Mycena News

The Mycological Society of San Francisco February, 2011, vol. 62:02

February 15th MSSF Meeting Speaker



Thomas S. Jenkinson Mushrooms and the Micronesia Biodiversity Project

Epacific islands. Thomas Jenkinson will highlight recent results from the ongoing project to document the mushroom diversity of the Federated States of Micronesia. Travel to the islands of Pohnpei and Kosrae, Micronesia, within the Micronesia/ Polynesia biodiversity hotspot for conservation. The habitat these islands provide for unique organisms is under increasing threat, and forest habitats on these islands have been lost at an alarming rate in the last few decades. The Micronesia Biodiversity Project incorporates information from surveys of plant, invertebrate and mushroom diversity in investigating the effects of deforestation on these islands. This presentation will focus on the new and interesting mushrooms documented over two seasons of field surveys in 2009 and 2010.

Thomas Jenkinson is a council member of the Mycological Society of San Francisco. He received his M.S. in Biology from San Francisco State University, and is currently a lecturer and laboratory co-coordinator of the introductory biology laboratory course at San Francisco State University. Thomas has collaborated on research projects in mycology in California and around the world.

MycoDigest: Engineered Fungi and the Mushrooming Production of Biofuels

Todd Osmundson

As the price of petroleum rises and the geopolitical and environmental consequences of oil addiction grow increasingly evident, markets and political support for 'biofuels' – fuel products derived from biological organisms – are growing rapidly. With their prodigious capacity for enzymatic magic and chemical conversion, fungi play a central role in biofuel production, and

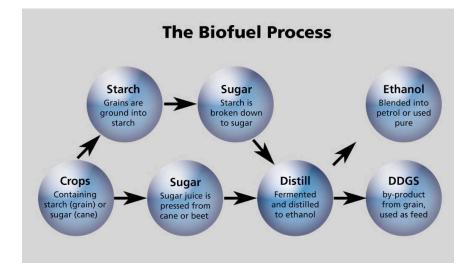


Diagram of the ethanol (1st-generation biofuel) production process (http://www.labnews. co.uk)

recent efforts in genetic, cellular and metabolic engineering (together part of the field known as "synthetic biology") have yielded fungal strains that may improve both efficiency and the range of plant materials that can be used in commercial processing.

Currently, the most commonly used biofuel is ethanol derived from plant materials. Ethanol is the product of ethanol fermentation, a process by which some organisms gain energy from sugars, producing ethanol and carbon dioxide as byproducts. Ethanol fermentation is a well-practiced procedure, the heart of brewing and baking. Sugars from plant materials such as grains, grapes, sugar cane, or potatoes are fermented, and the ethanol and carbon dioxide are either used or released. In brewing, the ethanol is harvested, and the carbon dioxide is either retained (producing the carbonation present in some alcoholic beverages) or released. In baking, the carbon dioxide causes bread and other baked goods to rise, and the ethanol is evaporated away during baking. The process of making ethanol biofuel is fundamentally similar to brewing, and the tool that enables the production of alcoholic beverages – the brewer's yeast, *Saccharomyces cereviseae*

MycoDigest is dedicated to the scientific review of mycological information.

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President's Post

February begins a change in our season with longer days and our favorite winter fungi. MSSF had several great forays last month starting with a beginner's foray led by Pat George and Alice Sunshine in the east bay. Given the very foggy conditions we still had a great turnout and collected many interesting species.

On the same weekend J.R. Blair and Bill Freedman led the annual Mills Canyon foray in Burlingame. You can see photos of this foray from the photos link on the Website, look for the Phleger Estate 1/8/11 photo album.

The third foray of the month, Al Carvajal's San Mateo Amble, is in progress as I write this.

Speaking of photos, we have started to have more photos submitted by members. This makes a great addition to the site, since having photo content allows members and potential members to see what MSSF is up to. "A picture says a thousand words," as they say. I encourage each of you to try to upload at least one photo to the site this month. Uploading is easy and if you have trouble I will personally help you. Please contact me by email if you have a large number of photos to post and we can upload your images as a batch to save you time. As an added attraction your photos may appear in future issues of the Mycena News!

