little enhancement by the reporter.



Diluting Isotopes

Bill Motzer

In my previous articles (see Dec. 2018 through June 2019 Vortex) of per- and poly-fluoroalkyl substances (PFAS), I discussed the chemistry of a “family” of manufactured (anthropogenic) chemicals used in products from the 1940s to the early 2000s that resist heat, oils, greases, stains, and water. Such surface-active agents were included in aqueous firefighting foams (AFFF), stain-resistant products, coating additives (i.e., polytetrafluoroethylene or PTFE also known as Teflon™), and cleaning products. Industrial uses were widespread spanning aerospace, automotive, chemical, construction, semiconductor, and textile companies. Under typical environmental conditions PFAS do not hydrolyze, photolyze, or biodegrade. Therefore, they are extremely environmentally persistent with potential to bioaccumulate and biomagnify in wildlife because they are readily absorbed upon ingestion, primarily accumulating in blood serum, kidneys, and liver. Animal toxicological studies have indicated potential developmental, reproductive, and systemic effects.

The most well-known and researched PFAS compounds are PFOA (perfluorooctanoic acid – $C_8HF_{15}O_2$; CAS No.: 335-67-1) and PFOS (perfluorooctane sulfonic acid – $C_8HF_{17}O_3S$; CAS No.: 1763-23-1). Within the U.S., PFOS and PFOA were the two PFAS compounds produced in the largest commercial amounts. PFOA is a perfluoroalkyl carboxylate synthetically produced as a salt and its ammonium salt is the most widely produced form. PFOS is commonly used as a simple salt (such as potassium, sodium, or ammonium) or is incorporated into larger polymers.

There are approximately 4,700 known PFAS compounds and these occur in almost all global environments including remote places. There are now major environmental concerns for surface-water and groundwater contamination in urban industrial areas. Health-based advisories or screening levels for PFOA and PFOS in drinking water have been developed by the U.S. Environmental Protection Agency (U.S. EPA) and state regulatory agencies, including California. Surface and groundwater sampling protocols have also been developed; analytical detection methods include high-performance liquid chromatography (HPLC) and tandem mass spectrometry (TMS). U.S. EPA analytical methods, specifically Method 533 for drinking water, can now determine 14 branched and linear isomer and 11 unique PFAS compounds. This method has detection and reporting limits in the ng/L (parts per trillion) range and because of the very low concentrations requiring a precise and accurate analysis that includes isotope dilution analysis (IDA) usage.

So why is IDA done and why is it important? First, IDA: (1) allows for accurate recovery correction and (2) normalizes instrument performance across different matrices. Second, and more importantly, IDA methodology allows accurate determination of the amount or quantity of an element or chemical compound by adding known amounts of an isotopically-enriched substance (either a stable or radioactive isotope) or standard to the sample to be analyzed (see Figure 1). Mixing of an isotopic standard with the sample essentially “dilutes” the sample’s isotope composition. Therefore, this is also considered as a method of internal standardization, because the standard (an isotopically-enriched form of analyte) is added directly to the sample. Additionally, unlike traditional analytical methods relying on signal intensity, IDA employs signal ratios. Therefore, IDA is regarded as one of analytical chemistry’s measurement methods having the highest metrological standing.

IDA is almost exclusively used with TMS

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Research was omitted from landmark paper claiming natural origin of SARS-CoV-2

Published: 26 May 2020 GM Watch Report: Claire Robinson

Chinese and US scientists have been collaborating for years in dangerous gain-of-function experiments that involve genetically engineering coronaviruses from bats and other animals, as revealed by a series of scientific publications. The coronaviruses are related to the SARS viruses that cause severe respiratory diseases in humans. The scientists were based at the Wuhan Institute of Virology (WIV) in China, the lab suspected by some of accidentally releasing the SARS-CoV-2 virus that caused the COVID-19 pandemic, and at the University of North Carolina (UNC) in the US.

