July 2009
Volume 7, Issue 7

The Herbal Dispatch

The Medicinal Botanicals Program
Mountain State University (304) 929-1630

ISSN 1548-6052 (Print)
ISSN 1548-6044 (Electronic)

MSU and USDA, ARS Held Successful Workshop

On July 11, 2009, the Mountain State University Medicinal Botanicals Program (MSU-MBP), Beckley, WV, and the USDA, ARS, Appalachian Farming Systems Research Center (AFSRC), Beaver, WV, held a workshop and research update on improving small ruminant grazing practices for Appalachia. The day-long event was held at the AFSRC facilities. The event started with an introduction by Dr. Joyce Foster, AFSRC Research Biochemist, followed by a presentation on finishing small ruminants on pasture by Dr. Ken Turner, AFSRC Research Animal Scientist. Dr. Jim Neel, also an AFSRC Research Animal Scientist, discussed the development and use of silvopasture for small ruminant grazing in Appalachia. Dr. Kim Cassida, AFSRC Research Agronomist, talked about non-traditional forages for use in central Appalachia.

USDA-ARS scientists Dr. Ken Turner (far left), Dr. Joyce Foster (with name tag) and Dr. Jim Neel (third from right) discuss aspects of small ruminant production with some workshop participants during the tour of the AFSRC research farms.

After a coffee break at mid morning, Dr. Foster spoke about opportunities to control *Haemonchus contortus*, an economically important gastrointestinal parasitic nematode of small ruminants, using selected plant constituents. She was followed by Dr. Mario Morales, Director of the MSU MBP, who discussed the cultural and nutritional qualities of American potato bean (*Apios americana* Medikus) and its potential as forage or dietary supplement for sheep and goats. The final presentation was given by Dr. Jorge Ferreira, AFSRC Research Horticulturist, who spoke about potential applications for Artemisia in small ruminant production.

After lunch, participants visited the AFSRC research farms where they had the opportunity to observe research plots containing condensed-tannin-containing legumes, a meat-goat grazing trial using traditional and non-traditional forages, and a study that compares forage and small ruminant production on silvopasture and traditional pasture. The farm tour provided first-hand exposure to many of the plants and procedures described during the morning session and enabled participants to interact among themselves and with the researchers. Attendees expressed enthusiasm for information presented during the course of the event and interest in implementing some of the techniques that were demonstrated. To enhance the learning experiences, the MSU-MBP produced a workshop proceedings, Improving Small Ruminant Grazing Practices, that was distributed to participants, A copy of this publication, on CD, can be obtained by submitting a request to Dr. Mario Morales, MBP, MSU, 410 Neville Street, Beckley, WV 25801-4511. Requests can also be submitted by email to mmorales@mountainstate.edu or by phone (304-929-1683). Alternatively, the Proceedings can be downloaded from the MSU-MBP web site at http://www.mountainstate.edu/usda.

The Herbal Dispatch

A monthly publication of the Medicinal Botanical Program

The goal of this newsletter is to inform readers of the Program's educational, research and outreach activities and events; and of results of the latest research on the chemistry, cultivation, processing and preventive and therapeutic use of botanicals.

The views expressed in The Herbal Dispatch are those of the authors and do not necessarily reflect those of MSU or the Medicinal Botanical Program staff. Authors are solely responsible for their articles.

Mario R. Morales
Editor/Publisher

The Soil Biology Primer-Part VII

By Elaine R. Ingham
Oregon State University

Chapter 3: BACTERIA

THE LIVING SOIL: BACTERIA

Bacteria are tiny, one-celled organisms – generally 4/100,000 of an inch wide (1 µm) and somewhat longer in length. What bacteria lack in size, they make up in numbers. A teaspoon of productive soil generally contains between 100 million and 1 billion bacteria. That is as much mass as two cows per acre.