Several other great mycological events happened around the Bay area in January. Many of our members attended the Santa Cruz Fungus Fair put on by the Fungus Federation and a number of MSSF'ers were also at the always great SOMA camp as I can personally attest.

We are still actively seeking a volunteer for the council position of Librarian. The library has been freshly re-organized and the library catalog is now available on the MSSF website so this great club asset is ready for a new caretaker. We will be glad to help you get up to speed so don't delay, contact me today !

In closing I thank you for your ongoing support and I encourage you to contact me if you have questions, suggestions, or want to talk about the council positions the Society is presently trying to fill. Stay tuned to the calendar for upcoming forays soon to be announced.

Hope to see you all soon.

-Lou president@mssf.org



Culinary Corner

Despite the rumor that the mushroom season is fast winding to a close, lots of fungi have been showing up as of late January. Especially prominent amongst the culinary fungi have been yellowfeet (*Cantharellus tubaeformis*) and black trumpets (*Craterellus cornucopoides*), called "trompettes des morts" by the French because they have a funereal, somber color, not because they have any sinister effects on one's health. Plenty of self-satisfied looking mushroomers came into the big room at the SOMA Camp with baskets of Craterellus. Unfortunately I missed the rich harvests. Fortunately dried ones can be reconstituted and used in this recipe. It is still chilly at night in February and a rich, hearty soup served with a very good bread makes life cheerier. Here's a recipe from France using an ancient grain, Triticum spelta, called Spelt.

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Cream of Spelt Soup with Black Trumpet Mushrooms

Ingredients for 4 generous servings:

3/4 cup spelt
1 1/2 cup of water
2 T. olive oil
1 small onion, diced
1 carrot, diced
1 leek, white part only, diced
1 tender rib celery, diced
bouquet garni of thyme and bay leaf
6 cups of rich chicken or turkey stock
1 T. olive oil
1 small shallot, finely chopped
1/3 to 1/2 pound of cleaned Craterellus pulled into filaments
leaves from one branch of thyme
4 T. of creme fraiche

2 T snipped chives

Combine the spelt with the water, bring to a boil, lower the heat, cover and simmer for about 30 minutes or until done.

In a medium large casserole, heat 2 T. olive oil, add the leek, onion, carrot and celery. Cook over medium low heat until soft but not colored. Add the stock, the cooked spelt and the bouquet garni and simmer for 30 minutes. Remove the herbs and pass the soup through the fine blade of a food mill. Return to the casserole, whisking to smooth. Heat gently, adding a bit more stock if the soup is too thick, and correct the seasoning.

In a medium heavy skillet, heat the olive oil and saute the shallot for a few minutes. When soft, raise the heat to high and add the mushrooms. Cook, stirring delicately, until they give up their liquid and it evaporates. Salt lightly. Cooking time will be about 4 or 5 minutes. Scape the mushrooms to the side of the skillet and drop the thyme leaves onto the bare spot. When they pop, stir them into the mushrooms, remove from heat and correct the seasoning.

Ladle the soup into warmed bowls. Float the mushrooms on top (divided into 4 portions). Top with a dollop of creme fraiche and a sprinkle of chives.

Note: Other kinds of mushrooms including chanterelles, shiitakes, morels or cepes can be used. Oyster mushrooms are a bit too bland for this recipe.

The MSSF Culinary Group started the New Year with an Italian winter repast which featured wild boar ragu over polenta with cheese, a beautiful salad, panna cotta for dessert and rich coffee. Thanks to all you cooks: Sandy of the salad fame, Norm, Lisa G., Lou, Lisa B for the ragu and polenta, Honoria and Monique for panna cotta and our faithful Remo for the coffee. Of course, we had our grand raffle, too, with Curt at the spinning cage. Our dinners are delicious and fun! See the website for details on how you can join us. And for the menu and other details, of course. - Pat

What's Bookin'?

