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Bio II/ Prof. Kollar
Oct. 8, 2014

Subterranean Immunology: Re-examination of Autumn Olive

Elaeagnus umbellata, commonly known as Autumn Olive, is considered to be an invasive, non-native species on the American continent. This shrub is native to Eurasia and was brought to North America in 1830 for the purposes of preventing erosion of un-vegetated dry, sandy soils with low-pH and as a wildlife attractant. In the 1940s the U.S. Soil Conservation Service advised the planting of Autumn Olive as windbreaks and to stabilize strip mine soil. According to the Michigan Department of Natural Resources Autumn Olive provides a winter food source in the form of lycopene rich berries to a variety of songbirds such as thrushes, cardinals, Cedar waxwings, Evening Grosbeak, sparrows, Bobwhite, Ruffed grouse, Ring-necked Pheasants, wild turkeys, and Mallards. When flowering this plant provides nectar to Ruby-Throated Hummingbirds and Sphinx Moths, and provides nesting habitat and protective cover for songbirds. The University of West Virginia Extension Service details this shrub as a food source for upland game birds, two migratory game birds, twenty non-game birds, and four mammals. In contrast, the Missouri Department of Conservation states that Autumn Olive is an invasive shrub that "degrades native wildlife habitat" and recommends chemical control with the herbicides 2,4-D, dicamba, or a 50/50 mix of triclopyr and diesel fuel.

This conflicting information entreats one to investigate further the role of this plant in our local environment. This shrub represents a complex symbiotic relationship below ground as well as above, which involves this plant, nitrogen fixing gram positive, aerobic Frankia actinomycetes, and aerobic arbuscular mycorrhiza belonging to phylum Glomeromycota. In fact, with further examination, one can see that this plant represents a complex beneficial triad of symbiotic organisms, the products of which may benefit humanity both directly and indirectly.

Upon observation it is easy to understand the above ground symbiosis that takes place between Autumn Olive and these diverse wild birds populations. The plant provides a carbohydrate rich fuel along with complex beneficial molecules such as lycopene and fatty acids that help birds maintain a healthy immune system against microbial infections and environmental stresses and toxins (Senar, Juan Carlos, Anders Pape Moller, et al., 2010). And in return these wild birds assist in the propagation of this plant. However, below ground the intracellular endosymbiosis that takes place between the plant, the mycorrhiza, and the bacteria is not easily observed nor understood. One can see the root nodules formed by the Frankia bacteria in the shrub's root mass but the filamentous arbuscular hyphae that extend between the plant's root cells are delicate haustoria and break down quickly on exposure to UV light.

These three organisms have a relationship based on extreme need and communicate via a complex molecular language of isoprenoid based stimulants and inhibitors. The gene CCD8 (carotenoid cleavage dioxygenase), which facilitates carotenoid metabolism, may have initiated the evolution of this symbiotic language and was present in the first phototrophic bacteria 3700 mya. These proteobacteria used carotenoid metabolism to synthesize bactiro-rhodopsin and passed this gene CCD8 to cyanobacteria. In plants, CCD8 has a regulatory function in branch production, root growth, flower development, and leaf senescence. CCD8 is a member of the same superfamily (RPE65) as the membrane proteins that bind retinal pigments in the eye. (Thomas, Huang,Young, Ougham, 2009).

In this three way endosymbiosis, the nitrogen fixing bacteria provide necessary nitrogen in the form of ammonia, and phosphorous to both mycorrhiza and to the plant. The mycorrhiza increase water absorption and the plant's uptake of potassium, copper, iron, nickel, sulfur, and zinc while at the same time provide protection against uptake of heavy metals such as cadmium and manganese. These mycorrhiza also protect the host plant and Frankia bacteria from disease through the production of antibiotics that give the Frankia colonizing bacteria the advantage over pathogenic microbes and protect plant roots from pathogenic fungal infections (Ianson, David and Jeff Smeenk, 2010). In return for these nutritional inputs, the plant provides the mycorrhiza and Frankia with fixed carbon molecules in the form of carbohydrates from photosyntheis and flavonoid compounds derived from the phenylpropanoid pathway (Abdel-Lateif, Didier Bogusz, and Valerie Hocher, 2012).