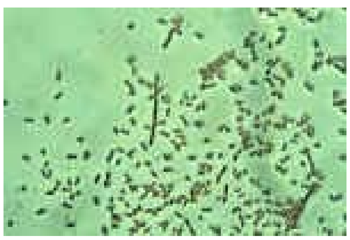

Figure 1: A ton of microscopic bacteria may be active in each acre of soil. ***Credit***: *Michael T. Holmes, Oregon State University, Corvallis.*

Figure 2: Bacteria dot the surface of strands of fungal hyphae. ***Credit:*** *R. Campbell. In R. Campbell. 1985.* Plant Microbiology*. Edward Arnold; London. P. 149. Reprinted with the permission of Cambridge University Press*

Bacteria fall into four functional groups. Most are decomposers that consume simple carbon compounds, such as root exudates and fresh plant litter. By this process, bacteria convert energy in soil organic matter into forms useful to the rest of the organisms in the soil food web. A number of decomposers can break down pesticides and pollutants in soil. Decomposers are especially important in immobilizing, or retaining, nutrients in their cells, thus preventing the loss of nutrients, such as nitrogen, from the rooting zone.

A second group of bacteria are the *mutualists* that form partnerships with plants. The most well-known of these are the nitrogen-fixing bacteria. The third group of bacteria is the *pathogens*. Bacterial pathogens include *Xymomonas* and *Erwinia* species, and species of *Agrobacterium* that cause gall formation in plants. A fourth group, called *lithotrophs* or *chemoautotrophs*, obtains its energy from compounds of nitrogen, sulfur, iron or hydrogen instead of from carbon compounds. Some of these species are important to nitrogen cycling and degradation of pollutants.

What Do Bacteria Do?

Bacteria from all four groups perform important services related to water dynamics, nutrient cycling, and disease suppression. Some bacteria affect water movement by producing substances that help bind soil particles into small aggregates (those with diameters of 1/10,000-1/100 of an inch or 2-200µm). Stable aggregates improve water infiltration and the soil's water-holding ability. In a diverse bacterial community, many organisms will compete with disease-causing organisms in roots and on aboveground surfaces of plants.

A Few Important Bacteria

Nitrogen-fixing bacteria form symbiotic associations with the roots of legumes like clover and lupine, and trees such as alder and locust. Visible nodules are created where bacteria infect a growing root hair (Figure 4). The plant supplies simple carbon compounds to the bacteria, and the bacteria convert nitrogen (N2) from air into a form the plant host can use. When leaves or roots from the host plant decompose, soil nitrogen increases in the surrounding area.

Nitrifying bacteria change ammonium (NH4+) to nitrite (NO2-) then to nitrate (NO3-) – a preferred form of nitrogen for grasses and most row crops. Nitrate is leached more easily from the soil, so some farmers use nitrification inhibitors to reduce the activity of one type of nitrifying bacteria. Nitrifying bacteria are suppressed in forest soils, so that most of the nitrogen remains as ammonium.

Denitrifying bacteria convert nitrate to nitrogen (N2) or nitrous oxide (N2O) gas. Denitrifiers are anaerobic, meaning they are active where oxygen is absent, such as in saturated soils or inside soil aggregates.

Actinomycetes are a large group of bacteria that grow as hyphae like fungi (Figure 3). They are responsible for the characteristically "earthy" smell of freshly turned, healthy soil. Actinomycetes decompose a wide array of substrates, but are especially important in degrading recalcitrant (hard-to-decompose) compounds, such as chitin and cellulose, and are active at high pH levels. Fungi are more important in degrading these compounds at low pH. A number of antibiotics are produced by actinomycetes such as Streptomyces.

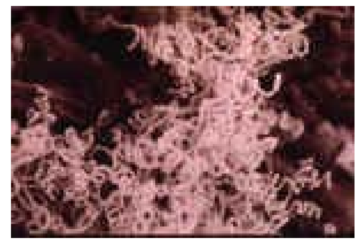

Figure 3: Actinomycetes, such as this Streptomyces, give soil its "earthy" smell. ***Credit:*** *No. 14 from* Soil Microbiology and Biochemistry Slide Set*. 1976. J.P. Martin, et al., eds. SSSA, Madison, WI*

Figure 4: Nodules formed where Rhizobium bacteria infected soybean roots. ***Credit:*** *Stephen Temple, New Mexico State Univ.*

WHERE ARE BACTERIA?

