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### Preface

## Glyphosate interactions with physiology, nutrition, and diseases of plants: Threat to agricultural sustainability?

The transition from biologically based to intensive, chemical-based agricultural production systems advanced in North America and Europe soon after World War II as inorganic fertilizers and organically synthesized pesticides became widely available. This modern or conventional-type agriculture was adopted by other major crop production areas throughout the world with a sharp increase in adoption generated by the input-intensive “Green Revolution” of the 1960s and 1970s. In general, conventional cropping systems are characterized as large-scale production enterprises that utilize high-yielding crop varieties often in monoculture or short-term rotations planted on the most fertile, productive soils available with high inputs of chemical fertilizers and pesticides. Little emphasis is given to managing soil organic matter through use of traditional legume-based rotations, cover crops, or organic soil amendments that are central to maintaining the biological activity and inherent fertility of soils in biologically based cropping systems. By abandoning the biological management component, many conventionally managed fields have experienced severe disease, insect, and weed infestations (Drinkwater et al., 1995); serious declines in soil organic matter, nitrogen, and carbon contents (Khan et al., 2007); and alterations in the balance of beneficial and detrimental biological activities due to drastic changes in soil and rhizosphere microbial communities (Dunfield and Germida, 2004).

One of the most significant inputs necessary for successful conventional crop production are herbicides for management of the variety of weed infestations especially encountered in row-cropping systems. This technology was rapidly adopted because most weeds could be controlled when matched with selective herbicides, which were compatible with the crop, and was considered more cost-effective than cultural methods of weed management. Glyphosate, the active ingredient in the herbicide Roundup, became very popular after introduction in the 1970s for non-selective weed control in fallowed fields and non-cropped areas of orchards, vineyards, and timber plantations. The development of no-tillage systems (“no-till”) for row-cropping systems greatly expanded the use of glyphosate as it became standard practice to apply glyphosate to growing vegetation in fields prior to planting. This “burndown” application eliminated the need for tillage and allowed farmers to plant crop seeds directly into soil beneath a mulch of dead plant residues. The no-till practice contributed to reductions in soil erosion and energy consumption for field preparation and to expansion of grain production (primarily corn and soybean) in many areas suitable for row-cropping throughout the world.

Although glyphosate is the most widely used herbicide worldwide (Woodburn, 2000), several problems associated with

glyphosate interactions with plant nutrient availability, transfer to and effects on susceptible crops, indirect effects on rhizosphere microorganisms and plant pathogens, and development of glyphosate-resistant weeds have raised serious concerns regarding the sustainability of cropping systems in which glyphosate is the primary weed management strategy. Within this context it was our mandate to assemble a selection of papers for submission to *European Journal of Agronomy*. This Special Issue contains peer-reviewed papers based on contributions presented at the international symposium on “Mineral Nutrition and Disease Problems in Modern Agriculture: Threats to Sustainability?” held in Piracicaba-SP, Brazil, 20–21 September 2007. The symposium, organized by Dr. T. Yamada of the International Plant Nutrition Institute-Brasil (IPNI) continued discussions of issues presented in a previous symposium on herbicide impacts on plant nutrition and disease convened in 2005 and was held under the auspices of IPNI, The Agrisus Foundation – Sustainable Agriculture, ESALQ-University of Sao Paulo, and European Society of Agronomy.

The strategic location of Piracicaba was ideal for convening a symposium on experiences with nutrition and disease problems in modern agriculture. Sao Paulo is among Brazil’s leading states in land use devoted to sugarcane, citrus, and coffee plantations; the majority of these enterprises are managed following modern and intensive management, which include use of glyphosate to manage vegetation in these perennial crops. To the south, in subtropical southern Brazil, no-till agriculture was developed to reduce extensive soil erosion resulting from intensive row-cropping; adoption of no-till accelerated with the introduction of glyphosate to Brazil in the mid-1970s. To the north in the tropical savannah region (cerrado) of central Brazil, no-till was introduced in the 1980s as large farms devoted to soybean, cotton, and maize were established (Bolliger et al., 2006). After many years of frequent applications of glyphosate in both plantation and row crops in Brazil, several problems in plant health and productivity developed, which are representative of similar situations encountered in conventional cropping systems all over the world.

