Improving Phosphorus Use Efficiency Through Organically Bonded Phosphorus

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Improving Phosphorus Use Efficiency Through Organically Bonded Phosphorus

Micheal Warner Hill

A thesis submitted to the faculty of
Brigham Young University
in partial fulfillment of the requirements for the degree of
Master of Science

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December 2012

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ABSTRACT

Improving Phosphorus Use Efficiency Through Organically Bonded Phosphorus

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Current maximum efficiency of phosphorus (P) fertilizers that is utilized by plants in the same year of application ranges from near zero to thirty percent. Despite low utilization of P in crop production, yields are often limited by P deficiencies. Innovative technology is requisite to achieve greater efficiency as fertilizer demands are increasing, while phosphorus mineral resources are rapidly being depleted. A growing environmental concern for nutrient pollution of surface waters also carries significant weight. A novel new product, Carbond\textsuperscript{®}P, is promising technology to increase P use efficiency. Research is needed to understand its capabilities and the functioning mechanisms imbedded within its technology.

Several research studies were conducted to evaluate Carbond\textsuperscript{®}P (CBP) against traditional fertilizers ammonium polyphosphate (APP) and monoammonium phosphate (MAP). A soil column leaching study was conducted to determine P mobility through three soils, at two rates (20 and 30 kg P ha\textsuperscript{-1}) in either a banded or mixed soil application. Mobility of P was evaluated at 24, 48, 110 and 365 d after fertilization. CBP showed significantly greater total P leachate values across all soil types and application rates averaged across all readings taken until 365 daf for both application types. In the banded applications, CBP generally produced significantly greater solubility than MAP or APP up until 110 daf. For applications mixed with soil, CBP and MAP had greater solubility than APP at 24 days after application, but by the later evaluation dates (48 and 110 daf) the CBP was significantly higher than both MAP and APP. No statistical significance was found in the leachate P 365 daf in either the banded or mixed applications.

One glasshouse study on maize (\textit{Zea mays} L.) grown in three soils were conducted at 0, 5, 10, 20, 40, 80, and 160 kg P ha\textsuperscript{-1} comparing CBP and APP fertilizer impacts on early season growth. CBP produced significantly more biomass in two soils when averaged across rates (and at the 20 kg P ha\textsuperscript{-1} rate in a third soil), increased petiole P in one soil and thicker stems in another. Two field trials showed similar physiological advantages with CBP over APP at later growth stages. CBP maize responded with significantly more biomass and P uptake at the V12-V18 growth stages in one field, as well as plant height in another. At the R2-R3 growth stages, CBP also produced thicker stalks in both fields than APP. These growth enhancements were strongest in medium to highly calcareous soil (6-12 %) low in P (7 mg kg\textsuperscript{-1}). These observations warrant the use of CBP and further investigation to understand its benefits and limitations.

Keywords: phosphorus, P, organic acid, Carbond P, maize, \textit{Zea mays}, fertilizer, phosphorus use efficiency, PUE
ACKNOWLEDGEMENTS

I would like to express my gratitude to Bruce Webb, Matthew Vickery, and Rachel Buck for their consultation and training in the BYU Soil and Plant Analysis Laboratory. There have been many fellow students who have contributed significantly the success of this project. Their hard work, positive attitudes, attention to detail, and insightful ideas have helped each project run smoothly, as well as elevate morale and friendship within the lab. I would like to acknowledge Joshua LeMonte, Curtis Ransom, Matthew Hamilton, Steven Bergsten, Tabitha Brown, Sean Summerhays, Tobin Story, Chelsea Katseanes, Austin Hopkins, Tyler Hopkins and all other members of the Biophysical Soil Chemistry Laboratory at BYU for their assistance. I am most appreciative to my graduate committee for their guidance throughout this experience. I have gained many insights into achieving not only academic excellence through their mentorship, but have been uplifted by their exemplary character. Last, I would like to thank my wonderful wife Amanda, loving daughter Leanna and other family members for their continual support and inspiration through this journey.
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Chapter 1

Phosphorus Mobility Through Soil Increased With Organic-Acid Bonded Phosphorus Fertilizer (Carbond® P)

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Manuscript accepted for publication in the Journal of Plant Nutrition, September 2012
ABSTRACT – ARTICLE 1

Phosphorus fertilizer is integral for maximizing crop production and is used abundantly to achieve desired yields. However, reduction of P fertilizer is warranted, as it is the primary source of nutrient pollution in surface waters (eutrophication leading to hypoxia) and is derived from non-renewable mineral resources. A research study was conducted in 2009 to evaluate a new fertilizer product, Carbond® P (CBP, 7-24-0) against traditional fertilizers ammonium polyphosphate (APP, 10-34-0) and monoammonium phosphate (MAP, 11-52-0). The study was conducted to determine P mobility through three soils using 0.18 m soil columns with fertilizers applied either in a concentrated band or mixed application rates of 20 and 30 kg P ha⁻¹. Mobility of P was evaluated at 24, 48, 110 and 365 d after fertilization (daf) by applying two pore volumes of water to the soil columns and analyzing the collected leachate for P. CBP showed significantly greater total P leachate values across all soil types and application rates averaged across all readings taken until 365 daf for both application types. In the banded applications, CBP generally produced significantly greater solubility than MAP or APP up until 110 daf. For applications mixed with soil, CBP and MAP had greater solubility than APP at 24 days after application, but by the later evaluation dates (48 and 110 daf) the CBP was significantly higher than both MAP and APP. No statistical significance was found in the leachate P 365 daf in either the banded or mixed applications. These results help explain increased P uptake by plants in field and greenhouse conditions and could lead to an overall reduction of P to produce crops.
INTRODUCTION

Efficient fertilization is essential for providing adequate food, fiber, and fuel for society (Hopkins et al., 2008). Phosphorus is an essential plant nutrient involved in every phase of growth and found in all plant parts (Bennett, 1993). It is an integral component to nutrient management for achieving maximum crop yields (Hopkins et al., 2008). The most important role of P is bioenergetics, as it is a component of the adenosine phosphates (AMP, ADP, and ATP) used in photosynthesis to convert light energy to chemical energy and in respiration activities. Consequently, adequate P concentration in plant tissue is critical for all energy requiring reactions. In addition, P is used to modify the activity of various enzymes by phosphorylation and for cell signaling. Furthermore, phosphate esters are structural components of DNA, RNA, and phospholipids.

Despite the universal requirement of P in plant nutrition, inefficient acquisition of P often limits potential growth. Chemical reactions in soil precipitate the plant available forms of P applied as fertilizer into forms poorly available for plant uptake (Hopkins et al., 2008; Mengel and Kirkby, 2001). These reactions are biphasic, or are known to occur rapidly upon fertilization and then slow as equilibrium of fixed P with solution P is reached (Larsen et al., 1959; Pierzynski et al., 2005). Several experiments show precipitation reactions reach equilibrium in approximately 48 hours (Black 1943; Larsen et al. 1959). The relatively short period of temporarily increased soil solution P and the somewhat static nature of the equilibrium greatly contributes to the inhibition of plant P uptake throughout the growing season.

Increasing the percentage of P fertilizer that is utilized by a crop (P use efficiency or PUE) has always been an obstacle for achieving high yields. Despite current cultural
practices to increase PUE (Hopkins et al., 2008; 2010a, b, c; 2012; Mengel and Kirkby, 2001; Randall et al., 1985), plants generally utilize between near 0 to 30% of P applied as fertilizer amendments in the first year after application (Randall et al., 1985; Syers et al., 2008). As a result, growers find it necessary to apply superfluous amounts to meet crop demands.

The rapid nature of P precipitation reactions with soil clays and oxides render this nutrient relatively immobile in the soil under prevalent mineral soil conditions with agronomically acceptable fertilization rates based on soil recommendations (Sims et al., 1998). Fertilization of P into lower sections of the profile low in residual P can be difficult or impractical where established perennial rooting systems exist. Inefficient fertilization may antagonize plant growth and yield. Poor P mobility through the soil is also a problem as very little P moves to plant roots via mass flow and uptake is largely dependent upon root interception and diffusion over short distances. Root growth can be slow during the various times of the growing season (especially during the initial phases of growth) for physiological, environmental conditions, or pest damage reasons.

Discovering innovative technology to increase the mobility and plant availability of chemical P fertilizers is paramount to the long-term sustainability of modern agriculture and consequently to providing food and fiber for the projected population increase during the next century (United Nations, 2007).