Various species of bacteria thrive on different food sources and in different microenvironments. In general, bacteria are more competitive when labile (easy-to-metabolize) substrates are present. This includes fresh, young plant residue and the compounds found near living roots. Bacteria are especially concentrated in the

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Wild Ginger (*Asarum canadense* L.)

By David C. Carman
Grower and Collector
Princeton, West Virginia

Wild ginger, a member of the birthwort family, and a native medicinal perennial, must not be consumed internally due to containing liver damaging aristolochic acid. Older references must be ignored in this respect, when referring to the plant being eaten raw, boiled with brown sugar and preserved as a candy, added to stews, made into tea, etc.

Also known locally as Canada snakeroot, Canada wild ginger, black snakeroot, Vermont snakeroot, Southern snakeroot, heart snakeroot, coltsfoot snakeroot, black snakeweed, snakeroot, as arum, Indian ginger, colicroot, colt's foot, false colt's foot, broad leaved asarabacca, cat's foot, and catfoot.

Wild ginger is quite common in our local woodlands where forest shade, rich humus soil, and soil moisture are present, and it is easily grown in similar garden settings when propagated by transplanting wild roots. They spread readily by self seeding, and form a dense, beautiful ground cover of large, heart shaped, dark-green leaves with petioles six to 12 inches tall.

A single purple/brown, foul smelling flower appears inconspicuously in early spring on a short pedicel in the crotch of two leaf petioles at ground level and, quite often, unseen under fallen leaf litter.

If you have a shade garden, I recommend growing a few wild ginger transplants. They are a beautiful native plant.

Ricin

Submitted by G. Sujatha and R. Balakrishnaraja

Dept. of Biotechnology, Bannari Amman Institute of Technology, Sathyamangalam – 638 401. Tamil Nadu. India

Ricin is a protein toxin that is extracted from the castor bean (*Ricinus communis* L.). It can be either a white powder or a liquid in crystalline form and is a very dangerous poison. The ricin protein is formed by the subunits A and B. Castor oil is found in many commonly used substances such as paints, varnishes, and lubricating oils, such as brake and hydraulic fluids. It is also used as a purgative. Its production spans the globe, but primary sources are India, China, and Brazil.

R. communis was cultivated for centuries in ancient Egypt and Greece for the lubricating and laxative effects of its oil. Both the oil and whole seed have been used in various parts of the world for disease treatment as well as for malicious mischief and homicidal purposes. Due to its excellent lubricating properties, castor oil was utilized by the aircraft industry during World War I. Castor oil shortages during World War II motivated the US government to subsidize agricultural production of castor beans in the San Joaquin Valley of California. These subsidies persisted until the 1960s, when synthetic oils replaced castor oil in the aircraft industry. The first work on the toxicology of ricin was performed by Hermann Stillmark at the Dorpat University in Estonia for his 1888 thesis. Stillmark determined that ricin was a protein and suggested the name. He purified it to a very high degree and found that it agglutinated erythrocytes and precipitated serum proteins. For years, these effects were considered to be the mechanism of action of ricin, although later work showed that the toxicity and agglutination effects were separate properties. In 1891 Paul Ehrlich studied ricin and abrin in pioneering research that is now recognized as the foundation of immunology.

Biochemistry

Ricin is classified as a type-2 ribosome-inactivating protein (RIP). Whereas Type-1 RIPs consist of a single enzymatic protein chain, Type-2 RIPs, also known as holotoxins, are heterodimeric glycoproteins. Type-2 RIPs consist of an A chain that is functionally equivalent to a Type-1 RIP, covalently connected by a single disulfide bond to a B chain that is catalytically inactive, but serves to mediate entry of the A-B protein complex into the cytosol. Both Type-1 and Type-2 RIPs are functionally active against ribosomes *in vitro*, however only Type-2 RIPs display cytotoxicity due to the lectin properties of the B chain. In order to display its ribosome inactivating function, the ricin disulfide bond must be reductively cleaved.