A basic understanding of the behavior of glyphosate in plants and the environment is necessary to set the foundation for the investigations upon which the Symposium was based. The mode of action, or the sequence of events leading to plant injury and/or destruction after herbicide treatment, has been described in numerous reports as the binding of glyphosate with and inactivation of 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS), the critical enzyme in the shikimate pathway required for synthesis of a variety of aromatic plant metabolites including essential amino

acids, phenylalanine, tryptophan and tyrosine (Franz et al., 1997). Tryptophan is necessary for the synthesis of indolylacetic acid (IAA), the main growth promoter, that can explain the widespread field observation of reduced in depth root growth of plants. Because a single, specific enzyme restricted to one plant metabolic pathway is inactivated by the herbicide resulting in eventual plant death, glyphosate was proclaimed as environmentally friendly (Franz et al., 1997). Additional standard assays identified other general properties of glyphosate including immobilization by soil colloids and organic matter and rapid degradation by soil microorganisms, which suggested the herbicide posed no adverse impacts on the environment or toward desirable plants.

More intensive evaluations revealed that glyphosate was translocated within plants, accumulated in roots, and was eventually released into the rhizosphere (Coupland and Casely, 1979). Subsequent research on glyphosate interactions with soil microorganisms demonstrated that although glyphosate was metabolized by a segment of the microbial population, it was also toxic to several bacteria and fungi; the net effect of glyphosate appeared to be a disruption of soil and root microbial community composition because selected components of the microbial community were stimulated while others were suppressed (Wardle and Parkinson, 1992; Busse et al., 2001). Interestingly, many studies showed that glyphosate was not entirely and immediately immobilized by soil colloids because a portion was available for metabolism by soil and rhizosphere microorganisms (Haney et al., 2000). In the 1980s, Rahe and coworkers documented that severe root infection associated with glyphosate-treated plants was due to disruption of synthesis of plant defense compounds, or phytoalexins, through the shikimate pathway thereby predisposing plants to attack by soilborne fungal pathogens (Johal and Rahe, 1988; Lévesque et al., 1987). Thus, infection by soilborne pathogens caused by the inability of plants to synthesize phytoalexins contributed to the overall herbicidal efficacy of glyphosate and was considered a “secondary mode of action” of glyphosate. These findings were significant because the release of glyphosate into the environment was found to have considerably more and far-reaching effects than the original notion that was limited to only the localized disruption of a specific metabolic pathway within a target plant. As glyphosate use increased in a number of agricultural systems, substantial evidence accumulated on multiple adverse effects on crop health and productivity and soil–plant–microorganism interactions mediated by this herbicide.

Based on the concerns of multiple agricultural and environmental effects associated with widespread use of glyphosate during the past decades, the Symposium was convened to present and discuss up-to-date research on problems of plant nutrition and disease linked to glyphosate use, present and critique sustainable alternative management strategies, and propose future research efforts. Key themes of the Symposium included interactions of glyphosate with nutrient availability to crop plants; interactions of glyphosate with plant pathogens and disease development in crop plants; and impacts of glyphosate on plant nutrition and microbial interactions in transgenic, glyphosate-resistant cropping systems (i.e., Roundup Ready).

Each presenter addressed one or more of the key themes of the Symposium. Römheld (Tesfamariam et al., 2009) reviewed the consequences of glyphosate transfer to non-target (crop) plants via the rhizosphere after herbicide application to target plants (undesirable vegetation). Evidence is provided that such transfer occurs in orchards when weeds in alleys are sprayed with glyphosate, which is subsequently released through the dying roots to be taken up through the living roots of trees. This was demonstrated with glyphosate-killed grass mulch simultaneously grown with citrus saplings, which contained significantly high contents of shikimate indicating glyphosate uptake by citrus from the dying grass roots

(Neumann et al., 2006). The cation-chelating ability of glyphosate, which previously received little attention regarding impacts on plant nutrition, was highlighted by several presenters as a critically important factor in nutrient deficiencies of crops observed in production systems heavily reliant on glyphosate for weed management. Römheld and Cakmak (Cakmak et al., 2009; Senem Su et al., 2009; Tesfamariam et al., 2009) discussed impaired micronutrient uptake and transport in plants exposed to glyphosate either through root transfer or by simulated drift of sub-herbicidal rates was due to the ability of glyphosate to form immobile stable complexes with Fe and Mn (Eker et al., 2006; Neumann et al., 2006). The possibility of interactive effects of glyphosate with other micronutrients was presented by Wood who suggested that the occurrence of Ni deficiency in pecan (Bai et al., 2006) and other orchard replant diseases might be exacerbated by release of glyphosate from killed vegetation in orchards, which then complexes with Ni making it unavailable for root uptake by trees.