Land View Fertilizer Inc. (Rupert, ID, USA) has developed an organic acid N and P fertilizer (Carbond® P or CBP; 7-24-0). Preliminary research with $^{31}$P nuclear magnetic resonance has confirmed that this product is not simply a mixture of organic acids with N and P, but a bonding of the P to organic molecules is occurring (Hill and Hopkins,
unpublished data). Preliminary glasshouse work showed enhanced early season growth by application of CBP to maize (*Zea mays* L.), dry bean (*Phaseolus vulgaris* L.), sugar beet (*Beta vulgaris* L.), and wheat (*Triticum* spp.). For maize, the CBP fertilized plants had greater height (5 cm), darker green color, 12% more chlorophyll, and thicker stems (14%) than ammonium polyphosphate (APP:10-34-0) fertilized plants for all but the highest rates used in that preliminary study (Hill et al., 2011). The CBP fertilized plants also had significantly greater (40-64%) dry matter yield than the APP plants at the two lowest rates. These growth increases were likely associated with improved P uptake (average of 50% increase in P concentration). This is especially remarkable considering the CBP fertilized corn was compared at equal liquid volume application rates, which translates into 37.7% less total P applied with CBP than with APP. When comparing across equivalent rates of P, the P uptake would be even more dramatic, as would be the biomass increase at the lowest rates. However, the second highest rate showed no difference in biomass and the highest rate showed a significant loss of biomass for CBP in comparison to APP when comparing equal volumes of fertilizer applied. This decline in biomass was possibly due to a P induced micronutrient deficiency (Barben et al., 2011; Nichols et al., 2012) or binding of the micronutrients to organic acids in the CBP. These results suggest that at least 30% less P can be applied to achieve equal or greater early season growth yields. However, no additional benefit is observed at higher fertilizer rates. These findings were confirmed in follow up studies where response was linked to soil chemistry (rather than interactions within the plant), especially in calcareous soils with low soil test P and organic matter concentrations (Hill et al., 2013; Summerhays et al., 2013a, b). Additionally, several field studies also confirmed these results, with increases
in plant tissue P concentration and yield when P was supplied as CBP compared to traditional P sources (Hopkins et al., 2013). Studies with maize grown under field conditions resulted in greater than 10% increases in yield when soil test and fertilizer rates were relatively low. In all cases, even when yield increases were not observed due to the absence of P deficiency, the P concentration in plant tissue increased—again suggesting that CBP is more efficient in delivery of P to plant roots.

These findings demonstrate that P applied as CBP rather consistently delivers more P into plants even when applying less total P fertilizer. Understanding the mechanisms involved in enhanced PUE would be invaluable. Little is known about the solubility and mobility of soil solution P with this organically bonded P fertilizer (CBP).

Although P is generally immobile in soils, P can leach through soils under certain situations. These include preferential flow, long–term P over-fertilization (especially with high rates of manure and similar biosolids), leaching in deep sandy soils and high organic matter soils where mobility of P is presumed to be increased by the lack of soil constituents primarily responsible for P retention (clays, oxides of Fe and Al, and carbonates; Sims et al. 1998). A decrease in P sorption capacity with organic P applications (Bache and Williams, 1971) may be achieved with CBP especially in a concentrated band application where the organic components may protect through chemical bonding to the organic molecules or alleviate P retention by occupation of fixation reactions with soil constituents previously mentioned. The objective of this study is to investigate ability of CBP to maintain P soluble over time compared to monoammonium phosphate (MAP; 11-52-0) and APP. Enhanced solubility and mobility
compared to traditional products could explain observed growth enhancements and address mechanisms of P movement through soil profiles.

**MATERIALS AND METHODS**

Three fertilizer P sources (APP, CBP, and MAP) were evaluated in the laboratory in a soil column leachate study (29 March 2009 to 29 March 2010). Each of the three fertilizer sources was applied as either a banded or mixed application to alkaline sand (310 g pot⁻¹), calcareous sand (310 g pot⁻¹), and calcareous loam (245 g pot⁻¹) at 20 or 30 kg P ha⁻¹ and compared to an untreated control. Soil was placed in 18 cm by 5 cm diameter soil columns and packed to uniform bulk density similar to native soils. Soil was retained in the column with landscape fabric (DeWitt weed-barrier fabric, Sikeston, MO, USA) fastened securely to the bottom with a zip tie. The banded treatments were injected 5 cm below the soil surface and the mixed treatments were applied to the bulk quantity of soil and mixed thoroughly before being placed and packed in soil columns. Three replicates of each treatment were established in a complete randomized design. After fertilizer placement or mixing, each soil column was initially wetted to field capacity with minimal leaching and then cycled between this moisture level and a nearly constant mass air-dry state throughout the trial—avoiding loss of moisture from the bottom of the column. Mobility of P was evaluated successively at 24, 48, and 110 daf by applying two pore volumes of water to each of the soil columns and collecting the leachate after passing through a Whatman #2 filter. After 110 d, each soil was removed from the column, mixed thoroughly to simulate a tillage operation, and then repacked into each column. Soil columns were stored in a building without heat or insulation to
simulate natural field conditions with freezing and thawing over the duration of the winter season. Maximum temperature reached 40°C while the minimum temperature achieved was -23°C. As with the initial extraction conditions, soils were periodically wetted to simulate normal cycles of wetting and drying. After 365 d of treatment (original placement or mixing), soils were leached a final time. Following this final leaching, soils were removed from soil columns and mixed thoroughly before being extracted with sodium bicarbonate (Olsen et al., 1954). The P and other nutrient concentrations from each collected leachate and the P from the soil extraction was analyzed by inductively coupled plasma atomic emission spectroscopy (Johnson and Ulrich, 1959; ICP-AES, Thermo Electron Corporation, Franklin, MD, USA) Statistical analyses were performed using Statistical Analysis System (Version 9.1, SAS, SAS Institute, Cary, NC, USA). The data were analyzed using analysis of variance (ANOVA) with mean separation using Duncan Waller Multiple Range Test at the 0.05 level of significance.

**RESULTS AND DISCUSSION**

The increases in P movement through the soils for CBP compared to APP and MAP were similar for all three soils ($P > 0.272$) and both rates of fertilizer ($P > 0.183$) and, with the exception of the source by method of application interaction ($P = 0.002$), none of the interactions were significant for any combination of parameters ($P$ values ranged from 0.256 to 0.772). Consequently, results are combined across soil type and fertilizer rate and discussed in terms of fertilizer source and method of application.

The concentrated band applications, simulating a starter fertilizer application, showed that CBP generally produced significantly greater P movement through the soil
columns than both APP and MAP (Figure 1). The P concentration in the leachate was significantly greater in the order CBP > APP >> MAP at 24 daf. Despite having less total P remaining in the soil after the first leaching event, this trend generally continued for the next two leaching events; with CBP leachate P concentrations being significantly greater than MAP at 48 daf and significantly greater than both APP and MAP at 110 daf.

Although the APP initially had greater P movement through the soil than MAP, these sources did not differ in leachate P concentration after the initial 24 daf leaching event. As expected the magnitude of the P concentration decreased with time and there were no differences among sources at 365 daf (soils were tilled after the 110-d leaching). Because leaching events were applied successively, it is important to examine the total summed P mobilized, which showed the P concentration in the leachate was significantly greater in the order CBP > APP >> MAP.

When fertilizers were mixed thoroughly into soils, simulating a broadcast-incorporated application, CBP again generally produced significantly greater P movement through soil columns than APP and MAP (Figure 2). At 24 daf, leachate P content with CBP was numerically greater than both MAP and APP, but only CBP leachate was significantly higher in P than APP. The leachate P content from the MAP application was similar statistically to both CBP and APP at 24 daf. At 48 and 110 daf, CBP produced significantly greater leachate P than both MAP and APP. Phosphorus leachate contents from MAP and APP were never statistically different from each other (24, 48 or 110 daf). Similar to the band application, there were no differences in leachate P concentrations among fertilizer sources at 365 daf (Figure 2). In addition, both fertilizer applications methods resulted in CBP mobilizing significantly more P than APP and
MAP when the total leachate was examined (Figures 1 and 2). In contrast with the banded application (Figure 1), the mixed application method for APP and MAP did not produce different P leaching concentrations when comparing total P leached from these soils (Figure 2).

It is also interesting that orthogonal comparisons for each fertilizer source comparing application methods showed that MAP produced significantly more leachate when mixed rather than banded ($Pr > 0.001$; Figures 1 and 2). It is possible that this observation is due to physical proximity of some of the MAP fertilizer pellets randomly being placed near the bottom of the soil columns—resulting in less distance for the P to travel and, consequently, interacting with less soil than the banded applications where distance was uniformly closer to the top of the column. In contrast, both APP and CBP produced significantly greater P leachate with the band compared to mixed applications ($P = 0.012$ and 0.009, respectively; Figures 1 and 2). However, the magnitude of the difference was not as great as that with MAP.

Based on these findings, as well as other studies showing increased plant P uptake (Hill et al., 2011; Hopkins et al., 2013) with CBP compared to conventional P sources, the CBP technology may be having various effects on P plant uptake. One possibility is the potentially beneficial properties of organic acids utilized in CBP are working to mobilize P and sequester cations that would normally precipitate P out of the soil solution (such as Ca, Mg, Fe, and Al). The general benefits for organic acids in nutrient acquisition have been observed by several researchers. Plants deficient in P have been shown to up-regulate root exudation of organic acids into soils (Grierson, 1992; Zhang et al., 1997). It is well documented that various organic acids help mobilize poorly soluble
mineral nutrients—with citrate, malate, and oxalate the most common and effective at mobilizing P (Hoffland, 1992; Oburger et al., 2009). The ability of organic acids to reduce P precipitation (Grossl and Inskeep, 1991) and even solubilize poorly soluble phosphates (Singh and Amberger, 1998) is potentially valuable in meeting sufficient plant P demands. However, the effectiveness of each organic acid in maintaining P accessible to plants is based on: 1) functional-group content and geometry (Grossl and Inskeep, 1991), 2) pH (Kirk, 1999), 3) adsorption site affinity (Strom et al., 2002), and 4) degradation resistance (Andrade et al., 2007). The variety of organic acids utilized by CBP, in theory, would maintain P mobile for a longer period than APP and MAP. Each organic molecule utilized would work on its own level of effectiveness, which may vary slightly depending on the soil conditions.