Structure

The tertiary structure of ricin was shown to be a globular, glycosylated heterodimer of approximately 60-65 kDa. Ricin toxin A chain (RTA) and ricin toxin B chain (RTB) are of similar molecular weight, approximately 32 kDa and 34 kDa respectively. This basic structure of ricin is similar to those of the botulinum toxin, cholera toxin, diphtheria toxin, tetanus toxin, and insulin.

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Appalachian Plant Profile: Burdock

By Dean Myles, Coordinator Medicinal Botanicals Program Mountain State University

Arctium lappa L., is a introduced herbaceous biennial commonly known as great burdock. Great burdock can achieve heights on 2 to 9 feet [1]. The lower stems (petioles) are solid with celery like grooves along the stem. The lower leaves are very large and become smaller as they progress up the stem. The heart shaped wooly leaves have an alternating arrangement with an entire margin. The thistle-like flowers are a reddish purple. The flowers are 1 to 1 ½ inch across and occur in flat-topped clusters. The fruit is an achene with a pappus of short bristles. The brown seed pods are burr-like with hooked barbs to facilitate seed dispersal. *A. lappa* can be found growing in waste places throughout most of North American but is not reported in West Virginia [2, 3].

Arctium minus L., is also an introduced herb known as common burdock. *A.minus* is smaller (2 to 5 feet in height) with hollow leaf stems without the grooves along the stem [1]. The flowers are also smaller and occur in loose raceme. *A.minus* is common in all areas of North America including WV [2, 3]. Both species have been used medically.

Traditionally, the roots, leaves, and seeds of *A. lappa* have been used in China, Japan, Europe and North America [1]. A root tea has been used as a blood purifier, diuretic, to stimulate bile secretions, digestion and sweating. The root has also been used for gout, liver and kidney ailments, rheumatism, and gonorrhea. The root is also used for diabetes due to high inulin content. Inulin is simply fructose polysaccharide that is not absorbed into the blood stream like sucrose therefore is well tolerated by diabetics [Encarta Dictionary]. The leaves are used in TMC for vertigo and rheumatism [1]. The seeds have been used for abscessed teeth, tooth aches, sore throat, insect bites, snakebites, flu, and constipation. Externailly the root and leaves have been used as a wash for burns, hives, eczema, and other skin conditions. Studies suggest the root may curb cell mutations and be effective for cancer.

A. Lappa is a principal herbal ingredient in the Essiac cancer remedy and Hoxsey formula. [4]. Preliminary study has shown that burdock may have anti-cancer effects and increase quality of life in cancer patients. Based on traditional use, burdock is generally believed to be safe when taken by mouth in recommended doses for short periods of time. However, burdock may also cause changes in potassium or sodium levels in the blood due to diuretic effects. Burdock may increase the risk of bleeding when taken with anticoagulants and NSAID's. Burdock should be avoided if pregnant or taking diuretic drugs. Caution is advised in patients with diabetes or hypoglycemia, and in those taking drugs, herbs, or supplements that affect blood sugar.

Cultivation of either *Arctium* species is quite easy. Simply collect dried seeds and plant in prepared beds in early spring. Burdock is grown in full to partial sun in varying soil conditions. However is best in deep, loose moist soil [5]. Thin plants to 6 inch apart in rows about 2 feet apart. The roots are usually harvested in July when they are 12 inches or more in length and about 1 inch thick [6]. Yields of 1,500 to 2,000 lbs of dry roots per acre can be expected in optimal conditions. Remember when planting burdock that it is highly invasive and due to its deep tap root and mode of seed dispersal is difficult to control. Please contact your state's agency concerning harvesting of wild plants or contact your local native plant program or the National Plants Database at http://plants.usda.gov/ for species status.

