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Soil Microbiology: With emphasis on plant nutrition.

Introduction:

Some bad news and some worse news. First the bad news. Soil chemistry is too dynamic and complex to ever be fully understand. The worse news. Soil microbiology is more complex than the chemistry and varies as functions of the chemistry, the structure, the plants and its own history.

The best way to handle a system as complex as the soil's biology is to ignore most of the components. Such a solution works (at best) only part of the time and that is why we often get conflicting data for soil-born disease studies. This is the reason that almost every biological control paper you can find reports success in one of the two years the study was conducted. Many studies have no success and those don't get published.

Lacking precession, we use metaphors such as "buffer" (as in pH) and "inertia" from physics. The buffering or the microbial-inertia in soil communities seems to be related to the number of component species. This is usually expressed (again metaphorically) as "richness of bio-diversity". If you could sterilize a soil and observe its re-colonization you would see that the first species to arrive increased its biomass dramatically as it exploited all nutrient sources present without competition. There would be a lot of amplitude in the biomass per species per unit time. As more and more species arrived the amplitudes of mass per each species would become more stable. There would be more resistance (inertia) in the soil to the arrival of new species. If plants are established in sterile soil and a pathogen arrives before there is sufficient microbial-inertia its may increase enough to wipe out all the plants.

Soil microbes don't like each other. We know and exploit this in our use of microbial antibiotics. The antibiotics Penicillium(s) from that genus of fungi and Streptomycin(s) from "bacteria" are examples of products that are common in soil communities. The most powerful natural toxins to us humans are produced by anaerobic soil bacteria that would like to be rid of all us oxygen using organisms. The point is that in soils inertia is a function of complexity. In simple language this is reflected in two sayings popular among ecologists: "Nature abhors a vacuum"and "diversity is stability".

Soil microorganisms are important to the breakdown of organic amendment. Various organic amendments favor different groups of microorganisms as a function of the ratios of complex to simple carbon sources and the ratios of N to C etc. It has long been a dream of pathologist to promote a "healthy"soil community of microorganisms (or at least a very high inertia one) through the proper use of organic soil amendments. So far, not much to report.

Bacteria:

There is no practical way to sterilize soil enough to remove the spore forming bacteria. Repeated autoclaving can but that is not practical (or desirable). Fungi are easier to kill but fumigation does not sterilize soil even as far as they are concerned, and again it would not be desirable. Without spending any more time on general soil bacteria, just remember they are there and they are important but there are so many we can't deal with them.

Plant Growth Promoting Rhizobacteria (PGPR).

Bacteria, and their metabolites, are more abundant in the rhizospere than in the rest of the soil. There are more carbohydrates there. Some of these bacteria produce products that can be used by the roots and some produce products that inhibit organisms that may be root pathogens (it may be the same product that does both). Either way, roots benefit and so do the rest of the plants. The bacteria are in the rhizosphere and they promote growth so they are PGPR. Most of what I know about PGPR is now 10-years-old and the knowledge base has increase a lot in that time. Scott Enebak has done a lot of research with PGPR in pine nurseries and has come to the conclusion that their affects vary so much from place to place that we are can not make much use of them at this time.

When I worked with "PGPR" in Florida it was in regard to compounds (called soderphores) they produce to chelate iron (and some other divalent cations). Once chelated, the iron is also available to plant roots and so they help plant nutrition (at least near neutral pH). There has also been speculation that this may deprive some pathogenic organisms of sufficient iron to make good growth and that this is a possible mechanism of disease suppression in the soil.

Fungi:

When soil fungi are monitored it is generally at the genus level. When pathogens are monitored it is usually at the sub-species level. An assumption that is made is that the species within a genus respond similarly to treatment. When populations of soil fungi are monitored the report usually lists numbers of Trichoderma, Fusarium, Rhizoctonia and Pythium. It should be remembered that there are unknown hundreds of species of these fungi and that the sub-species of interest are usually too few to be found by survey techniques.