A second impact that CBP technology is possibly having on P nutrition is physical bonding of P to organic acids to prevent rapid P precipitation with the soil as in traditional fertilizers. Instead, P is held in a bonded organic form that may contribute to the enhanced solubility of P for several weeks, as seen in this study. Although held away from soil reactions, plants are apparently still able to access the P as shown in other studies (Hill et al., 2011; Hopkins et al., 2013). The P-organic acid compound would have a relatively large molecular size and would likely be unable to transport across cell membranes. We theorize that the compound moves into the intercellular space in roots where the P is dislodged and made available for uptake. Research is needed to confirm the action for uptake, whether bonded forms are absorbed directly through leaves or roots and then utilized, or if plant chemical alterations of soil chemistry assist in breaking the bonds just before entering the plant.
The residual soil test P levels 365 daf showed bicarbonate extractable P concentration was significantly greater in the order of MAP > CBP > APP for the banded applications (Figure 3). As predicted, MAP, which allowed the least amount of P to be leached in the banded application, produced the greatest amount of P carry over. However, that same reasoning would lead one to expect that APP would have the second highest levels of residual P but this was not the case. It is possible that some of the functional groups on the slowly degradable organic molecules incorporated in CBP technology continue to work together in a concentrated enough band to help make insoluble P more labile in the soil (Singh and Amberger, 1998). No definitive explanation can be given without further experimentation. The same numerical trend was observed in the mixed application but only MAP was significantly greater than APP, with CBP not significantly different from MAP or APP (Figure 3). When taken out of a concentrated band, the fertilizers appear to react differently over time and are less “plant available”. Relative to the banded treatments, all residual soil P values were greatly reduced.

Although total soil soluble P was enhanced with CBP over traditional fertilizers APP and MAP, further examination is needed to assess the vertical distance the soluble P will travel with CBP. Enhanced P mobility using CBP may create similar environmental concerns with P as with N movement into waterways leading to eutrophication or into underground waters. However, in cases where P mobility is desired as in perennial crops, CBP may be more environmentally friendly than heavy manure or over fertilization of traditional fertilizers. If only slightly enhanced mobility is achieved, movement below easily erodible soils could help reduce P transport to waterways.
CONCLUSION

These data show highly significant differences among solubility of CBP and the traditional fertilizers, APP and MAP, over a period of at least 110 daf. If P is remaining in a more soluble state in the soil solution, it should also be more readily available for plant uptake; thus ameliorating the impediment of rapid precipitation of applied P fertilizers. Results of preliminary glasshouse experiments bolster the hypothesis that bonded P is accessible to plants for uptake. Further research is needed to determine how CBP is actually incorporated into plant matter.
REFERENCES


Available at: http://www.plantmanagementnetwork.org/cm


FIGURES

Figure 1. Banded Application Leachate Extractions. Concentrations of water extractable P in leachates obtained 24, 48, 110 and 365 d after fertilization (daf). Fertilizer treatments consisted of equal P levels applied as monoammonium phosphate (MAP, 11-52-0), ammonium polyphosphate (APP, 10-34-0) or Carbond P (CBP, 7-24-0) in a 0.05 m band applied to three soils at rates of 20 or 30 kg P ha\(^{-1}\) (results shown are combined across fertilizer rates and soil types and adjusted by subtracting leachate P obtained from an untreated control). Data bars sharing the same letter within leaching date are not significantly different from one another (\(P = 0.05\)).
Figure 2. Mixed Application Leachate Extractions.
Concentrations of water extractable P in leachates obtained 24, 48, 110 and 365 d after fertilization (daf). Fertilizer treatments consisted of equal P levels applied as monoammonium phosphate (MAP, 11-52-0), ammonium polyphosphate (APP, 10-34-0) or Carbon P (CBP, 7-24-0) mixed evenly in three soils at rates of 20 or 30 kg P ha$^{-1}$ (results shown are combined across fertilizer rates and soil types and adjusted by subtracting leachate P obtained from an untreated control). Data bars sharing the same letter within leaching date are not significantly different from one another ($P = 0.05$).
**Figure 3. Soil Extracted P Concentrations.**
Increase in bicarbonate extractable P adjusted for the untreated control. Soils for extraction were obtained from the bulk soil (thoroughly mixed) 365 days after fertilization (daf) with equal P levels applied as monoammonium phosphate (MAP, 11-52-0), ammonium polyphosphate (APP, 10-34-0) or Carbond P (CBP, 7-24-0) in three soils at rates of 20 or 30 kg P ha\(^{-1}\) (results shown are combined across fertilizer rates and soil types and adjusted by subtracting leachate P obtained from an untreated control). Data bars sharing the same letter within leaching date are not significantly different from one another ($P = 0.05$).
Chapter 2

Maize In-Season Growth Response to Organic Acid-Bonded
Phosphorus Fertilizer (Carbond P®)

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Manuscript accepted for publication in the Journal of Plant Nutrition, September 2012
ABSTRACT - ARTICLE 2

Phosphorus is essential for profitable crop production, but soil interactions antagonize efficient uptake. Increased P use efficiency is desirable for effective production and for reducing potential for P pollution in surface waters (eutrophication). Conservation of non-renewable P mineral deposits is an additional long-term concern. An organic acid based P fertilizer, Carbond® P (CBP; 7-24-0), increases P solubility through direct bonding of P, theoretically rendering it less likely to precipitate with soil cations. One glasshouse study on maize (Zea mays L.) was conducted in three soils at 0, 5, 10, 20, 40, 80, and 160 kg P ha⁻¹ comparing CBP and ammonium polyphosphate (APP; 10-34-0) fertilizer impacts on early season growth. Maize grown with CBP produced significantly more biomass in two soils when averaged across rates (and at the 20 kg P ha⁻¹ rate in a third soil), increased P concentration in one soil and produced thicker stems in another. Two field trials also showed similar physiological advantages of CBP over APP even at later growth stages with significantly more biomass and P uptake at the V12-V18 growth stages in one soil and an increase in plant height in the other. At the R2-R3 growth stages, CBP produced thicker stalks in both fields than APP. The physiological growth enhancement responses were strongest in medium to highly calcareous soil (6-12 %) low in P (7 mg kg⁻¹). Improvements in early and late season growth parameters using CBP compared to APP warrant its use and further investigation to understand its benefits and limitations.
INTRODUCTION

Efficient fertilization practices are essential for providing adequate quantity and quality of food, fiber, and fuel for society (Hopkins et al., 2008). Phosphorus is an essential plant nutrient involved in every phase of growth and found in all plant parts (Bennett, 1993) and an integral component in nutrient management for achieving maximum crop yields (Hopkins et al., 2008). The most important role of P is bioenergetics, as it is a component of the adenosine phosphates (AMP, ADP, and ATP) used in photosynthesis to convert light energy to chemical energy and in respiration activities. Consequently, adequate P concentration in plant tissue is critical for all energy requiring reactions. In addition, P is used to modify the activity of various enzymes by phosphorylation and for cell signaling. Furthermore, phosphate esters are structural components of DNA, RNA, and phospholipids.

However, efficient delivery of P to plant roots is an obstacle in crop production. Chemical reactions in soil precipitate or adsorb plant available forms of P applied as fertilizer into forms poorly available for plant uptake. These reactions begin immediately upon soil contact and occur rapidly after fertilizer is applied and, depending upon soil physical chemistry, result in plants utilizing near 0% to 30% of fertilizer P in the first year after application (Hopkins, 2012; Randall et al., 1985, Syers, 2008). The solubility of P is considerably reduced in strongly acid and alkaline soil conditions (optimal at slightly acidic to neutral pH), and lack of solubility is further exacerbated in the presence of excess lime (CaCO₃) (Westermann 1992).

To meet peak crop demands, growers often apply superfluous amounts of P fertilizer. This has created major concerns as fertilizer prices, environmental issues associated with
P use, and fears of depletion of finite P mineral resources rise. Various conservative reports estimate current P mineral resources will be exhausted within the next few centuries (Cordell et al., 2009; Gilbert, 2009; Marschner, 2012; U.S. Geological Survey, 2012). Nutrient pollution of surface waters resulting in eutrophication and leading to hypoxia already is a serious concern closely related to high soil P levels (Sharpley et al., 2003). Discovering and implementing innovative technology to improve the efficiency of P fertilizers are paramount to long-term sustainability of modern agriculture and consequently to providing food and fiber for the projected population increase during the next century.

There are a number of cultural practices relating to rate, timing, placement, and source options that can be used to increase plant utilization of applied P (P use efficiency or PUE) (Hopkins et al., 2010a, b, c; Hopkins 2012; Mengel and Kirkby, 2001). These practices include, but are not limited to, the following best management practices: 1) applying the correct rate based on experimentation under various environmental conditions, 2) insuring peak nutrient availability at the time of greatest uptake, 3) concentrating nutrients in the soil at locations where there is a large volume of roots, and 4) using sources that are soluble and have release patterns conducive for plant uptake. Fertilizer manufacturers and scientists are constantly attempting to develop new fertilizer technology or improve upon existing products to enhance PUE (Hill et al., 2011; Hopkins et al., 2008; Hopkins, 2012). Another practice to improve PUE has been to increase P solubility through the use of organic acids (Andrade et al., 2007). Plants deficient in P have been shown to up-regulate root exudation of organic acids into the soil (Grierson, 1992; Zhang et al., 1997), and it is well documented that various organic acids help to
mobilize poorly soluble mineral nutrients with citrate, malate and oxalate the most common and effective at mobilizing P (Hoffland, 1992; Oburger et al., 2009). Their ability to reduce P precipitation (Grossl and Inskeep, 1991) and even to solubilize poorly soluble phosphates (Singh and Amberger, 1998) is potentially valuable in meeting plant P demands. However, the effectiveness of each organic acid in maintaining P accessibility to plants is based on: 1) functional-group content and geometry (Grossl and Inskeep, 1991), 2) pH (Kirk, 1999), 3) adsorption site affinity (Strom et al., 2002), and 4) degradation resistance (Andrade et al., 2007).