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Photograph courtesy of: Janice Stiefel at http://wisplants.uwsp.edu/scripts/detail.asp?SpCode=ARCLAP

Ricin *(Cont'd)*

Description of the Agent

Ricin is a 66-KD globular protein that typically makes up 1% to 5% of the dry weight of the castor bean, although the yield can be highly variable. The toxic form is a heterodimer consisting of a 32-KDa A chain connected to the 32-KDa B chain through a single disulfide bond. As such, it is a member of the type II family of ribosome-inactivating proteins (RIPs), which possess enhanced *in*

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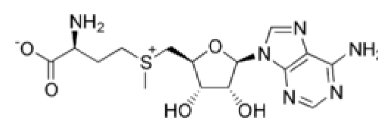
S-Adenosylmethionine: Molecular, Biological, and Clinical Aspects

CS Lieber and L Packer. 2002. Am J Clin Nutr, 76 (suppl):1148S–50S.

ABSTRACT

In clinical research, a novel approach has emerged: some of the essential nutrients are being used to treat pathologic conditions. Many of these nutrients, including methionine, must first be activated in the liver or in other tissues before they can exert their key functions. However, this activating process is impaired in disease states and, as a consequence, nutritional requirements change. For instance, for methionine to act as the main cellular methyl donor, it must first be activated to *S*-adenosylmethionine (SAMe; also known as ademethionine). SAMe is required and is of fundamental importance for the metabolism of nucleic acids and polyamines, the structure and function of membranes, and as a precursor of glutathione. These processes are often seriously altered in various pathologic states, but they cannot be restored by simply administering methionine. For instance, in liver disease associated with impairment of the enzyme that activates methionine to SAMe, supplementation with methionine is useless and may even become toxic as it accumulates because it is not used. Accordingly, one must correct the lack of SAMe by bypassing the deficiency in enzyme activation; this is done by providing the product of the defective reaction, namely SAMe. Under these pathologic conditions, SAMe becomes crucial for the functioning of the cell. Thus SAMe, which is found in all living organisms, becomes the essential nutrient instead of methionine.



Ricin *(Cont'd)*

vivo toxicity because of the presence of the B chain that facilitates uptake by the cell. Type I RIPs lack the B chain, and cellular toxicity is much less; uptake depends on endocytosis. Both chains are glycoproteins containing multiple mannose residues on their surfaces; association of both chains is required for toxicity.

Purification and characterization is not difficult and the crystal structure has been determined to be 0.25 nm. E ach chain is a globular protein, with the A chain tucked into a gap between two roughly spherical domains of the B chain. A lactose disaccharide moiety is bound to each of these spherical domains. The disulfide bond links residue 259 of the A chain with residue 4 of the B chain. The crystal structure demonstrates a putative active cleft in the A chain, which is believed to be the site of enzymatic action. A functional lipase active site at the interface of the two subunits was recently identified. This site is thought to be important for intracellular A chain translocation and subsequent intracellular trafficking.

Toxicity

Ricin is poisonous if inhaled, injected, or ingested, acting as a toxin by the inhibition of protein synthesis. While there is no known antidote, the US military has developed a vaccine. Symptomatic and supportive treatments are available and long term organ damage is likely in survivors. Ricin causes severe diarrhea and victims can die of shock.

Therapeutic Applications of Ricin

Some researchers have speculated about using ricins in the treatment of cancer, as a so-called "magic bullet" to destroy targeted cells. Ricin could be linked to a monoclonal antibody to target malignant cells recognized by the antibody. The major problem with ricin is that its native internalization sequences are distributed throughout the protein. If any of these native internalization sequences are present in a therapeutic, then the drug will be internalized by, and kill, untargeted epithelial cells as well as targeted cancer cells.