Each of the members of this three way collaborative synthesizes isoprenoid molecules derived from isopentenyl diphosphate, and the arbuscular mycorrhiza provide the phosphorous for this process. Both the Frankia bacteria and arbuscular micorrhiza make isoprenoids using the mevalonic acid pathway, while the plant uses both the MVA and alternate MEP pathways. The MVA pathway occurs in the cell's cytoplasm and is not light sensitive; however, the MEP pathway occurs in the cell's plastids and is light sensitive (Vranová, Eva, Diana Coman, et al., 2012). The Frankia use hopanoid lipids from MVA metabolism to form the diazo-vesicles, which are thick, spherical lipid-enveloped cellular wall structures that protect the nitrogenase used to convert nitrogen gas to ammonia from reactive molecular oxygen gas (www.microbewiki.Kenyon.edu).

The mycorrhiza produce sulfated and non-sulfated lipochitooligosaccharides that initiate plant root symbiosis as well as root growth and branching (Maillet F., Poinsot V., et al., 2011). The plant uses flavonoids to stimulate symbiotic exchange with mycorrhiza and Frankia. It synthesizes these chemical messengers through the phenylpropanoid pathway, transforming phenylalanine into 4-coumaroyl-CoA, which then enters the flavonoid biosynthesis pathway (María, L. Falcone Ferreyra, et al., 2012). Although the plant produces these flavonoid molecules, both mycorrhiza and Frankia bacteria reciprocate by serving as a subterranean immune system for the plant since these compounds have antimicrobial properties that strengthen the resistance of all three organisms against pathogenic microbes, insect pests and environmental stress.

Above ground avian wildlife and mammals benefit from the high carotenoid content in the berries, which helps these animals maintain healthy vision and immunity (Safran, Rebecca J., Kevin J.McGraw, et al., 2010). All three organisms use the shikimic acid pathway to synthesize the aromatic amino acids tryptophan, phenylalanine, and tyrosine. Tryptophan is necessary in the production of melatonin, which is also present in the berries that both birds and mammals need in order to maintain circadian sleep cycles.

When considering the fruit of Autumn Olive contains 40 to 50mg/l00g of lycopene, compared to 3mg/100g for fresh raw tomato and 10mg/100g for canned whole tomato (Black, Fordham 2007), one can see why this plant is valuable to wildlife throughout North America even though it is far from its native Himalayan realm. The discovery of *Elaeagnus tibetensis,* a fossil dating from the late Miocene at least 5.3 mya, indicates that the Qinghai-Tibet Plateau was likely the center of diversification for *Elaeagnaceae* (Su, Tao, Peter Wilf, et. al 2014). In the forests of Himachal Pradesh *Elaeagnus umbellata* is known as 'ghain,' growing wild at altitudes ranging from 1,200 to 2,100 meters commonly on dry, exposed hillsides and is used to treat pulmonary infections and cardiac ailments. It represents a source of vitamins A, C, E, and potassium as well as lycopene and melatonin in areas that will only support xeric fruit trees (Parmar, C. and M.K. Kaushal, 1982).

In conclusion, plants which offer an abundance of antioxidants such as lycopene and melatonin help increase herd immunity among wild bird populations. This plant is also host to moths such as the Speckled Casebearer Moth, *Coleophora elaeagnisella* that represent an important source of carotenoids for insectivorous bats. As in wild bird populations, these carotenoids can also improve immune response in bats to devastating pathogens such as *Pseudogymnoascus destructans*. During the Joseon Period in 19th Century Korea, bat imagery was popular in ceramics and furniture. *Elaeagnus umbellata* is also native to Korea and was introduced to the U.S. during this period. Thus, further study could focus on the plant's relationship with insectivorous bats in the process of determining its ability to benefit *Myotis lucifugus* as well as control insect pests of crops and forests. *Elaeagnus umbellata* represents a complex endosymbiotic relationship between plant, Frankia bacteria, and mycorrhiza that facilitates the high biosynthesis of lycopene. Further study of the production of bacteriocins by Frankia bacteria and Glomeromycota in this symbiosis is needed (Hassan M, Kio M, et al., 2012) as well as an analysis of flavonoids present in the seeds, juice, leaves, flowers, and phloem of Autumn Olive using gas chromatography-mass spectrometry. Beyond all the likely, however, little acknowledged benefits this plant offers to human populations it also fixes atmospheric nitrogen some of which comes from vehicle emissions. Therefore this plant's ability to filter our air is justification enough for its presence on the HCC campus.

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