Trichoderma:

Trichoderma is a very cosmopolitan soil fungus and apparently an important part of a soil's microbial-inertia. More has been written about bio-control using Trichoderma than for any other fungus. It is a good saprophyte, capable of getting energy from many organic sources, including cellulose and lignin, and is common in almost all soils. At least some strains are myco-parasites and produce metabolites very toxic to other (like Rhizoctonia). In addition, it is very tolerant to many broad-spectrum pesticides and generally increases in abundance after fumigation or treatment by some fungicides (it is sensitive to Benomyl). Trichoderma is believed to be more abundant in soil with good aeration and high organic matter and may play a part in the role of organic matter (composted) in disease suppression

In our recent work, and that of many others, populations of Trichoderma have been monitored after fumigation. We assume this indicates a resistance to the unwanted increases of potential pathogens. However, it should be stated clearly that we don't know the degree to which Trichoderma is important in any of the soilborn diseases in forest tree nurseries. There is better understanding of its function in some diseases of agronomic crops where it reduces the impact of Rhizoctonia and of some water moulds. The weakening of pathogenic fungi by pesticides increases the effectiveness of Trichoderma in these instances.

Nitrogen and Damping-off.

Damping-off is familiar to all nurserymen and is one of the first subjects in introductory plant pathology. Most pathologists think they know more about it than they do and so give false impressions to the growers they work with. **Damping-off** is the name given to a **symptom** that occurs when plant tissues without structural defenses are parasitized and collapse. It occurs during the time between germination and the plant's cuticle development (and anything that increases that time period increases the amount of damping-off). **Cool weather** and **deep sowing** retard hardening off and **moist conditions** can favor fungal development and increase damping-off, increasing the amount of **nitrogen** also tends to increase succulence and therefore damping-off.

Most pathologist and most plant diagnostic laboratories identify the fungi involved with the damped-off seedlings. Although proper identification can be important, when working with true pathogens, the fungi that cause damping-off may always be specialized pathogens. If they were specialized, then from the food base of the damped-off seedling, disease would continue to spread after the seedlings harden off. Experience will convince you that is rare in pine seedling nursery beds.

The fungi that cause damping-off are almost always species of ***Fusarium, Rhizoctonia*** or ***Pythium***. If it is consolation to know which of these fungi can be isolated from your seedlings then send have them identified. However, I believe most damping-off is a statistical probability that is simply associated with the length of the succulent period. If "x" damping-off would have occurred from a five day hardening off period then more damping-off would occur for each extra day required to harden off. Even soil fumigation does not appear to greatly affect the amount of damping-off.

In 1993, Westvaco evaluated a slow-release, fertilizer that was applied immediately after sowing. The fertilizer was accidentally damaged during processing and released much or most of its N soon after application. A sound study plan for the fertilizer combined with a lot of damping-off that year (a common but unpredictable event) to produce an opportunity to evaluate the affects of N on damping-off. There were five fertilizer treatments: the regular operational treatment, a 125 lb/ac application, a 250 lb/ac application, and two formulations containing 200 lbs/ac. Because the fertilizer study utilized two Latin Squares (each treatment in each row and column across the study area), one with a pre sow treatment of 150 lbs/ac NH4SO4, it was a powerful design to assess differences within the study area both by treatment and field position. The average damping-off was greater in that Latin Square were the NH4SO4 was applied (8 vs 6 per frame) and was greater in the plots with the "slow-release" fertilizer. Damping-off did not vary consistently between fertilizer treatments within the two Latin Squares but the nurseries operational fertilization practice (small weekly applications of N) which was no pre-treatment N, always had the least damping-off. Within both Latin Squares, damping-off differed significantly across the study area. The amount of damping-off increased steadily across the field in one direction (from column 1 to column 5).

Table 1. Damping-off by fertilizer and field position in a 1993 study in SC.

By Slow Release N, Combined for Pre Sow			By Field Position and Pre Sow NH4SO4		
Treatment	D-O / frame		Field Position	No Pre Sow	Pre Sow
Operational	2.4 a		1	14.0 a	16.0 a
250	7.9 b		2	7.4 b	13.2 a
200S	8.4 b		3	4.2 bc	6.2 b
200	8.7 b		4	2.6 c	2.8 b
125	7.9 b		5	2.2 c	2.0 b
Lsd	2.9			4.5	4.9

When evaluating the data in Table 1, notice the importance of filed position. Less balanced experimental designs would have confounded the affects of filed position with that of treatment. The differences between columns are probably is due to some consistent change across the field (moisture, OM etc) but the cause is not known. Coincidentally, I had a fumigation study in adjoining beds and damping-off varied similarly across my study plots in the same manner with respect to position but with no significant differences between fumigation treatments.

In the spring of 94, I observed differences in damping-off among container grown longleaf pine seedlings in Alabama. Damping-off differed among two types of containers used at the nursery and within container types between two media preparations. Although both media were mostly peat, one was a premixed composition and was mixed at the nursery. The two media differed for many nutrients (I failed to analyze either the media or the seedling properly) but the aim of the nursery had been to increase the nitrogen and have a greener crop. The nursery mix did produce a greener crop but with significantly more damping-off. Had this scenario occurred more recently, I would have utilized its potential better to learn about the affects of nutrition on damping-off. However, I acted like a pathologist and identified the fungi that were present. These were species of *Fusarium* if it means anything to you.