Some researchers hope that modifying ricin will sufficiently lessen the likelihood that the ricin component of these immunotoxins will cause the wrong cells to internalize it, while still retaining its cell-killing activity when it is internalized by the targeted cells. Generally, however, ricin has been superseded for medical purposes by more practical fragments of bacterial toxins, such as diphtheria toxin, which is used in denileukin diftitox, an FDA-approved treatment for leukemia and lymphoma. No approved therapeutics contains ricin. A promising approach is also to use the

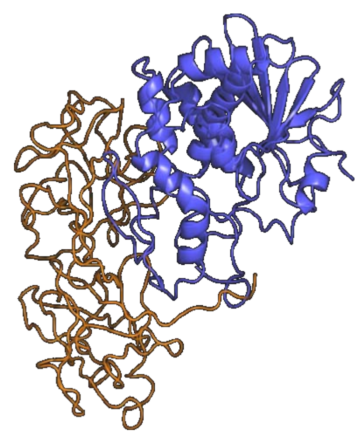


Figure 2. Structure of Ricin. Ricin is made of two protein chains linked by a disulfide bond. The A chain (blue) is an N-glycoside hydrolase that inhibits protein synthesis. The B chain (orange) is a lectin that helps ricin bind to a cell.

non-toxic B subunit as a vehicle for delivering antigens into cells thus greatly increasing their immunogenicity. Use of ricin as an adjuvant has potential implications for developing mucosal vaccines.

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About the Medicinal Botanical Program

This Program was created as result of a Specific Cooperative Agreement between Mountain State University and the USDA/ARS-Appalachian Farming Systems Research Center in Beaver, WV. The establishment of this agreement came through the efforts of Senator Robert C. Byrd and a Congressional Appropriation. The mission of the Program is to promote the medicinal plant industry in WV through research, education and outreach. The Program conducts research aimed at the identification and development of native plants as specialty vegetable/forage crops. Educational offerings include symposia, workshops and farm visits.

The Soil Biology Primer-Part VII *(Cont'd)*

rhizosphere, the narrow region next to and in the root. There is evidence that plants produce certain types of root exudates to encourage the growth of protective bacteria.

Bacteria alter the soil environment to the extent that the soil environment will favor certain plant communities over others. Before plants can become established on fresh sediments, the bacterial community must establish first, starting with photosynthetic bacteria. These fix atmospheric nitrogen and carbon, produce organic matter, and immobilize enough nitrogen and other nutrients to initiate nitrogen cycling processes in the young soil. Then, early successional plant species can grow. As the plant community is established, different types of organic matter enter the soil and change the type of food available to bacteria. In turn, the altered bacterial community changes soil structure and the environment for plants. Some researchers think it may be possible to control the plant species in a place by managing the soil bacteria community.

BUG BIOGRAPHY

By Ann Kennedy, USDA Agricultural Research Service, Pullman, WA

Bacteria That Promote Plant Growth: Certain strains of the soil bacteria Pseudomonas fluorescens have anti-fungal activity that inhibits some plant pathogens. P. fluorescens and other Pseudomonas and Xanthomonas species can increase plant growth in several ways. They may produce a compound that inhibits the growth of pathogens or reduces invasion of the plant by a pathogen. They may also produce compounds (growth factors) that directly increase plant growth.

These plant growth-enhancing bacteria occur naturally in soils, but not always in high enough numbers to have a dramatic effect. In the future, farmers may be able to inoculate seeds with anti-fungal bacteria, such as P. fluorescens, to ensure that the bacteria reduce pathogens around the seed and root of the crop.

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Contributions

Dear reader:

Would you like to share your knowledge, skills and experience with us? Do you know how to produce, process, market and/or use herbs and medicinal plants? Would you like to share this knowledge with our readers? It is quite simple. Just write your ideas on a piece of paper and mail it to us. We will type it and make sure that it gets published in our newsletter.

Please send contributions to the addresses indicated above.

MBP in Pictures

Coltsfoot plant growing outside the MSU MBP greenhouse

Coltsfoot is one of the most widely used herbs for the treatment of coughs and other lung complaints. It is the basis for many of the herbal cough preparations sold in Europe. It has expectorant, anti-tussive, anti-spasmodic, demulcent, anti-catarrhal and diuretic properties, which make it extremely helpful in the case of racking coughs such as those that accompany chest colds, asthma and emphysema.

Coltsfoot contains pyrrolizidine alkaloids but the amounts are extremely small that the beneficial effects are generally believed to outweigh the miniscule risk.