Based on extensive field surveys and large-scale experiments, Fernandez et al. (2009) demonstrated that previous glyphosate applications (ranging from 18 to 36 months prior to planting) was the most important agronomic factor in development of diseases, primarily *Fusarium* head blight, in wheat and barley crops. Higher *Fusarium* colonization of wheat and barley roots was also associated with glyphosate burndown applications prior to planting (Fernandez et al., 2007). An unknown but interesting aspect of these observations is the apparent persistent effect of glyphosate on plant growth two or more years after application. Huber (Johal and Huber, 2009) reviewed various microbial interactions with glyphosate including those documented for toxicity toward beneficial microorganisms (i.e., rhizobia, Mn-reducers, mycorrhizae) and stimulation of detrimental microorganisms (Mn-oxidizers, pathogenic fungi). Through these interactions, glyphosate changes nutrient availability and alters pathogen virulence to plants. Some of the more notable diseases in which glyphosate might be implicated include *Corynespora* root rot in soybean, *Marasmius* root rot of sugarcane, citrus variegated chlorosis (*Xylella fastidiosa*), and take-all (*Gaeumannomyces graminis*) in cereal crops. Many of the pathogens causing these diseases are stimulated either by glyphosate exuded from roots, by the altered composition of root exudates caused by glyphosate treatment, or through a combination of both exudation processes.

The final key symposium theme addressed the impacts of glyphosate on plant nutrition and microbial interactions, and development of herbicide-resistant weeds in transgenic, glyphosate-resistant (GR) cropping systems (i.e., Roundup Ready). One of the most significant advancements in intensive agriculture is the introduction of GR crops in the mid-1990s. By 2008 GR-resistant soybean occupied 65.8 million ha (53% of the global area planted to biotech crops), followed by maize (37.3 million ha at 30% of global area), and cotton (15.5 million ha at 12% of global area) (James, 2008). The GR cropping system provided a more cost-effective option for farmers, allowing them to spray a broad spectrum of weeds with glyphosate on “as needed” basis and reducing the need for pre and post emergence herbicides. However, the repetitive and dedicated use of glyphosate within a growing season and over the past decade has resulted in selection for resistance in several weed species (Johnson et al., 2009) and development of similar crop nutrition and health problems as those observed in non-GR systems. Previous findings that glyphosate and high concentrations of soluble carbohydrates and amino acids were released in root exudates of glyphosate-treated GR soybean (Kremer et al., 2005) suggested that impacts on micronutrient uptake and root microbial interactions might mirror those described for glyphosate interactions in non-transgenic cropping systems. Indeed, presentations at the Symposium indicated decreased uptake of micronutrients and subsequent development of deficiency symptoms in some

GR soybean cultivars (Tesfamariam et al., 2009; Johal and Huber, 2009); only limited information has been previously reported on depressed uptake of Mn and Fe in GR soybean (Gordon, 2007; Jolley et al., 2004), suggesting that genetic modification in the GR soybean and/or glyphosate released into the rhizosphere affected micronutrient uptake. Valuable information on increased colonization by potential fungal pathogens, increases in Mn-oxidizing microorganisms, and decreases in beneficial bacterial populations (fluorescent pseudomonads, rhizobia) in the rhizosphere of GR crops is presented to aid in understanding some of the production problems often reported for GR-cropping systems (Johal and Huber, 2009; Kremer and Means, 2009).

In summary, the Symposium provided a forum to bring together a current understanding of the numerous factors – physiological, nutritional, soil chemical, phytopathological, and biological – that interact with glyphosate management whether situated in conventional (plantation, orchard, row crops) or in transgenic agroecosystems. This understanding is essential for developing alternative approaches within management systems to overcome the constraints to crop productivity and health. Some of the recommendations that emerged from the Symposium included reducing the use of glyphosate in perennial culture by using mulching systems to suppress weeds, which has been successful in many citrus plantations in Brazil. Development of efficient methods for using cover crops in annual crops for weed suppression and possible increased availability of soil micronutrients was discussed. Several approaches for improving productivity in GR cropping systems included selection of cultivars with high Mn-uptake efficiency, delayed application of micronutrients (Mn, Zn, Fe and Cu) after glyphosate treatment, and cultural practices, including the use of gypsum + Mo, and roller knife mulching that minimize glyphosate impact on crops. The use of biological products such as those containing the plant defense compound salicylic acid and amino acids by foliar application to improve resistance to root pathogens was also suggested. We trust that the articles in this Special Issue will serve as valuable information sources for those interested in a better understanding of the interactions of glyphosate with crop plants and that this information will be used to develop more sustainable agricultural production systems.

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