Larry Stickney's book donation to the MSSF

Larry Stickney wished that his book collection be left to the Mycological Society of San Francisco. Recently, Monique Carment, (MSSF Librarian) and I spent many hours sorting through the numerous books Larry donated. The majority of the books donated were duplicates of books already listed in our extensive library. However, we were able to identify approximately 30% of them that were not duplicates and they have been added to the library.

The remainder of the donated books, and publications that were recently

purged from the library were included in the used book sale that was held at the January general meeting. The MSSF library has outgrown our current storage space, so new cabinets have been procured to assist in storing future book purchases, donations, plus merchandising and hospitality supplies. The used book sale raised over \$560.00 in needed funds to help offset the added cost of the new cabinets.

A list of the books donated by Larry will be available for members to view in the MSSF "File Archives" section of the website where they will be included in the minutes of the February council meeting. MSSF members can also contact me at: <u>lingking@sbcglobal.net</u> and I will e-mail a copy of the list to you.

Larry's books that were selected for inclusion in the library, plus new books reviewed over the last year, will soon be cataloged and numbered, and will be available for check out from the MSSF library.

~ Curt Haney





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Past issues of *Mycena News* can be read online at www.mssf.org.

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MycoDigest continued

- is also one of the primary tools of choice for producing ethanol biofuel. However, the scale of fuel demand and the particular characteristics of fuels and fuel production present several problems that must be addressed in order for biofuel production to become more efficient and environmentally sustainable.

The first problem is that S. cerevisiae is very good at fermentation - i.e., converting simple sugars (monosaccharides and disaccharides, composed of single sugar molecules or pairs of sugar molecules, respectively) to ethanol but relatively few of the carbohydrates in most plants occur in these forms. Some plants, such as sugar cane (widely used in Brazil for biofuel production) contain abundant simple sugars and are therefore relatively good biofuel sources; others, such as corn (the biofuel plant of choice in the United States) are highly inefficient sources. In most plants, most of the carbohydrates are tied up in the form of starch or cellulose, long chains of sugar molecules that function in storage and structure, respectively; the latter often occurs with several other compounds including other carbohydrates (hemicelluloses, pectins), proteins, and extremely recalcitrant (difficult-to-digest) lignin. The process of making biofuels thus is a 3-part process: digesting longchain carbohydrates into simple sugars that can be fermented, followed by fermentation itself, followed by recovery and chemical conversion of the fermentation products to usable fuels. Saccharomyces is good at the fermentation step, but not at the first step. Present methods use mechanical (e.g., heating), chemical (e.g., treatment with dilute acids), or enzymatic treatments to break down the plant material, but these methods are relatively expensive and time-consuming. Here, fungi also play a role, as the enzymes used for these treatments are derived from plant-degrading fungi

A second problem is that *S. cerevisiae* is a relatively picky eater. It has a strong preference for glucose (it is also capable of fermenting galactose and mannose), but leaves other sugars alone. One of these ignored sugars is xylose, (a.k.a., "wood sugar"), a common component of wood and other high-lignocellulose materials. The ability to ferment xylose and other sugars would result in substantial increases in fuel production efficiency.

A third problem with current production methods is that the use of corn as a biofuel presents not only issues of inefficiency, but more serious societal and environmental issues as well. The success of the biofuel industry in the United States (much of this success a direct result of generous government subsidies) has led to a market shift where a significant amount of corn production has been diverted from food to biofuel markets. This shift has led to higher food prices and concerns over food scarcity [1]. The market shift has also set off a chain of events where conversion of American croplands to corn production for ethanol has displaced soybean production to Brazil, leading to widespread clearing of biodiverse Amazon rainforest and endangered Cerrado forest to make way for agricultural cultivation there. Two recent studies have shown that, when calculations account for the amount of greenhouse gasses released by additional land clearing to augment or compensate for biofuel crop production, the carbon sequestration benefits of corn ethanol are negative compared to fossil fuels [2, 3]. The high amounts of agricultural inputs (fertilizers, tractor fuel, water, etc.) necessary to grow corn exacerbate this problem, and habitat loss may have severe impacts on biodiversity. The efficiency, food security, climate change, and biodiversity concerns related to ethanol fermentation have inspired the search for methods of producing fuels from the more recalcitrant carbohydrates - i.e., "cellulosic fuels" or "2nd-generation biofuels." These fuels not only include the recalcitrant components of current biofuel plants such as corn, but also other agricultural byproducts and plants that require low fertilizer and water inputs that can grow on marginal or degraded agricultural lands. A number of alternative cellulosic fuels, including a number of native grass species and algae, are currently being researched.