Mycorrhizae

The word mycorrhiza comes from the Greek words for fungus (mykes) and root (rhiza). So it actually means the symbiotic combination and not just the fungus. Much good science and a lot of pure BS is printed about mycorrhizae. There are two main types of mycorrhizal fungi. These designation are based on the physical appearance of the root/fungus interaction, but for the most part differentiate types of fungi. Ectomycorrhizae ("ectos" for informal discussions) are common to pines, oaks, and hickories and the endomycorrhizae (endos) are common to most other hardwoods and many other species of plants. Not all plant species have mycorrhizae. Many of the endos are species of *Endogone*. (a zygomycete). Most of the important ectos are basidiomycetes such as Thelephora, Pisolithus (the P in Pt) and Rhizopogon but some, such as the morels (which we eat if we can) are ascomycetes. Many fungi are capable of "casual" relations with roots that could rise to the description for fungus root interaction.

When you think about biological interactions, remember there is no free lunches. This goes for mycorrhizae too. The plant gets access to more soil resources through the large area of soil in contact with fungal hyphae. Under most conditions the plant benefits positively from the relationship. The largest benefit is usually increased phosphorus (P). However, if conditions become harsh the relationship can be detrimental. The fungus gets carbohydrates from the plant root and particularly with the endos may be able to get more than is optimal for the plant

A question that is answered differentially by various scientists is, how do different mycorrhizae influence seedlings and plantation establishment and how can nurserymen profit by knowing about mycorrhizae?

Endomycorrhizae:

Some plant species are obligate mycorrhizal. That is, they will not grow without their mycorrhizal symbionts. Sweetgum is probably the most important HW species in our nurseries in this category. On occasions, where we assume fumigation has been too affective, sweetgum seedlings are severely stunted by a lack of mycorrhizae. These stunted seedlings are (P) deficient and do not survive long once the weather becomes hot. Dogwood responds similarly but is grown less extensively and apparently has so many other problems that it is harder to pin down why it fails. At the Chiefland Nursery in FL they found by practice that it is best not to fumigate ground where they are going to sow dogwood. The mycorrhizal fungi associates of both sweetgum and dogwood are endos. These fungi do not produce spores for aerial dissemination (the ectos do) and this slows their recolonization of soil compared to the ectomycorrhizae.

Endomycorrhizae produce spores within root tissue that increase the long term survival in the soil. However, mycorrhizal hyphae in the soil appear to be the most important means of colonizing new plants. Mechanically working the soil reduces the viability (infectivity) of this mycelium. Periods of bare-fallow and the growth of non-host crops (like pine seedlings) also reduces the inoculum potential of soil with respect to endos.

Although I have seen a few situations where the application of endomycorrhizae spores would probably have been cost affective these situations were too few to balance the normal situation where amendments were not needed. I don't know how to predict or determine a situation where natural inoculum is inadequate and so on the whole I don't think that regularly adding inoculum would be cost affective.

Ectomycorrhizae:

Ectomycorrhizae are common and abundant throughout the region to which the southern pines are native. Their aerially disseminated spores so abundant that even in recently fumigated soil seedlings almost always become adequately mycorrhizal soon after germination. So why is there so much discussion about mycorrhizae with respect to pine seedlings? There are two answers and the one you get depends on whom you are listening to. It is difficult (perhaps impossible) to discuss ectomycorrhizae without touching on personality. This is because various researchers, and research groups, are associated with the advocacy of specific mycorrhizae.

Ectomycorrhizae are a persistent focus of discussion in forestry because some species may be "better" than others. And "better" in this context has meant that after outplanting, seedlings with one species grew and or survived better than those with another species. More recently there are claims for increased seedling growth in the nursery.

Discussion of ectos in the South eventually come around to *Thelephora terrestrius* (Tt). If you want to sell another ecto, something bad has to be said about poor old Tt. What nutsedge is to weeds, Tt is to ectomycorrhizae. You can't get rid of it. Therefore, any ecto you want to promote has to be "better" than Tt . The argument for another mycorrhizae usually cites greater adaptability to a range of cites after outplanting. Pt, the most famous mycorrhizae in the South, thrives in very acid soil conditions and was shown to be a to enhance seedling growth and survival on mine spoils. On more typical cites (and almost all sites are more typical) in the South, the data are less "dramatic". It is likely that most trees have a succession of mycorrhizae through their life in the field.