Longer chain organic acids, such as humic and fulvic acids, have received considerable attention in their role of maintaining plant available P because of their complex structures and chemically active functional groups. “Humic matter (primarily macromolecules of humic and fulvic acids) is assumed to be the most widely distributed organic C containing material on the earth’s surface, is present in soils, in water of streams, lakes and oceans, and in their foam and sediments, from the tropics to the arctic regions” (Tan, 2003). They exert a number of important functions including slow release of plant nutrients, pH buffering, and interactions with micronutrients, toxic metal ions and xenobiotic organic molecules (Brunetti et al., 2007; Clapp et al., 2005; Hayes and Clapp, 2001; Senesi and Loffredo, 1999; Stevenson, 1994). “They are the most chemically active compounds in soils, with electrical charges and exchange capacities exceeding those of the clay minerals” (Tan, 2003). By being adsorbed to the surfaces of clays and oxides, they block P sorption sites and leave more P in soil solution (Grossl and Inskeep, 1991; Larsen, 1959; Pierzynski et al., 2005).
A major criticism for the utilization of organic acids in P fertilization is that most are transitory in nature, mineralize quickly (Andrade et al., 2007), and may not produce a long lasting effect on P bioavailability. Consequently, many studies have been carried out over a period of only a few weeks. However, others including Andrade et al. (2007) have shown that humic acids can withstand degradation over relatively long periods of time (Qualls, 2004; Stevenson, 1994) and can maintain their beneficial functions of keeping P free in soil solution compared to smaller, less stable organic acids, such as citric and acetic acids.

The functional groups that make up fractions of humic and fulvic acids are critical in determining their structural properties and the resultant chemical reactions. The main functional groups are: carboxyl, carbonyl, aliphatic hydroxyls, phenolic hydroxyls, and amine groups (Stevenson, 1994). The protonation of amino groups will create positive charges (Tan, 2000), making them amphoteric compounds (carry both positive and negative charges), but this reaction is expressed mainly at extremely low soil pH (< 4). Most of the negative charges from humic and fulvic acids are produced through deprotonated carboxylic groups in near neutral pH conditions, as well as deprotonated phenolic hydroxyl groups at high pH (Mengel and Kirkby, 2001), which permit them to act as weak bases (Stevenson, 1994). These functional groups are also important in inhibiting P precipitation in soils. The carboxyl and phenolic alcohol groups can complex or chelate ions known to form precipitates with P such as Fe, Al, Ca, and Mg. They can also occupy binding sites of clays and oxides through water and metal bridging. Although these functional groups are present in both humic and fulvic acids, the percent composition of each group and relative size of molecules varies between humic and
fulvic acids, with fulvic acids being smaller in size and having relatively greater concentration of aliphatic and carboxylic functional groups (Stevenson, 1994; Tan, 2003).

There are distinct discrepancies in the literature concerning this area of study—primarily with regard to how these acids are formed and if they maintain a replicable structure by source and location. Tan (2003) reviews the current science of humic and fulvic acids. As the fertilizer industry utilizes humic/fulvic acids, discretion is required despite the positive scientific examples cited in the literature previously discussed. One must consider reliability of such heterogeneous acids when considering any attempt to use organic acids to increase PUE or any other claimed impact on plant growth.

Land View Fertilizer Inc. (Rupert, ID, USA) has engineered a new P fertilizer, Carbond® P (CBP; 7-24-0) that is bonded with a suite of organic acids through a controlled manufacturing process (as opposed to the relatively common practice in the western USA of adding and mixing an organic acid additive with P fertilizers). The organic acids used in CBP are known to bond with P and is assumed to increase P solubility across a variety of soil types. Preliminary studies showed highly significant increases in P solubility and mobility for up to 110 d after fertilization in three alkaline soils (Hill et al., 2013). The apparent longevity of maintaining soluble P found with CBP is dramatic compared to both monoammonium phosphate (MAP: 11-52-0) and ammonium polyphosphate (APP; 10-34-0) and is of great interest in our efforts to increase PUE. Further preliminary research with $^{31}$P nuclear magnetic resonance (NMR) confirmed that CBP is not simply a mixture of organic acids but an organic molecule chemically bonded with P (Hill and Hopkins, 2011, unpublished data).
This increased P solubility translated into enhanced early season growth in preliminary glasshouse studies (Hill et al., 2011) with maize (Zea mays L.), as well as with bean (Phaseolus vulgaris L.), sugar beet (Beta vulgaris L.), and wheat (Triticum spp.) (Hopkins, unpublished data). For example, CBP fertilized maize plants were taller, (5 cm), darker green, higher in chlorophyll (14%), and thicker stemmed (14%) compared to APP fertilized plants for all but the highest P rates used in this preliminary study (Hill et al., 2011). The CBP fertilized plants also produced significantly greater (40-64%) dry matter yield than the APP fertilized plants at the two lowest rates. These growth increases may have been related to improved P uptake (average of 50% increase in P concentration), which is especially remarkable considering that the CBP fertilized maize was compared at equal volume application rates (6, 17, 51, and 152 kg ha\(^{-1}\)) which translates to 30% less total P applied with CBP than with APP (APP is 34% and CBP is 24% P\(_2\)O\(_5\)). The second highest rate produced no difference in biomass and the highest rate gave a significant loss of biomass for CBP compared to APP. The latter may have been due to a P-induced micronutrient deficiency or the organic acids in CBP are binding micronutrients. These preliminary results suggest that equal or greater early season growth can be achieved with 30% less applied P from CBP. It is important to emphasize that the responses were limited to the lowest rates of fertilizer P, confirming a previously oft observed condition that once a plant has adequate P that more is not likely to further enhance growth (Hopkins et al., 2010b; Hopkins, 2012; Mengel and Kirkby, 2001).

Further investigation comparing PUE of APP compared to CBP on an equal P rate basis in both greenhouse and field conditions is needed to understand how PUE translates into enhanced uptake and growth with these fertilizers. Therefore, the objectives of this
study was to compare height, stem width, shoot biomass, and tissue nutrient concentrations of maize to which equivalent rates of APP and CBP fertilizers were applied to soils in both glasshouse and field conditions.

**MATERIALS AND METHODS**

**Glasshouse Trials**

A study on early season maize growth was conducted on three soils simultaneously in a glasshouse located on the Brigham Young University campus at Provo, Utah, USA (12T 444776 E 4455133 N; elevation 1398 m) over 22 d (6 – 28 July 2011). In each soil, six replications of each treatment were arranged in randomized complete block designs. The objective was to evaluate differences in early season PUE of APP and CBP fertilizers as measured by plant height, stem width, total shoot biomass and P concentration. The trial was performed on three soils of medium P bioavailability and ranging from moderately to highly calcareous (Table 1). Nitrogen was balanced across all P rates with urea (46-0-0). Equal amounts of other nutrients were applied to each treatment based on soil tests (Table 1; Gavlek et al., 2003). Best management practices for plant growth in a glasshouse were observed. Pest infestation was minimal and, therefore pesticide applications were not required.

Targeted daytime and nighttime air temperatures were 25° and 15°C, respectively, although limitations in heating/cooling capacity resulted in ranges of 20-29°C daytime and 11-16°C nighttime temperatures. Natural light was supplemented with high pressure sodium lamps to maintain a 16/8-h light/dark cycle.
Soils (1.5 kg) were placed in elongated square pots (10 cm width x 25 cm depth) lined with landscape fabric (Weed Block, Easy Gardener, Waco, TX, USA) to prevent soil loss and to allow drainage. The containerized soil was tapped repeatedly to achieve uniform soil density (soil filled to approximately 5 cm from the top of the pot). Then the topmost 7.5 cm of soil was displaced and P fertilizer treatments were applied in a concentrated band. Rates of 0, 5, 10, 20, 40, 80, and 160 kg P ha\(^{-1}\) of each fertilizer source were applied based on simulated 50-cm row spacing. Three maize seeds pot\(^{-1}\) of hybrid N27B-3111 (presoaked in water 4 h prior to planting) were planted 2.5 cm deep to achieve a P fertilizer banded placement 5 cm below and 5 cm to the side of the seed.

Plants in each pot were thinned after 15 d to the two healthiest plants and measurements were taken 22 d after planting. Plant height was measured from the soil surface to the uppermost leaf being extended for a maximum height, while stem thickness was measured at the soil surface to the widest diameter of the stalk. Total shoot biomass was determined by destructive harvest followed by drying to the point of constant weight at 65° C. Shoot tissues were ground uniformly (Wiley Mill, 1-mm sieve), a 1.0 g subsample was digested in nitric-perchloric acid, and analyzed for nutrient content by Inductively Coupled Plasma Atomic Emission Spectroscopy (Johnson and Ulrich, 1959; ICP-AES, Thermo Electron Corporation, Franklin, MD, USA).

