Soybean Cellular Homeostasis of Micronutrients as a Function of Soil pH

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ABSTRACT

The soybean [*Glycine max (*L.)Merr.] is a cash crop in the United States, Brazil, and many other countries. Soil pH is known to determine nutrient availability, which in turn, affects crop yield. Plants have homeostatic responses to the available levels of nutrients. In response to toxic levels of nutrients, some plants use mechanisms that sequester the offending nutrients in anatomical sites that render them less destructive to cell function. This experiment investigated the spatial deposition and concentration of micronutrients in leaves of soybean grown in varying soil pH. Concentration and deposition patterns give insight into soybean homeostatic response to varying nutrient availability. Element intensity, measured in fluorescent counts which are proportional to concentration, and intracellular distribution were analyzed by synchrotron X-ray fluorescence (sXRF) at beamline 13-ID-E. Leaves were analyzed in vivo directly sampled off living plants at the time of analysis. Measurements were made of soybean plants grown in soils with low pH, optimal pH, and high pH. Soybean grown in lower soil pH contain higher levels of manganese (Mn) versus soybean grown in higher soil pH with Mn deposited in greater quantities in veins, necrotic regions, and hairs. Plants grown in differing soil pH levels did deposit varying intensities of micronutrients in leaves. The intensities as detected by sXRF mirror those predicted by generally accepted patterns of nutrient availability as determined by soil pH.

METHODS

Soybean plants were grown in loess derived soil of varying pH as follows:

Sample 1- pH 7.6
Sample 2- pH 7.4
Sample 3- pH 6.5 , high OM
Sample 4- pH 5.2

Note- all soils have 5-10% organic matter (OM) except Sample 3 which has 49% OM

Elemental Analysis

Leaf samples were analyzed in vivo at the GSECARS 13-ID-E X-ray microprobe beamline at the Advanced Photon Source, Argonne National Laboratory.

Run conditions were as follows:

- Incident beam energy of 12 keV, tuned using Si(111) monochromator
- 1 µm focused beam was used for analyses, focused using 200 mm long, Rh-coated , Kirkpatrick-Baez focusing mirrors
- X-ray fluorescence collected using 4-element Vortex silicon drift detector
- Approximate 2mm square area of live leaf was compositionally mapped and then a summed Energy Dispersive Spectra for this area was used for elemental evaluation
- The fluorescent counts are considered to be directly proportional to elemental concentration in samples.

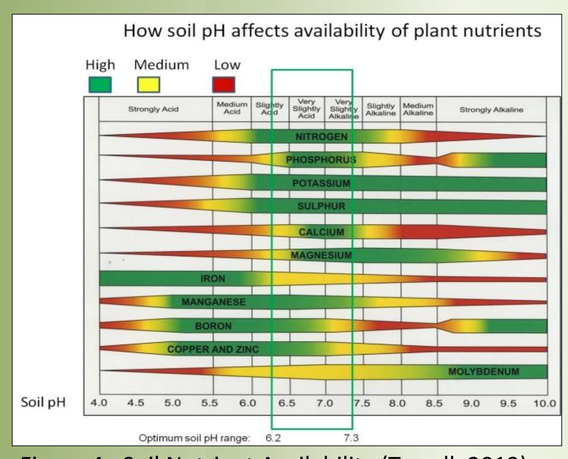

Figure 4: Soil Nutrient Availability (Towell, 2013).