A fourth problem is that ethanol is not optimal as a fuel. It has a relatively low energy density (the amount of energy released per unit weight of fuel; an especially important consideration in flight applications), is corrosive, and has a strong tendency to absorb water; these characteristics make distribution, transportation and storage more complicated [1, 4]. It is therefore quite apparent that a number of issues stand in the way of using biofuels as a viable replacement for petroleum-based fuels. In response to these issues, a significant amount of research effort has been expended toward engineering *S. cerevisiae* (and also the bacterium *Escherichia coli*) in the interest of (i) improving the efficiency of complex carbohydrate breakdown ("saccharification"); (ii) improving fermentation efficiency and increasing the diversity of sugars that can be fermented; and (iii) developing non-ethanol biofuels.

The two major pathways that have been taken toward improving the efficiency of complex carbohydrate breakdown are to either improve the speed, efficiency, and cost-effectiveness of pre-fermentation processing, or to engineer yeast cells that can carry out both the saccharification and fermentation steps. On the pre-fermentation processing side, one of the most interesting recent advances is a study by researchers at Caltech and the company DNA2.0 in which they took gene sequences encoding known fungal lignocellulose-degrading enzymes and used computer-based tools and simulations to combine various parts of these gene sequences and infer the structure, function, and stability of the resulting recombinant enzymes. They then engineered S. cerevisiae cells to express these chimeric genes and tested their compatibility (i.e., the ability of S. cerevisiae cells to express the protein in its active form), their thermotolerance (ability to withstand the relatively high temperatures and extended time periods used in industrial processing), and level of ability to hydrolyze cellulose. The researchers created 15 recombinant proteins with higher thermostability than the most stable natural enzyme, as well as proteins with higher temporal and pH stability [5]. The diverse chemistry of plants means that different enzymes will perform better in different applications; the approach used in this study identified hundreds of additional potential enzymes that have not yet been tested. Industrial applications often use more than one enzyme to achieve optimal results; the addition of a diverse set of recombinant enzymes to the approximately 20 known natural fungal enzymes is therefore likely to significantly increase the range of substrates and reaction conditions that can be used. The second pathway, unifying the saccharification and fermentation steps (a process known as "simultaneous saccharification and fermentation," or SSF) has also seen some notable successes, such as a recent study by researchers from UC-Berkeley and the Lawrence Berkeley Labs in which genes from the cellulose-degrading fungus Neurospora crassa were expressed in S. cerevisiae, allowing the yeast to import and metabolize cellodextrin - a midpoint product of cellulose degradation - thus representing a first step toward achieving SSF [6].

Improving the diversity of sugars that S. cerevisiae can ferment is an important element in the quest for more efficient ethanol production. Similar to the cellodextrin example, synthetic biology has been used to combine the desired features of more than one organism. Saccharamyces cerevisiae can tolerate ethanol well but most strains cannot ferment xylose; some strains (particularly winemaking yeasts) have recently been shown to have xylose fermenting ability, but are not particularly efficient at the task [7, 8]. In contrast, Scheffersomyces stipitis (formerly known as Pichia stipitis) is an outstanding xylose fermenter but is inhibited by ethanol. Engineering of S. cereviseae cells with xylose metabolism genes from S. stipitis and other fungi has produced effective xylose fermenting cells in a number of studies [e.g., 9, 10]. By adding additional genes from the fungi Trichoderma reesei and Ambrosiozyma monospora, a team at Purdue University recently generated S. cereviseae cells that can ferment arabinose (another sugar common in cellulosic breakdown products) in addition to glucose, galactose, mannose, and xylose [11]. Another recent paper reported the dual-engineering of S. cereviseae with both the xylose degradation genes from S. stipitis and the cellodextrin transport genes from N. crassa [12]. Cofermentation of sugars and SSF are complicated by a number of factors, including the much slower rate of cellulose hydrolysis than fermentation [1] and the inhibitory effects of glucose on xylose fermentation [12]; careful optimization and continued effort will therefore be required for production-level results. Metabolic engineering in general is complicated by the interconnectedness of metabolic pathways; in some cases improvements in a biofuel synthesis pathway may cause changes in other pathways such that yield is ultimately reduced. The use of in silico (computerized) metabolic models has shown promise in predicting such effects and designing alternatives [13].