Oddly, however, this long and high-profile research history was entirely omitted from the scientific paper, published in *Nature* in February this year, in which Shi Zhengli and her team at the WIV claimed to have identified a natural origin for SARS-CoV-2. The origin, according to the WIV scientists, was a bat virus, RaTG13, that was thought to have jumped from an animal to a human at a Wuhan seafood and wildlife market (the “zoonotic” theory – that is, coming from animals by a natural spillover event).

Why the omission? To understand the possible reason, we need to first understand the nature of the research work that was done by the WIV scientists and their US collaborators.

The purported benign aim of this line of research was to investigate the potential of bat coronaviruses to infect humans, to improve scientists’ ability to predict pandemics, and to develop vaccines or other therapies.

However, this is also gain-of-function research, which aims to make viruses more infective or transmissible. Such research has come under increasing criticism by scientists for many years, due to its tendency to pose huge risks for little benefit. This fear is borne out by the results of a particularly risky gain-of-function experi-

ment carried out in the US and published in 2015 by scientists from the UNC in collaboration with WIV scientists, including Shi Zhengli, dubbed China’s “bat woman” for her work with bat coronaviruses. The work was funded by: The National Institute of Allergy & Infectious Disease (NIAID) of the US National Institutes of Health (NIH). These funds went to the U.S. Agency for International Development (USAID) and Chinese Institutions. The director of the NIAID is Dr Anthony Fauci, who currently heads up the US COVID-19 response. The NIH’s money was directed through the US-based Eco-Health Alliance, headed by Dr Peter Daszak;

In the published paper reporting the risky experiment, the scientists state that they began their work before the 2014 US temporary moratorium on virus gain-of-function studies, which was prompted by several high-profile biosafety failures at US labs. But in spite of the moratorium, as stated in the paper, the NIH gave permission for the study to continue. Dr Fauci of the NIAID “outsourced” the research to the WIV in China, in the words of one media article.

In the experiment, the scientists took a mouse coronavirus and exchanged its spike protein – the part on the surface of the virus that determines infectivity – for one from a bat coronavirus that was similar to the virus that causes the human epidemic disease SARS. They kept the mouse virus “backbone” – its basic RNA and protein molecular structure. The bat coronavirus, in its natural state, was unable to infect humans as its spike protein was inadequate – it was not able to dock onto the ACE2 receptor on human cells.

Infectivity is supposed to be determined just by the spike protein. So joining the bat spike protein with the mouse virus back-

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bone should have resulted in a virus that was non-infectious to humans.

But the resulting genetically engineered chimeric virus unexpectedly turned out to be highly infectious to humans. In fact, its infectivity, tested in human airway cells, was comparable to the human epidemic-causing virus strain SARS-CoV Urbani.

The scientists were clearly surprised and alarmed by this finding. As they state, “based on previous models of emergence”, the creation of this chimeric virus “was not expected to increase pathogenicity”. They deduced that the nature of the spike protein alone was not enough to determine infectivity – the backbone of other protein components is also important.

The researchers then tried – but failed – to develop a vaccine or antibody therapy. The antibodies were unable to block the receptor binding domain (RBD – the part of the spike protein that binds to the human ACE2 receptor, resulting in infection) of the bat-mouse chimeric virus.

The researchers conclude their publication with a caution and a question left hanging in the air. They write that their findings “represent a crossroads of GOF [gain-of-function] research concerns; the potential to prepare for and mitigate future outbreaks must be weighed against the risk of creating more dangerous pathogens. In developing policies moving forward, it is important to consider the value of the data generated by these studies and whether these types of chimeric virus studies warrant further investigation versus the inherent risks involved.”

In short, the very research that is claimed by some to be necessary to develop vaccines and other interventions risks creating a pandemic.

While there are serious risks involved in carrying out such research, there are also risks involved in publishing it. In this case the researchers examined the amino acid sequences of the bat virus spike protein and identified the sequences required for human infectivity – and published information on them in their paper.

The London-based molecular geneticist Dr Michael Antoniou commented, “The information on amino acid sequences pro-

vided in this paper is crucial to designing a virus that is infective in humans. Anyone with access to a standard laboratory would be able to use the information to estimate the amino acid sequence needed to engineer an RBD that would be highly likely to infect human cells.”