RESULTS

Note- For all figures: Mn- Red Ca- Blue

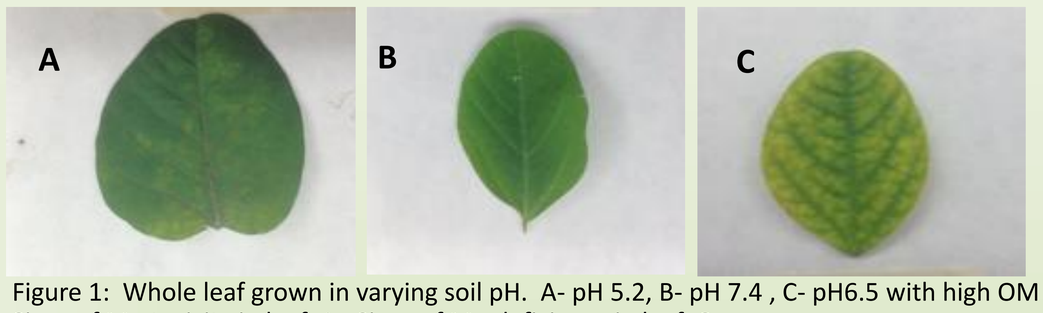

Figure 1: Whole leaf grown in varying soil pH. A- pH 5.2, B- pH 7.4 , C- pH6.5 with high OM Signs of Mn toxicity in leaf A. Signs of Mn deficiency in leaf C.

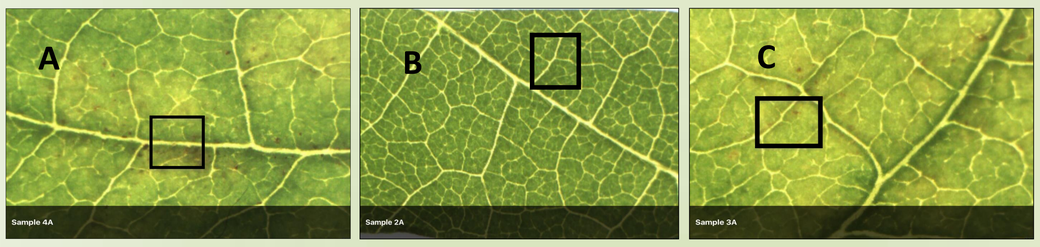

Figure 2: Leaf mesophyll (100x) of plants grown in varying soil pH. A- pH 5.2, B- pH7.4, C- pH 6.5 with high OM. Elemental analysis and maps below of boxed areas.

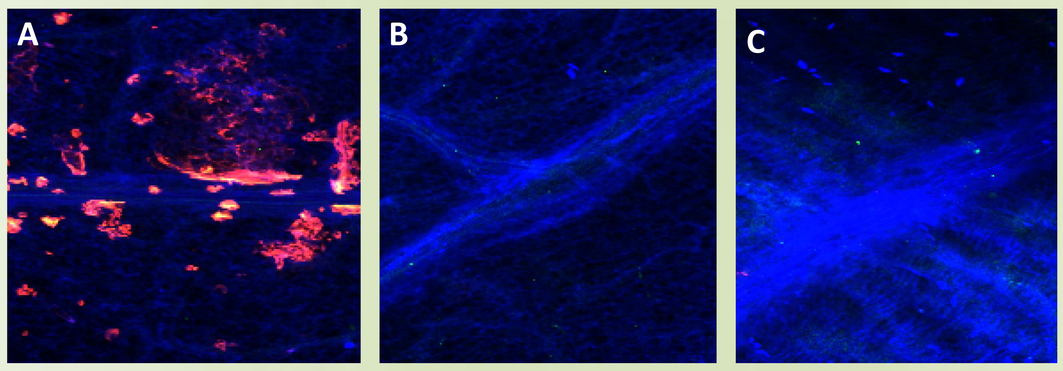

Figure 3: Elemental distribution maps of leaf mesophyll of plants grown in varying soil pH: A- pH 5.2, B- pH 7.4, C-pH 6.5 with high OM, All maps are scaled with equal sensitivity to Mn and Ca.