Given its significant downside, ethanol is considered by many biofuels researchers to be a temporary fix. Although scientists still busily at work on improving the efficiency of ethanol fermentation, there is also a significant amount of research on "3rd-generation biofuels" that have more desirable chemical characteristics. A diverse number of alternative fuels are under development, and have the potential to viably replace gasoline, diesel, and jet fuels; the development of these fuels for commercial-scale production is inextricably linked to advances in synthetic biology, and the near future will no doubt yield a substantial crop of new developments in genetic, cellular and metabolic engineering as biofuels research continues [1, 4, 13-16]. In terms of natural biofuel sources, a recent study reported that an endophytic fungus isolated from a Patagonian tree produces short-chain hydrocarbons such as those found in biodiesel; the authors suggest that such fungi have the potential to produce "myco-diesel," to be used as biofuel [17]. However, an independent assessment of the finding expressed skepticism about the possibility of production-scale yields without bioengineering [18].

The level of research activity and both governmental and corporate interest in biofuels indicate that widespread production in the near future is nearly certain. But will biofuels fulfill their promise of environmental sustainability? One factor that does not seem to enter the current discussion is the prospect that production of cheaper fuels would create additional demand beyond simply replacing existing fossil fuel demand, much in the same way that highway expansion projects - intended to reduce traffic congestion - tend to attract increased numbers of motorists by facilitating access. The sharp rise in gasoline prices in the summer of 2008 and their subsequent fall - and the concurrent fall and rise in automobile usage, respectively - illustrate an important yet basic economic concept: price and demand are interconnected. There is little reason to expect that demand will decrease or stay constant if fuel is more plentiful and less expensive. And although 2nd- and 3rd-generation biofuels may themselves be carbon-neutral, the associated industrial and transportation infrastructure will still bear significant environmental costs. It is also unclear whether current cultivated land area and additional degraded or marginal agricultural lands will be able to produce sufficient biomass to meet increased demand, or whether additional land clearing would occur. The amazing advances in engineering S. cereviseae are clearly good news for the biofuel industry and the goal of national energy security, but the benefits of these modified fungi for their forest-inhabiting relatives are much less certain.



Todd Osmundson is a postdoctoral researcher in the department of Environmental Science, Policy and Management at the University of California, Berkeley with research interests in fungal systematics, biodiversity, evolution, ecology and conservation.

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MSSF Calendar February 2011

February 7th: February Culinary Dinner

Special Mardi Gras theme with Chef Edwin Caba. 7PM at the San Francisco County Fair Building, Golden Gate Park. Reservations required no later than February 4, 2011 with a strict limit of 60 so don't delay.

February 15th: Speaker Thomas Jenkinson

This month's General Meeting will feature council member and San Francisco State lecturer Thomas Jenkinson who will discuss the mushrooms of the remote Pacific islands. 7PM Randall Museum.

Call for Volunteers

We have one remaining volunteer position to fill:

Librarian Chairperson

The library has been purged, revised and organized. The catalog is now available on the MSSF website, and this great club asset is ready for a new caretaker.

Don't be shy! Monique Carment will help you get started. Remember, our great organization would not survive without volunteers.

If you are interested in volunteering for this position contact Lou Prestia at: <u>President@mssf.org</u>

Check the MSSF online calendar at: http://www.mssf.org/calendar/index.php for full details, latest updates and schedule changes. The submission deadline for the March, 2011 issue of Mycena News is Sunday, February 20th. Please send your articles, calendar items, and other information to: mycenanews@mssf.org

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