**Field Trials**

Two field trials were conducted during 2011 on similar soils as used in the glasshouse study to compare CBP and APP applications on maize (P8107 hybrid) grown in two commercial fields in South-Central Idaho.
One trial was conducted on a portion of a commercial field (interspersion of soils 1a and 1b in Table 1, although the majority was more characteristic of soil 1b) near Rupert, ID (12T 272936 E 5083540 N; elevation 1305 m). Planting occurred 24 May 2011 with fertilizer applied by large glass pipette in a concentrated band approximately 5 cm below and 5 cm to the side of the seed shortly after plant emergence (31 May 2012). Rates of 0, 1.4, 3.3, 7.6, 17.6, 40.4, and 93 kg P ha\(^{-1}\) of APP and CBP were applied (treatments were applied at multiples of 2.3 to be similar to rates used in the greenhouse and preliminary field trials). Each treatment was applied to four 7-cm wide twin rows 9 m long. Treatments were organized in a randomized complete block design replicated six times. The N in the P fertilizer sources used in this field trial was balanced across all treatments with urea ammonium nitrate solution (UAN; 32-0-0) applied in conjunction with the P fertilizers for a total of 62 kg N ha\(^{-1}\) applied with the P. At day 50, biomass, height, stem width, and total shoot nutrient contents were measured. Twenty plants were randomly selected for stem width measurement at the widest diameter 2.5 cm above the ground using high precision calipers. Height was measured from the ground to maximum height of hand stretched upper most leaves. These same plants were randomly harvested by cutting 2.5 cm above the ground. The plants were dried to the point of constant weight at 65° C and then analyzed as previously described. The height and stem thickness data were reassessed on d 70. Unfortunately, this cooperating grower applied CBP to the entire field shortly after our 70 d measurements, which reduced confidence in final harvest biomass measurements (data not shown).

The second field trial was conducted on a whole 12-ha irrigated field (soil 2, Table 1) near Gooding, Idaho (11T 671597 E 5082740 N; elevation 1,036 m). Plot sizes
were 24-twin rows wide (56-cm row widths) and approximately 600 m long. Control plots (0 kg P ha\(^{-1}\)) were randomly embedded within the treatments with the same width but only 25 m long. Treatments were organized in a randomized block design replicated three times. Rates of 0, 10, 15 and 20 kg P ha\(^{-1}\) of both fertilizer sources were applied at planting (13 May 2011) in cooperation with the grower with an automated applicator mounted on the planter in a concentrated band approximately 5 cm below and 5 cm to the side of the seed. The N found in the P fertilizer sources used in this trial were balanced across all treatments by banding urea ammonium nitrate (UAN; 32-0-0) for a total of 25 kg N ha\(^{-1}\) applied with the P. Also included in the same band was 11 kg K ha\(^{-1}\) (KCl; 0-0-12) and 1.1 kg Zn ha\(^{-1}\) [ZnSO\(_4\) chelated with ethylenediaminetetraacetic (EDTA)]. At 47 d after planting, biomass, height, stem width, and total shoot nutrient content were measured on subsets of these plots as previously described. The stem thickness measurements were reassessed on 88 d.

**Statistics**

All results were statistically analyzed using analysis of variance procedure in JMP software (Version 9.0.2, SAS Institute, 2010, Cary, NC, USA) with significance indicated at \(P < 0.05\). Mean separation was performed by LS Means Studentized t Tests for source parameters and the standard error was used when comparing rate by source interactions.

**RESULTS**

**Glasshouse Trials**

There was a highly significant orthogonal contrast between fertilized and unfertilized plant responses for all measured parameters for all soils \((P < 0.001,\) data not
Shoot biomass, stem width, plant height, and P concentration were all impacted to varying degrees by P source and rate (Tables 2-4). Increasing rate of fertilizer P resulted in significant increases in P concentration for maize grown in the three soils (Tables 2 and 3). This increase in P concentration as a function of increasing P rate resulted in increases in maize stem width in two soils and a biomass increase in one. The general trend of the significant measurements taken across all soils was curvilinear, with peaks occurring typically at 20-40 kg P ha\(^{-1}\) and then generally a plateau thereafter (Table 3).

Responses to P source were more numerous and generally greater for soil 1a than for the other two soils. Soil 1a is a highly calcareous soil (12% CaCO\(_3\)) with low soil test P (7 µg g\(^{-1}\)). Comparatively, soils 1b and 2 contained 7 and 14 µg g\(^{-1}\) soil test P and 50% and 73% less CaCO\(_3\), respectively. In soil 1a, the CBP fertilizer source resulted in significantly greater P concentration than with APP and also resulted in greater maize stem width and shoot biomass (Tables 2 and 4). The increase in P concentration is especially impressive given the potential dilution impact from producing more biomass. The CBP source also resulted in significant biomass increase in soil 2 (Table 2 and 4). Compared to APP application, there was a trend for increased stem width of maize grown in soil 2 as well, but the magnitude was not as great as in soil 1a and the difference close to being significant \((P = 0.1759)\). Plant height was not impacted by fertilizer source with any of the soils. Important with regard to the objectives of these trials is a significant increase in early season maize biomass yield with CBP compared to APP fertilized plants in two of the three soils (Table 2 and 4). In the other soil (soil 1b), a close examination of a slightly significant rate by source interaction \((P = 0.0831)\) shows a trend similar to the
other two soils with CBP biomass greater than APP at the 10 kg P ha\(^{-1}\) rate but similar at other rates (data not shown).

In soil 1a, the most likely to develop a highly stressed environment for P availability (Table 1), CBP fertilized plants developed significantly thicker stems than APP fertilized plants when averaged across all rates (Tables 2 and 4), but no significant impact of CBP was observed on stem thickness in the other two soils. Examination of the significant interaction between rate and source in soil 1b (Table 2) shows a steep increase in stem thickness with increasing rate with both sources and then a plateau at higher rates (data not presented). Carbond P-treated maize reached maximum stem widths at a lower P rate than APP-treated maize with significantly thicker maize stems at the 20 kg P ha\(^{-1}\) rate where stem widths of APP-treated maize did not peak until 40 kg P ha\(^{-1}\). Stem widths were also numerically thicker with CBP than APP application at both rates (data not shown).

The percent P found in the shoot tissue was significantly higher in CBP fertilized plants than APP fertilized plants grown in soil 1a (Tables 2 and 4), but there was also a significant interaction between source and rate. These significant increases in percentage P in maize plants fertilized with CBP compared to APP fertilized plants were at the 40, 80 and 160 kg P ha\(^{-1}\) (Figure 1). Specifically, CBP treatments gradually produced increased percentage P tissue concentrations with increasing rates of P while APP treatments never produced higher P concentrations; a slight bump at the 10 kg P ha\(^{-1}\) rate was not significantly different than CBP but remained similar to the control (0.00 % P) at all other rates (Figure 1). Concentrations of other nutrients were not significantly and/or practically impacted by rate or source of P and trends observed were not consistent across
soils. Consequently, average maize nutrient contents of Ca, Mg, K, S, Cu, Fe, Mn, Na, and Zn are reported for each soil (Table 5).

**Field Trials**

Similar to the glasshouse study, there was a highly significant orthogonal contrast between fertilized and unfertilized plant responses for all measured parameters for soil 1 ($P < 0.001$) indicating P limiting soil conditions, but maize grown in soil 2 was not responsive to P fertilization.

Shoot biomass, stem width, plant height, and P uptake were all impacted to varying degrees by P source and rate (Tables 6-8). The P concentration in maize plants was not impacted with increasing P fertilizer rate (Tables 6 and 7). However, increased P application resulted in increases in maize biomass and, consequently, the total amount of P accumulated in the shoots significantly increased at the highest level of fertilization over two of the lowest three P rates (Table 7). Maize plant height and stem width were not impacted significantly as a function of P rate in soil 1, but both were impacted in maize grown in soil 2 (Table 6 and 7).

There were three parameters significantly impacted by P fertilizer source for soil 1 and two for soil 2 (Tables 6 and 7). As previously mentioned, soil 1 is highly calcareous soil (6-12% CaCO$_3$) with low soil test P (7 µg g$^{-1}$) compared to soil 2 (3.3% CaCO$_3$ and 14 µg g$^{-1}$ soil test P). The CBP fertilizer source resulted in significantly higher total P uptake but not higher maize P concentration than APP fertilizer. The CBP also produced thicker maize stems and higher shoot biomass than APP (Tables 6 and 8). Not surprisingly, there was no increase in P concentration, P uptake, or biomass in soil 2, possibly related to higher P soil test (Table 1). Despite high soil test P, stem width and
plant height were significantly increased with CBP compared to APP for maize grown in soil 2. Concentrations of Ca, Mg, K, S, Cu, Fe, Mn, Na, and Zn were not impacted in a consistent and significant manner, with the average concentrations reported in Table 9.

DISCUSSION

Maize biomass and P concentration and/or uptake were consistently equal to or, more frequently, greater with CBP than APP applications. This confirms results obtained in a preliminary glasshouse study (Hill et al., 2011). The CBP never performed worse than APP and frequently resulted in increases in the in-season growth parameters measured in this study. The apparent benefit for CBP over APP was more pronounced under conditions where expected P fertilizer response would be greatest (low soil test P and highly calcareous soil). In general, CBP application resulted in more P measured in the maize shoot tissue. This apparently resulted in greater in-season plant biomass. Although greater plant height was measured in one circumstance, this parameter was not as highly impacted as was stem width. Stem width was significantly thicker with CBP than APP in one soil in the glasshouse trials and in both soils in the field studies (and numerically higher in all soils in these studies). Maize plants deficient in P generally have lower shoot growth rates as carbohydrates are directed to the roots for nutrient exploration (Marschner, 2012; Mengel and Kirkby, 2001). Stem widths and heights were measured as an indirect assessment or estimate of this P deficiency symptom. The results of our study would suggest that P deficiency symptoms were present and that CBP was more efficient at correcting the deficiency than APP. Additional studies are needed to
verify these impacts and to evaluate root biomass and other physiological impacts of CBP on in-season growth.