In other words, the researchers have provided a guide to making a bioweapon.

Dr Antoniou explained how their data makes what would otherwise have been a laborious process far quicker and more efficient. If you start with no information, you could engineer a human-infective virus like SARS-CoV-2 by using a “directed iterative evolutionary selection process”. This would involve using genetic engineering in a mutagenesis procedure to generate a large number of randomly mutated versions of the SARS-CoV spike protein RBD, which would then be selected for strong binding to the human ACE2 receptor and consequently high infectivity of human cells.

However, using the information provided by the UNC and WIV researchers, Dr Antoniou says, “You don’t have to go in blind using a total ‘saturation’ mutagenesis of the RBD amino acid sequence. You don’t have to start from a black box of unknowns. You already have an insight into which amino acid sequence is needed for human infectivity, so that guides you as to how to engineer the virus.”

This raises the ethical question of whether gain-of-function research is ever worth the risk. Dr Antoniou believes that it is not: “Research of this type is not necessary to identify new targets for therapeutic intervention. An investigation of the basic mechanisms of how virus infection takes place and progresses is sufficient for this. Thus gain-of-function research with known dangerous pathogens such as coronaviruses should be banned.”

In spite of the dangers highlighted in the 2015 paper, and in the wake of the US temporary moratorium on virus gain-of-function work, the research with bat coronaviruses continued – this time in China. In 2017 WIV scientists, including Shi Zhengli, published a study with funding shared

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between Chinese and US institutions, the latter including the US NIH and USAID.

The researchers report the findings from virus infectivity experiments where genetic material was combined from different SARS-related coronaviruses to form novel chimeric versions. They were trying to find out which mutations were needed to allow certain bat coronaviruses to bind to the human ACE2 receptor. They found that two genetically engineered chimeric viruses replicated “efficiently” in human cells. The consequences of escape of such viruses could be serious.

Then, only this month, WIV scientists led by Shi Zhengli published a pre-print reporting work in which they investigated the ability of spike proteins from bat SARS-related coronavirus (SARSr-CoV), among other coronaviruses, to bind to bat and human ACE2 receptors. In other words, they examined how efficiently these coronaviruses infect humans and how human infectivity can be optimised.

The three papers examined above show that over a period of several years, Chinese and US scientists were using genetic engineering techniques for gain-of-function experiments with coronaviruses, resulting in the generation of viruses better adapted to infect humans.

Against this background Shi Zhengli published her landmark paper in the journal *Nature* in February this year, after the COVID-19 pandemic had spread across the globe. In this paper, Shi and her co-authors claimed to have identified the closest relative to SARS-CoV-2 and its “probable” origin, a natural bat coronavirus, which she called RaTG13. The paper highlights the natural origin zoonotic theory for SARS-CoV-2 – that it jumped from an animal into humans at the Wuhan seafood and wildlife

market. This theory has not subsequently been supported by emerging evidence.

All publications arguing for a natural origin for SARS-CoV-2 rely heavily on this one paper by Shi Zhengli and colleagues, describing the sequence of a purported natural bat coronavirus named RaTG13. But notably absent from the paper is any reference at all to Shi and her collaborators’ long history of gain-of-function genetic engineering research with bat coronaviruses, described above. That includes the important paper by UNC and WIV scientists of 2015, which had the alarming result of turning a harmless bat virus into a human pathogen.

It is as if this research background simply didn’t exist. Why? Could it be because drawing attention to it might raise the suspicion in people’s minds that SARS-CoV-2 might also have been intentionally or accidentally optimised in the lab during gain-of-function research?

After all, if the belief gained traction that the virus might have escaped from a lab, virologists could expect their research to be “impacted adversely by tighter laboratory controls”, as the leading vaccine researcher Professor Nikolai Petrovsky has pointed out in explaining why the majority of scientists seem to be supporting the idea that the virus originated in a wet market rather than a lab.

It would also, of course, almost certainly bring the “gravy train” of virus gain-of-function research to an abrupt halt, quite apart from causing a massive political storm. It might even awaken public doubts about the safety of other risky applications of genetic engineering.