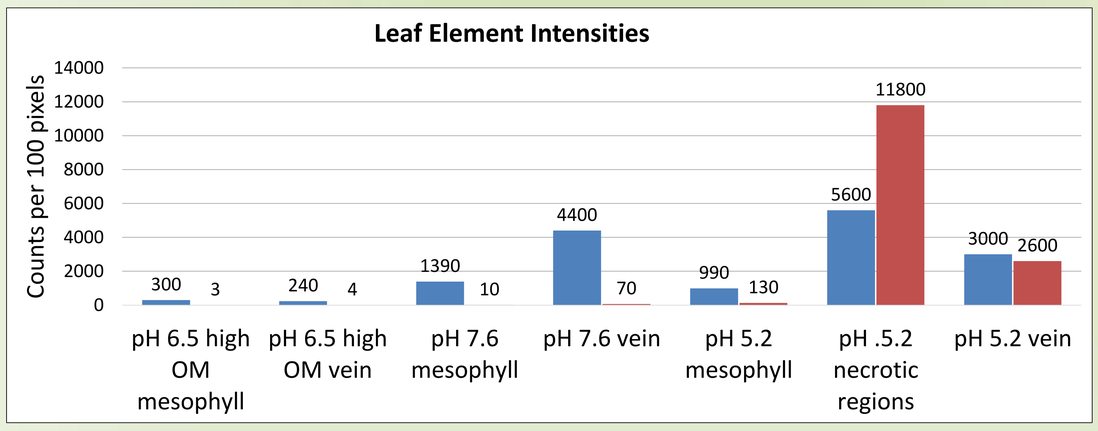

Figure 5: Leaf element intensities in various soil pH in mesophyll, veins and necrotic regions.

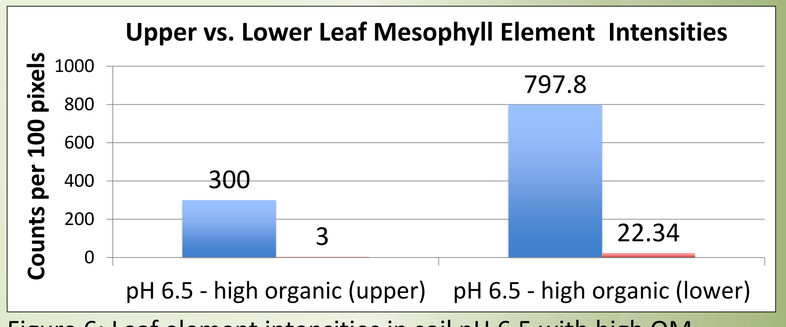

Figure 6: Leaf element intensities in soil pH 6.5 with high OM.