The strongest early season biomass response to CBP compared to APP was evident in soil 1a (glasshouse). The distinguishing characteristic of this soil compared to the other two soils studied is elevated CaCO$_3$ content (12.1%; Table 1). Increasing CaCO$_3$ content is closely associated with the capacity of a soil to form precipitates with soluble P (Westermann, 1992). In Idaho soils similar to those used in these studies, an additional 11 kg P ha$^{-1}$ for each percent increase in soil CaCO$_3$ is recommended to compensate for the loss of P to adsorption/precipitation reactions for potato production (Stark et al., 2004). The availability of P supplied to the plant from fertilization with APP in Figure 1 illustrates how typical fertilizers may not deliver adequate P to plants grown in CaCO$_3$ rich soils, even at moderately high P rates. The ability of CBP to maintain P solubility, presumably by ameliorating precipitation reactions, is evidenced in the increased maize P concentrations with increasing P rates (Figure 1). This is evidence that CBP is maintaining P in soluble forms longer than APP, which was also shown in preliminary soil laboratory trials (Hill et al., 2013). Similar studies to ours are needed to evaluate the performance of CBP in soils with low pH and/or CaCO$_3$ concentrations. Data from a hydroponic study suggests that CBP delivers more P to maize plants in acid than alkaline conditions and confirms that CBP impacts soil chemistry (and not plant physiology directly; Summerhays et al., 2013b). Studies also show that the effect is likely limited to low to moderate OM soils (Summerhays et al., 2013a).

The field studies on maize expand our observations from early- to mid-season growth responses under glasshouse conditions to actual field conditions. Although the
physiological growth demand for P nutrient uptake needs to be favorable at all stages of growth for optimal yields, observations were made around the V12 (12 leaf vegetative stage) to V18 growth (18 leaf vegetative) stages because of the critical demand that is placed on P supply for achieving maximum grain yields at these stages. During this time, the number of ovules (potential kernels) and size of the ear are determined, and nutrient deficient conditions may seriously reduce yield potential. During these critical periods of growth, enhanced maize P nutrient uptake and biomass yield from CBP compared to APP were observed on soil 1b. This is strong evidence that maize plants are receiving needed P nutrition from CBP for high yields even in the middle of the growing season.

Final grain and total biomass yields were obtained successfully for maize grown in the field trial on soil 2, along with yields of multiple crops and rates of APP and CBP, and showed positive responses (Hopkins et al., 2013). Unfortunately, final yields for soil 1 were lost due to application of CBP to the entire field by the cooperating grower after observing positive visual and other measurements early in the season (70 d). Final grain harvest for maize grown on soil 2 showed a highly significant increase from CBP application compared to APP—a 12% increase in grain yield with CBP at the 15 kg P ha\(^{-1}\) rate and an 8% increase at the 20 kg P ha\(^{-1}\) rate over APP. The CBP-treated maize also produced an 8.4% increase in biomass (silage) compared to APP. It is interesting to note that this responsive site did not have as great of differences in the early season measurements for both glasshouse and field studies as soil 1, yet final yield in the field was significantly impacted by CBP.

These results are largely consistent with additional field trials taken to final yield on potato (*Solanum tuberosum* L.), alfalfa (*Medicago sativa* L.), sugar beet, and dry bean
(Hopkins et al., 2013). In general, CBP outperformed APP when crop plants were grown in low soil test P soils, especially in calcareous conditions. In these studies, the difference between CBP and APP often diminished at high rates of P application. In a few cases where high rates of P were applied, yields using CBP declined compared to APP at the highest rates—this could possibly be attributed to P-induced micronutrient deficiency. This impact is observed at high P rates, but is not easily predicted or well characterized from a management standpoint (Barben et al., 2011; Hopkins, 2012; Nichols et al., 2012).

**CONCLUSION**

Carbond P application frequently resulted in more in-season maize biomass, thicker stems and increased plant P concentration than APP application—especially for P limiting soils (highly calcareous, low soil test P soils). Early season stem thickness and biomass were the most consistently observed enhancements from CBP. Additional data are needed to strongly conclude that enhanced early season growth parameters translate into improvements in final yields in the field.
REFERENCES


### Table 1. Soil Characteristics.
Select soil characteristics for soils used in glasshouse (three soils) and field studies (two soils).

<table>
<thead>
<tr>
<th>Soil Characteristic</th>
<th>Method</th>
<th>Soil 1a</th>
<th>Soil 1b</th>
<th>Soil 2</th>
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<tbody>
<tr>
<td>pH</td>
<td>1:1 Water-Soil</td>
<td>8.2</td>
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<td>EC, dS m⁻¹</td>
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</table>

¹In the field experiment, soil 1 is a combination of soil 1a and 1b with soil 1b dominating the landscape.
### Table 2. Glasshouse Maize Growth Parameters.

Statistical significance (P values) for early-season growth parameters (V6 growth stage) for maize grown in three soils under glasshouse conditions at seven P rates (0, 5, 10, 20, 40, 80 and 160 kg P ha\(^{-1}\)) applied as either ammonium polyphosphate or Carbond P. Values bolded are statistically significant at P < 0.05, ANOVA.

<table>
<thead>
<tr>
<th>Source</th>
<th>Shoot biomass</th>
<th>Stem width</th>
<th>Plant height</th>
<th>P concentration</th>
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<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

------------------------------------ soil 1a -------------------------------

| Rate | 0.2130 | 0.2112 | 0.3465 | 0.0298 |
| Source | 0.0092 | 0.0500 | 0.1232 | 0.0072 |
| Rate*Source | 0.1884 | 0.2725 | 0.3359 | 0.0281 |

------------------------------------ soil 1b ------------------------------

| Rate | 0.0339 | <0.0001 | 0.0756 | <0.0001 |
| Source | 0.9982 | 0.2505 | 0.7396 | 0.6508 |
| Rate*Source | 0.0831 | 0.0271 | 0.2819 | 0.4551 |

------------------------------------ soil 2 --------------------------------

| Rate | 0.5271 | 0.0111 | 0.4960 | <0.0001 |
| Source | 0.0391 | 0.1759 | 0.4369 | 0.5100 |
| Rate*Source | 0.1598 | 0.3537 | 0.1559 | 0.3639 |
Table 3. Glasshouse Maize Growth Parameters by Fertilizer Rate.
Early-season growth parameters (V6 growth stage) for maize grown in three soils under glasshouse conditions at seven P rates (0, 5, 10, 20, 40, 80 and 160 kg P ha⁻¹) applied as either ammonium polyphosphate or Carbond P. Values are the average across fertilizer source and are further adjusted by subtracting values obtained with the control (0 kg P ha⁻¹) from each P treatment. Values followed by the same letter are not statistically significant at $P = 0.05$, Tukey Honestly Significant Difference.

<table>
<thead>
<tr>
<th>Rate, kg P ha⁻¹</th>
<th>Shoot biomass, g pot⁻¹</th>
<th>Stem width, mm</th>
<th>Plant height, cm</th>
<th>P concentration, %</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Soil 1a</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>0.23 a</td>
<td>0.84 a</td>
<td>6.2 a</td>
<td>0.01 b</td>
</tr>
<tr>
<td>10</td>
<td>0.35 a</td>
<td>1.22 a</td>
<td>6.4 a</td>
<td>0.02 ab</td>
</tr>
<tr>
<td>20</td>
<td>0.51 a</td>
<td>1.89 a</td>
<td>10.7 a</td>
<td>0.05 a</td>
</tr>
<tr>
<td>40</td>
<td>0.28 a</td>
<td>0.97 a</td>
<td>4.0 a</td>
<td>0.06 a</td>
</tr>
<tr>
<td>80</td>
<td>0.11 a</td>
<td>0.33 a</td>
<td>3.7 a</td>
<td>0.03 ab</td>
</tr>
<tr>
<td>160</td>
<td>0.23 a</td>
<td>0.91 a</td>
<td>3.1 a</td>
<td>0.05 a</td>
</tr>
<tr>
<td><strong>Soil 1b</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>0.40 b</td>
<td>1.60 c</td>
<td>5.2 a</td>
<td>0.04 c</td>
</tr>
<tr>
<td>10</td>
<td>0.48 ab</td>
<td>2.36 b</td>
<td>3.8 a</td>
<td>0.10 d</td>
</tr>
<tr>
<td>20</td>
<td>0.57 ab</td>
<td>2.98 ab</td>
<td>5.0 a</td>
<td>0.18 c</td>
</tr>
<tr>
<td>40</td>
<td>0.71 a</td>
<td>3.06 a</td>
<td>8.7 a</td>
<td>0.28 b</td>
</tr>
<tr>
<td>80</td>
<td>0.65 ab</td>
<td>2.91 ab</td>
<td>8.8 a</td>
<td>0.30 ab</td>
</tr>
<tr>
<td>160</td>
<td>0.56 ab</td>
<td>2.74 ab</td>
<td>4.9 a</td>
<td>0.34 a</td>
</tr>
<tr>
<td><strong>Soil 2</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>0.21 a</td>
<td>0.00 ab</td>
<td>4.3 a</td>
<td>0.02 c</td>
</tr>
<tr>
<td>10</td>
<td>0.35 a</td>
<td>0.54 ab</td>
<td>4.3 a</td>
<td>0.04 c</td>
</tr>
<tr>
<td>20</td>
<td>0.19 a</td>
<td>0.33 ab</td>
<td>6.6 a</td>
<td>0.07 c</td>
</tr>
<tr>
<td>40</td>
<td>0.36 a</td>
<td>0.86 a</td>
<td>7.2 a</td>
<td>0.14 b</td>
</tr>
<tr>
<td>80</td>
<td>0.28 a</td>
<td>0.15 ab</td>
<td>6.1 a</td>
<td>0.18 ab</td>
</tr>
<tr>
<td>160</td>
<td>0.23 a</td>
<td>-0.49 b</td>
<td>4.9 a</td>
<td>0.23 a</td>
</tr>
</tbody>
</table>
Table 4. Glasshouse Maize Growth Parameters by Fertilizer Source.
Early-season growth parameters (V6 growth stage) for maize grown in three soils under glasshouse conditions at seven P rates (0, 5, 10, 20, 40, 80 and 160 kg P ha⁻¹) applied as either ammonium polyphosphate (APP) or Carbond P (CBP). Values are the average across fertilizer rates and are further adjusted by subtracting values obtained with the control (0 kg P ha⁻¹) from each P treatment. Values bolded are statistically significant at $P = 0.05$, ANOVA.