Bayleton, Mycorrhizae Suppression and Growth After Outplanting (From Kelley: SJAF)

		Ware, Co GA		Lee, Co AL					
Nursery	Bayleton (oz/ac)	Mycor %	Pt Index	Live %	dbh	ht	Live %	dbh	ht
AL (Camden)	0	77 a	25	91.4 a	3.1	17.6	80.0	3.8	24.9
	6	73 a	16	94.4 ab	3.3	17.9	88.8	3.6	25.3
	12	51 b	6	93.1 b	3.1	16.8	94.1	3.8	26.1
	24	36 b	6	90.2 ab	3.3	17.5	90.7	3.8	25.9
SC (Taylor)	0	77 a	-	93.2	3.3	17.8	92.7	3.7	26.2
	6	63 a	-	95.3	3.4	17.7	91.3	3.6	26.2
	12	42 b	-	94.2	3.4	18.8	91.3	3.7	26.8
	24	27 b	-	98.1	3.5	17.7	91.3	3.7	26.7

Mycorrhizae and Disease Control:

There are claims made for disease control by mycorrhizae. Although an influence is measurable, I doubt it is of insufficient magnitude to be of concern. Given the option to have abundant mycorrhizal inoculum or to fumigate to control disease I suggest fumigation is more cost affective. When you look at the table prepared by David South for the costs of different mycorrhizal inocula remember that fumigation cost about $2 /M seedlings so all but two mycorrhizal treatments cost more.

Costs of mycorrhizal inoculants (after David South, see AUSFNMC web page for more list)

Inoculum Type	Stock Type	$ / M seedlings
Pt spores	bare-root	0.43
Pt spores pellets	bare-root	2.75
Pt mycellium	bare-root	7.50
Pt spores + gel	bare-root	40.00 (Yes that is $40)
Pt spores + VA (VA = endo)	bare-root	51.00 for conifer or double rate for HW
Pt spores + humate	container	1.52
Pt spores spray	container	2.00

Even though pines may not have a true obligate requirement for their ectomycorrhizal symbionts they grow poorly without one. The first pines planted in Western Australia, Rhodesia and the

Philippines would not grow until reunited with mycorrhizae. Therefore, if you move to any of these places you will need additional information in order to make a financial about mycorrhizae.

Iron (Fe), Manganese (Mn), and Summer Chlorosis in Pine Seedlings:

Iron is an essential micronutrient for in all life forms. The chemical properties that make it biologically active, ironically cause it to be extremely insoluble at neutral or basic pH even though it is almost always abundant in the soil (iron is the 4th most abundant element in the earth's crust). Since we keep our bareroot nursery soils acidic for the production of pines, Fe should seldom be a problem. Neither should Mn. However, in nurseries where the irrigation water is near neutral a yellowing of the seedling may be common during the summer, especially when irrigation is the primary source of water. The exact cause of this is not know but I believe its due to slight, temporary, shifts in the pH of the soil solution. This is not to be confused with a shift in the pH of the soil. The observation that seedlings usually become greener when rainfall is more abundant seems to support this assumption.

Both iron and manganese are divalent cations when in the soil solution. Either or both can be the cause of summer chlorosis and the symptoms are indistinguishable. Both ions are taken up by plants using similar mechanisms and so may seem to "compete" with each other. That is, when Fe is much more abundant in the solution than is Mn, more Fe is taken up. This can make the chlorosis worse is Fe is added when Mn is deficient.

Mn deficiency is rare in bare root nurseries. However, Mn seems to be tied up by mill-sludge and I know of two instances where bareroot seedlings in sludge treated beds suffered Mn deficiency. Extremely chlorotic seedlings in sludge treated beds in GA had only 12 ppm Mn compared to seedlings in adjacent beds which contained 106 ppm. At an Alabama nursery, differences in Mn were measured between green (399ppm) outside and yellow (88ppm) seedlings inside and area treated with sludge more than 10 years earlier. Mill sludge is often high in Ca and this probably contributes both directly and indirectly to the low Mn availability. Ca both raises the pH and may compete for the mechanisms that uptake divalent cations.