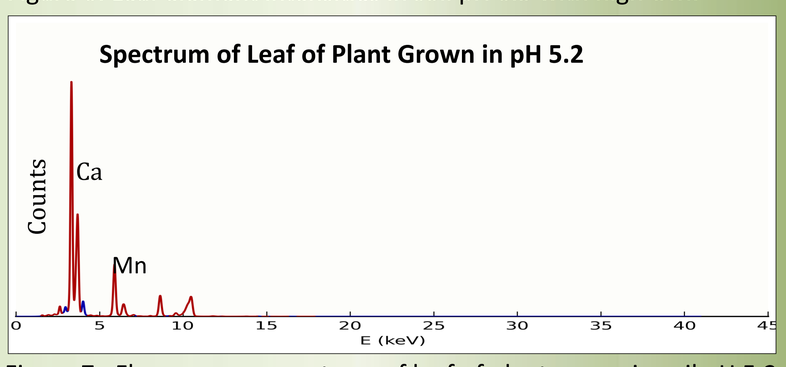

Figure 7: Fluorescence spectrum of leaf of plant grown in soil pH 5.2

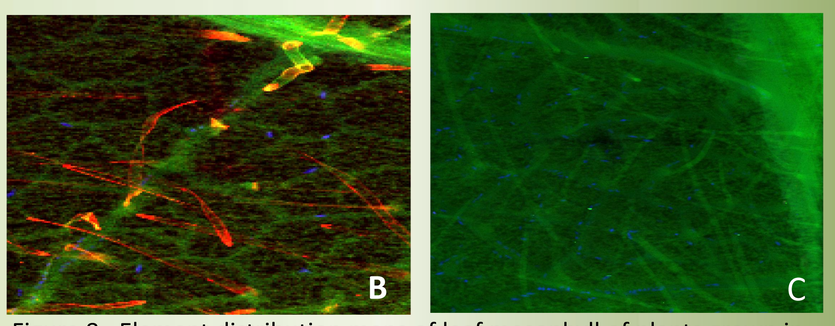

Figure 8: Element distribution maps of leaf mesophyll of plants grown in pH 7.4(B), and pH 6.5 with high OM (C). Hairs discernable by Compton correction (green) in sample C and Mn/Ca deposition of sample B. Both maps are Compton-corrected and scaled with equal sensitivity to Mn and Ca.

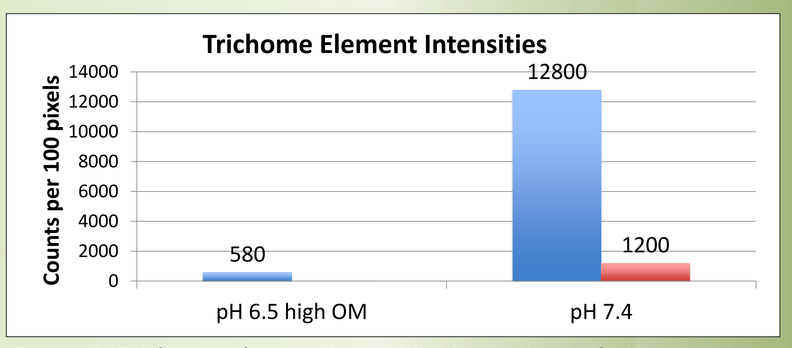

Figure 9: Trichome element intensities in varying soil pH.

RESULTS DISCUSSION

There is an expected inverse relationship between soil pH and Mn concentration and a direct relationship between soil pH and Ca concentration in soybean leaf (Figure 4). In this experiment, plants grown in lower pH had greater amounts of Mn and plants grown in higher pH had lesser amounts of Mn. Contrary to expected results, our data shows an inverse relationship between the intensity of Ca counts in leaves and soil pH (Figure 5). Mn is known to be an immobile nutrient. High OM soil creates nutrient deficient conditions for soybean and therefore shows great differences in deposition patterns. Our results indicate that when in these deficient conditions there were lower intensities of Mn in the younger, upper leaves versus the older, lower leaves (Figure 6). Lower soil pH allows for greater movement of Mn into plants and creates possible toxic conditions. According to Blamey et al., when soybean is grown in low pH, Mn will be deposited in greater amounts near veins and in necrotic regions. This research found similar patterns of Mn deposition as shown by sXRF elemental maps and total fluorescent counts (Figures 3, 5, 7). In addition, we found increased deposition of Mn in hairs of plants in optimal growth conditions (pH 7.4), but not in plants in deficient soil conditions (Figures 8, 9).

CONCLUSIONS

Soybean deposit elements in varying concentrations and deposition patterns in response to soil pH. The effects of the soil pH and resulting differing element intensities can be seen at the macroscopic scale. Figure 1, leaf C, exhibits interveinal chlorosis, which is symptomatic of Mn deficiency. Symptomology is also evident at the microscopic scale (Figure 2). We determined that there is an inverse relationship between soil pH and Mn deposition in soybean leaf. We also found evidence that Mn is an immobile nutrient in soybean. In plants grown in nutrient deficient soil, the intensity of Mn in younger leaves was less than that in older leaves, exhibiting this immobility (Figure 6). In agreement with Blamey et al., we found greater deposition of Mn near veins and necrotic regions in plants grown in soil pH 5.2 (Figures 3, 5, 7). This suggests there is an inability of soybean to sequester Mn, and therefore cannot prevent toxic levels from disturbing leaf cell homeostasis. There are many factors that contribute to cation exchange and uptake; these factors are not addressed in this research.

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