<table>
<thead>
<tr>
<th>P source</th>
<th>Shoot biomass, g pot⁻¹</th>
<th>Stem width, mm</th>
<th>Plant height, cm</th>
<th>P concentration, %</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>soil 1a</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>APP</td>
<td>0.15</td>
<td>0.53</td>
<td>3.6</td>
<td>0.01</td>
</tr>
<tr>
<td>CBP</td>
<td>0.40</td>
<td>1.44</td>
<td>7.2</td>
<td>0.06</td>
</tr>
<tr>
<td><strong>soil 1b</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>APP</td>
<td>0.56</td>
<td>2.49</td>
<td>6.6</td>
<td>0.21</td>
</tr>
<tr>
<td>CBP</td>
<td>0.56</td>
<td>2.72</td>
<td>5.5</td>
<td>0.21</td>
</tr>
<tr>
<td><strong>soil 2</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>APP</td>
<td>0.21</td>
<td>0.09</td>
<td>4.8</td>
<td>0.12</td>
</tr>
<tr>
<td>CBP</td>
<td>0.33</td>
<td>0.37</td>
<td>6.2</td>
<td>0.10</td>
</tr>
</tbody>
</table>
Table 5. Average Glasshouse Maize Nutrient Concentrations.
Early-season (V6 growth stage) nutrient concentrations for maize grown in three soils under glasshouse conditions at seven P rates (0, 5, 10, 20, 40, 80 and 160 kg P ha⁻¹) applied as either ammonium polyphosphate or Carbond P. Values are the average across fertilizer rate and source.

<table>
<thead>
<tr>
<th>Soil</th>
<th>Ca</th>
<th>Mg</th>
<th>K</th>
<th>S</th>
<th>Cu</th>
<th>Fe</th>
<th>Mn</th>
<th>Na</th>
<th>Zn</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a</td>
<td>1.22</td>
<td>0.73</td>
<td>4.0</td>
<td>0.22</td>
<td>9.4</td>
<td>115</td>
<td>124</td>
<td>103</td>
<td>41</td>
</tr>
<tr>
<td>1b</td>
<td>0.92</td>
<td>0.67</td>
<td>3.5</td>
<td>0.25</td>
<td>11.5</td>
<td>95</td>
<td>113</td>
<td>92</td>
<td>42</td>
</tr>
<tr>
<td>2</td>
<td>0.65</td>
<td>0.23</td>
<td>5.2</td>
<td>0.26</td>
<td>8.4</td>
<td>43</td>
<td>101</td>
<td>103</td>
<td>52</td>
</tr>
</tbody>
</table>
**Table 6. In-Field Maize Growth Parameters.**
Statistical significance ($P$ values) for in-season growth parameters (V12-R2 growth stages) for maize grown in two fields in South-Central Idaho at seven rates of P for soil 1 (0, 1.4, 3.3, 7.6, 17.6, 40.4, and 93 kg P ha$^{-1}$) and four rates of P for soil 2 (0, 10, 15, and 20 kg P ha$^{-1}$) applied as either ammonium polyphosphate or Carbond P. Values bolded are statistically significant at $P = 0.05$, ANOVA.

<table>
<thead>
<tr>
<th>Source *</th>
<th>Shoot biomass</th>
<th>Stem width</th>
<th>Plant height</th>
<th>P concentration</th>
<th>P uptake</th>
</tr>
</thead>
<tbody>
<tr>
<td>soil 1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Model</td>
<td>0.0081</td>
<td>0.0027</td>
<td>0.2904</td>
<td>0.0001</td>
<td>0.0020</td>
</tr>
<tr>
<td>Rate</td>
<td>0.0140</td>
<td>0.1034</td>
<td>0.2826</td>
<td>0.1537</td>
<td>0.0286</td>
</tr>
<tr>
<td>Source</td>
<td>0.0015</td>
<td>0.0001</td>
<td>0.1528</td>
<td>0.2541</td>
<td>0.0082</td>
</tr>
<tr>
<td>Rate*Source</td>
<td>0.6903</td>
<td>0.1165</td>
<td>0.4517</td>
<td>0.4764</td>
<td>0.3366</td>
</tr>
<tr>
<td>soil 2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Model</td>
<td>0.7043</td>
<td><strong>0.0003</strong></td>
<td><strong>0.0029</strong></td>
<td>0.3150</td>
<td>0.5894</td>
</tr>
<tr>
<td>Rate</td>
<td>0.9288</td>
<td><strong>0.0032</strong></td>
<td><strong>0.0449</strong></td>
<td>0.4019</td>
<td>0.8723</td>
</tr>
<tr>
<td>Source</td>
<td>0.9202</td>
<td><strong>0.0002</strong></td>
<td><strong>0.0217</strong></td>
<td>0.6078</td>
<td>0.9332</td>
</tr>
<tr>
<td>Rate*Source</td>
<td>0.8293</td>
<td>0.6779</td>
<td>0.5858</td>
<td>0.2502</td>
<td>0.6848</td>
</tr>
</tbody>
</table>
Table 7. In-Field Maize Growth Parameters by Fertilizer Rate.
In-season growth parameters (V12-R2 growth stages) for maize grown in two fields in South-Central Idaho at seven rates of P for soil 1 (0, 1.4, 3.3, 7.6, 17.6, 40.4, and 93 kg P ha\(^{-1}\)) and four rates of P for soil 2 (0, 10, 15, and 20 kg P ha\(^{-1}\)) applied as either ammonium polyphosphate or Carbond P. Values are the average across fertilizer source and are further adjusted by subtracting values obtained with the control (0 kg P ha\(^{-1}\)) from each P treatment. Values followed by the same letter are not statistically significant at \(P = 0.05\), Tukey Honestly Significant Difference.

<table>
<thead>
<tr>
<th>Rate, kg P ha(^{-1})</th>
<th>Shoot biomass, g plant(^{-1})</th>
<th>Stem width, mm</th>
<th>Plant height, cm</th>
<th>P concentration, %</th>
<th>P uptake, g plant(^{-1})</th>
</tr>
</thead>
<tbody>
<tr>
<td>soil 1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>9.5(^{ab})</td>
<td>2.9(^{a})</td>
<td>1.3(^{a})</td>
<td>-0.006(^{a})</td>
<td>2.5(^{ab})</td>
</tr>
<tr>
<td>3</td>
<td>7.3(^{b})</td>
<td>3.1(^{a})</td>
<td>1.7(^{a})</td>
<td>-0.010(^{a})</td>
<td>1.3(^{b})</td>
</tr>
<tr>
<td>8</td>
<td>9.0(^{b})</td>
<td>3.1(^{a})</td>
<td>1.7(^{a})</td>
<td>-0.013(^{a})</td>
<td>1.8(^{b})</td>
</tr>
<tr>
<td>18</td>
<td>16.3(^{ab})</td>
<td>3.5(^{a})</td>
<td>-2.5(^{a})</td>
<td>-0.025(^{a})</td>
<td>2.9(^{ab})</td>
</tr>
<tr>
<td>40</td>
<td>13.0(^{ab})</td>
<td>3.5(^{a})</td>
<td>7.6(^{a})</td>
<td>-0.012(^{a})</td>
<td>3.2(^{ab})</td>
</tr>
<tr>
<td>90</td>
<td>23.1(^{a})</td>
<td>5.0(^{a})</td>
<td>4.2(^{a})</td>
<td>-0.013(^{a})</td>
<td>6.2(^{a})</td>
</tr>
<tr>
<td>soil 2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>-3.4(^{a})</td>
<td>1.8(^{a})</td>
<td>63(^{a})</td>
<td>0.032(^{a})</td>
<td>-0.1(^{a})</td>
</tr>
<tr>
<td>15</td>
<td>-2.1(^{a})</td>
<td>1.8(^{a})</td>
<td>61(^{ab})</td>
<td>0.030(^{a})</td>
<td>0.5(^{a})</td>
</tr>
<tr>
<td>20</td>
<td>-3.2(^{a})</td>
<td>0.9(^{b})</td>
<td>59(^{b})</td>
<td>0.018(^{a})</td>
<td>-0.3(^{a})</td>
</tr>
</tbody>
</table>

60
Table 8. In-Field Maize Growth Parameters by Fertilizer Source.
In-season growth parameters (V12-R2 growth stages) for maize grown in two fields in South-Central Idaho at seven rates of P for soil 1 (0, 1.4, 3.3, 7.6, 17.6, 40.4, and 93 kg P ha\(^{-1}\)) and four rates of P for soil 2 (0, 10, 15, and 20 kg P ha\(^{-1}\)) applied as either ammonium polyphosphate (APP) or Carbond P (CBP). Values are the average across fertilizer rate and are further adjusted by subtracting values obtained with the control (0 kg P ha\(^{-1}\)) from each P treatment. Values bolded are statistically significant at \(P = 0.05\), ANOVA.