Although some texts suggest that the Fe:Mn ratio is important and that there may be an optimal ratio or range, apparently healthy pine seedlings often have had fairly broad ranges of both elements. I have not been able to identify a ratio that appears to be preferable to pine seedlings. For the most part, it seems sufficient to avoid extremes of abundance or deficiency for both elements. In healthy pine seedlings Fe is reported to range form 100 to 2,000 ppm and Mn from 85 to 1350 ppm. In Table 2, the lower Fe:Mn ratio usually is associated with the greener seedlings but the measured difference does not always seem to be biologically important or large enough to be reliably assessed.

Table 2. Foliar Mn and Fe where green and yellow seedlings from five different bareroot nurseries were compared and no more likely cause (like disease or N) for chlorosis was indicated.

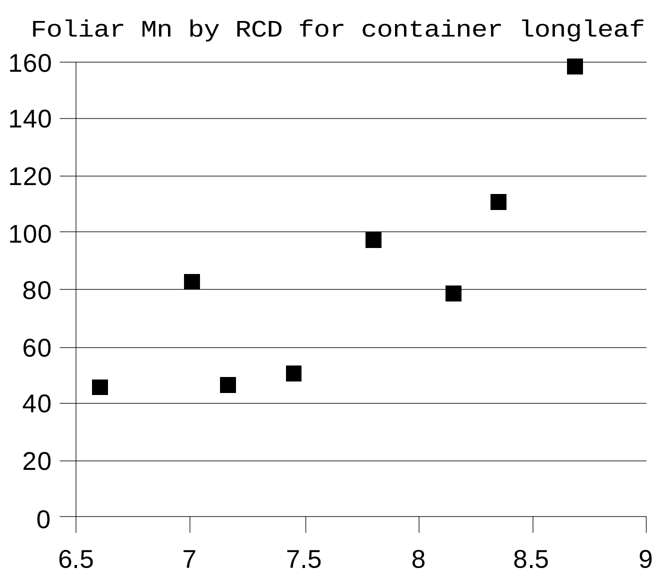
	Pair 1		Pair 2		Pair 3		Pair 4		Pair5*	
Color	Green	Yellow	Green	Yellow	Green	Yellow	Green	Yellow	Green	Yellow
Fe	118	206	70	100	223	334	84	109	153	156
Mn	83	62	436	518	482	620	541	944	399	88
Fe:Mn	1.42	3.32	0.16	0.19	0.46	0.53	0.15	0.11	0.38	1.78

* Pair 5 is an example of sludge induced Mn reduction.

There is some evidence (and that may be putting it too strongly) that Mn promotes the growth of longleaf pine. However, so far it is not clear whether more Mn in larger seedlings is a cause of or a reflection of better growth conditions (especially water availability).   There is very little Mn in peat, and this may be the reason that its deficiency has been more common (in samples to the disease lab) among container grown seedlings.   Although Fe is frequently added to bareroot beds at the first sign of chlorosis, a tissue analysis should proceed the addition of Fe to chlorotic seedlings growing in a peat based media.

The data in Figure 1 was produced by analyzing “families” growing at a GA nursery . It shows a significant linear relationship (r=0.86) between foliar Mn and RCD of container longleaf. What we can’t tell from the relationship is what is cause and what is affect as to the correlation of big seedling and more Mn (due to more water)..

Mn (ppm)



RCD (mm)

A “fumigation” study of bareroot longleaf at the Ashe Nursery produced the data in Table 2 for the affects of treatments on foliar nutrition (not statistically assessed) and RCD. In Table 2 the treatments are arranged, within each year, in order of increasing RCD except in year 1 where the Control and Pine Bark treatments are the same and seedlings/ft is used to select order. This order (by RCD) also puts the treatments in order of increasing foliar Mn and decreasing Fe:Mn ratio. My impression from these data is that is a good correlation between tissue Mn (but not Fe) and RCD. However, it should be understood that some persons would consider this the worst sort of “fishing for significance”. The fact that I sought out the data to investigate just these relationships is somewhat ameliorating of that “fishing” transgression.

Year	Treatment	Seedling/ ft	Soil pH	RCD	Soil Mn	Tissue Mn	Tissue Fe	Fe:Mn
1	Yard Waste	3.2	6.1	11.5	36	105	186	1.77
1	Control	2.5	5.8	13.2	42	122	219	1.79
1	Pine Bark	5.5	5.6	13.2	50	160	189	1.18
1	MBr	6.7	5.8	14.1	40	202	137	0.67
2	Yard Waste	1.7	6.2	8.2	73	115	327	2.84
2	Control	2.4	5.6	9.5	50	174	306	1.75
2	Pine Bark	2.9	5.6	9.6	61	190	230	1.21
2	MBr	3.4	5.6	12.9	58	213	234	1.09