But despite this array of vested interests, a forensic investigation needs to begin as soon as possible into the exact origins of a pandemic virus that, in the words of Professor Petrovsky, seems “like it was designed to infect humans”.



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analyses for applications where a high degree of accuracy is required; however, it can also be employed with every type of MS used in different environmental analytical fields and standards (e.g., all National Metrology Institutes rely on IDA when producing certified reference materials). For high-precision analysis, IDA is applied when an analyte's low recovery is required. In addition to stable isotopes usage, radioactive isotopes can also be employed; this is often required in biomedical applications (e.g., in estimating blood volume).

Because IDA involves same element isotopic measurements, differences in chemical compositions are eliminated. IDA's major advantage is that during the entire analytical process including sample preparation, analyte separation, and sample enrichment, no analyte quantitative recovery is necessary once equilibration occurs between the spike and sample. Therefore, when compared

to other analytical methods, IDA is more stable and less error-prone during chemical processing steps.

For trace and ultra-trace analysis, different elemental species accuracy is essential. For such analyses, IDA, allows for adding one, or two, highly enriched isotope tracers or spikes (the two spike method also known as the double-spike technique). This is accomplished by adding an element, with well-known concentrations to the sample, which is then mixed and homogenized with the solid sample or aqueous solution. Determination of the trace element concentration is then performed by measuring the change in the isotope ratios in the sample-spike mixture compared to those in the sample and highly enriched isotope tracer.

So, the next time you submit a sample for PFAS analysis using IDA, remember that the returned analytical data will have greater accuracy and therefore reliability than other methods.

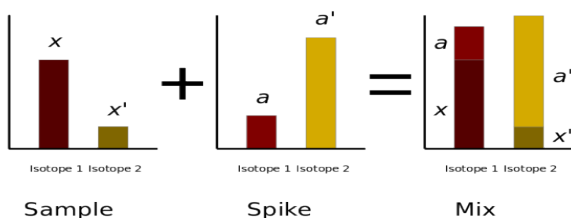


Figure 1: IDA basic principles require adding an isotopically altered standard to a sample (one spike method). This changes the analyte's natural isotopic composition and by measuring this change one can accurately calculate the amount of the analyte present in the sample.

Source:

https://en.wikipedia.org/wiki/Isotope_dilution

Help from ACS in a COVID-19 World

Marinda Li Wu, ACS Career Consultant

Most of us have been sheltering in place and working from home since March 16. In case you have not heard, ACS has been offering various new Member Services for the first time to help members as we all adjust to this new COVID-19 world.

ACS Webinars are a free member benefit. Interesting webinars on all kinds of topics used to be available once a week. Now these great webinars are available daily. Take a look at www.acs.org and search for the ACS Webinars.

Hundreds of ACS webinars presented by subject matter experts in the chemical enterprise are available in the archive along with live webinars coming up. Check it out for some fascinating topics including several on this new corona virus and how it differs from other viruses.

“Navigating Your Chemistry Career in a COVID-19 World” was recently presented as an ACS Webinar by Joe Martino, a fellow ACS Career Consultant. His talk is now available in the ACS Webinar Archive. I

found it full of good tips and suggestions. Joe, along with over seventy other ACS chemists, are certified as ACS Career Consultants.

Did you know that you can sign up online for a personal ACS Career Consultant to discuss your career and future options? You can get help with your resume, job search, and preparing for interviews (many now virtual).

Visit www.acs.org/careers to select your own ACS Career Consultant in various fields of expertise to offer career tips and suggestions to help you navigate in this new COVID world.

ACS Career Services is also now offering for the first time Virtual Office Hours for members to sign up to talk with ACS Career Consultants. I participated in the launch program where both ACS career consultants and job seekers tested using ZOOM technology for virtual consultations and break out rooms. Visit www.acs.org/careers for more information.

The CALACS Senior Chemists are also offering ZOOM training for all members interested. Visit www.calacs.org for more details on other activities for our California Section despite this new corona virus.





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