<table>
<thead>
<tr>
<th>Rate, kg P ha(^{-1})</th>
<th>Shoot biomass, g plant(^{-1})</th>
<th>Stem width, mm</th>
<th>Plant height, cm</th>
<th>P concentration, %</th>
<th>P uptake, g plant(^{-1})</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>soil 1</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>APP</td>
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<td>2.5</td>
<td>4</td>
<td>-0.011</td>
<td>1.83</td>
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<tr>
<td>CBP</td>
<td>18</td>
<td>4.3</td>
<td>1</td>
<td>-0.016</td>
<td>4.17</td>
</tr>
<tr>
<td><strong>soil 2</strong></td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>APP</td>
<td>-3</td>
<td>1.0</td>
<td>3</td>
<td>0.024</td>
<td>-0.01</td>
</tr>
<tr>
<td>CBP</td>
<td>-3</td>
<td>2.0</td>
<td>6</td>
<td>0.029</td>
<td>0.10</td>
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</tbody>
</table>
Table 9. Average In-Field Maize Nutrient Concentrations.
In-season (V12 growth stage) nutrient concentrations for maize grown in two fields in South-Central Idaho at seven rates of P for soil 1 (0, 1.4, 3.3, 7.6, 17.6, 40.4, and 93 kg P ha\(^{-1}\)) and four rates of P for soil 2 (0, 10, 15, and 20 kg P ha\(^{-1}\)) applied as either ammonium polyphosphate or Carbond P. Values are the average across fertilizer rate and source.

<table>
<thead>
<tr>
<th>Soil</th>
<th>Ca</th>
<th>Mg</th>
<th>K</th>
<th>S</th>
<th>Cu</th>
<th>Fe</th>
<th>Mn</th>
<th>Na</th>
<th>Zn</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.77</td>
<td>0.48</td>
<td>2.21</td>
<td>0.19</td>
<td>7.78</td>
<td>109</td>
<td>125</td>
<td>168</td>
<td>57</td>
</tr>
<tr>
<td>2</td>
<td>0.97</td>
<td>0.61</td>
<td>2.37</td>
<td>0.16</td>
<td>6.17</td>
<td>153</td>
<td>69</td>
<td>217</td>
<td>115</td>
</tr>
</tbody>
</table>
Figure 1. Concentration of P (%) was measured at the V6 growth stage of maize after banded treatments of 0, 5, 10, 20, 40, 80 and 160 kg P ha\(^{-1}\) as either Carbond P or ammonium polyphosphate in a glasshouse study. Values are for soil 1a. Statistics use values adjusted by subtracting values obtained with the control (0 kg P ha\(^{-1}\)) from each treatment. Error bars were constructed using the standard errors.
Prospectus

TITLE IMPROVING PHOSPHORUS USE EFFICIENCY THROUGH ORGANIC ACID BONDING

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M.S. Candidate
Department Plant and Wildlife Science
Brigham Young University
Provo, Utah

PROBLEM STATEMENT
Current maximum efficiency of phosphorus fertilizers that is utilized by plants in the same year of application ranges from near zero to 30%. Innovative technology must be developed to more efficiently deliver phosphorus fertilizer to plants since fertilizer demands are increasing, while phosphorus mineral resources are rapidly being depleted. A growing environmental concern for nutrient pollution of surface waters also carries significant weight for this movement. A novel product called Carbond® P holds promising technology to increase this efficiency. Research is needed to understand its capabilities and functionality mechanisms imbedded within the technology.
INTRODUCTION

Phosphorus (P) is an essential nutrient for plant growth and development (Bennett, 1993). However, inefficient P uptake is a common growth impediment. Chemical reactions within the soil hold much of the P unavailable to plants, and often release the P slower than the plant needs. From crops to sports turf grass, managers are faced with the inherent inefficiencies of P fertilization. Here, we will discuss the importance of P in the physiology of plants, how natural processes in the soil inhibit plant uptake, and finally present the technology behind a novel fertilizer that may enhance plant P uptake efficiency (Carbond P). Finally, we will propose experimentation to understand its capabilities as well as understand the unique underlying chemistry that sets it apart from other fertilizer products.

Phosphorus in Plant Physiology

Phosphorus (P) is an essential plant nutrient (Bennett, 1993) and is one of the three most commonly deficient nutrients (along with nitrogen and potassium) for plant growth and development. P is a component of DNA, RNA as well as the phospholipids that make up plant membranes (Tiaz, 2006). P plays a key role in plant bioenergetics. It is a component of adenosine triphosphate and adenosine diphosphate which is required for the conversion of light energy to chemical energy during photosynthesis. Adenosine triphosphate can also be used for biosynthesis of many molecules and, as such, is important for plant growth and reproduction. It is also used to modify the activity of various enzymes by phosphorylation and can be used in cell signaling.

In an agricultural setting, the heavy P demand required by plants to obtain perpetual maximum crop yields is deficient in most soil conditions. The lack of plant available P in the soil is common issue set by three primary reasons. First, P content in soils is often relatively low.
Secondly, the P compounds common in soils are poorly soluble and as a result are only limited in their availability for plant uptake. Lastly, when soluble sources of P are added to soils, they immediately begin forming poorly soluble compounds within the soil matrix (Brady, 2010). Understanding how P evolves in the soil, and in what forms plants take it up is essential for management of high yielding plants.

**Phosphorus in the Soil**

Phosphorus in the soil exists in both liquid and solid phases, with the amount of $\text{H}_x\text{PO}_4^{x-3}$ (or phosphates) dissolved in the soil solution existing in rapid equilibrium with the solid phase P forms. Mengel and Kirkby (2001) put soil P into three categories with relation to plant accessibility. The first group is the solution P (or liquid phase phosphates) which are readily available to plants. However, this makes up only a minute fraction of the total soil P. More than 90% of P in the soil is in non-labile solid phases, inaccessible to plants during the growing season. These forms include primary phosphate minerals, humus P, insoluble phosphates of Ca, Fe and Al and P precipitated by hydrous oxides, and silicate minerals. This fraction of soil P is released only very slowly and can be thought of as P in the bank, which can be used in future years. The third group of P is somewhere in the middle of the previously mentioned. It is referred to as the labile P, and is in solid phase forms that are semi-soluble. Kirkby and Mengal classify these as sparingly soluble phosphates of Ca in calcareous and alkaline soils, of Fe and Al in acidic soils. Also included are strongly adsorbed phosphates by Fe and Al hydrous oxides, occluded phosphate by Fe and Al compounds and organic phosphorus. The phosphate concentration (or equilibrium of liquid and solid P forms) depends on soil pH, as well as the types and quantities of solid phase minerals and organic constituents that make up the soil. Generally speaking, the highest solubility of phosphorus is found between soil pH ranges of 6-7,
and above 9. High pH soils can have a large quantity of apatite (aka rock phosphate) and yet have very little solution phase phosphorus available for plant uptake. The solubility of other mineral forms of P is also highly pH dependent and generally has higher soil solution equilibrium than apatite. The presence of large quantities of non-mineral forms of P (such as manure) tends to have relatively greater soil solution P concentration, often with a very long memory effect. It is common to find soils with a history of heavy manure application to have elevated solution P concentrations many decades after the fact.

As plants take up P in the soil solution, the solid phase P compounds in the labile and non-labile pools are no longer at equilibrium. The most soluble forms in the labile pool will begin to dissolve slowly until equilibrium is reached. The rate of dissolution has a large impact on whether or not plants are able to take up adequate P to meet their needs. The reverse is true when highly soluble forms of P fertilizer are added to soil, with the P precipitating to a mineral form until equilibrium is reached. Phosphate precipitation reactions are kinetically biphasic; or known to occur rapidly upon initial P additions and slow as they reach equilibrium of fixed P with solution P (Pierzynski, 2005; Larsen, 1959). Several experiments show these reactions reach equilibrium in approximately 48 hours (Larsen 1959; Black 1943). This relatively short time frame of temporarily increased soil solution P and somewhat static nature of the equilibrium greatly contributes to the inhibition of plant P uptake throughout the growing season. In fact, plants only utilize from near zero to about 30% of P applied in P fertilizer amendments in the first year after application (Hopkins, 2012; Randall et al., 1985). Despite the inherent inefficiencies with P fertilization, effectual delivery of P from fertilizers to crops is integral for maximizing crop yields (Hopkins et al., 2008). As a result, growers find it necessary to apply superfluous amounts in order to meet crop demands.
Why Improve Phosphorus Use Efficiency?

Increasing the percentage of P from fertilizer that is utilized by plants (P use efficiency or PUE) is important not only for reaching high crop yields, but also because P is derived from limited nonrenewable mineral resources and can be a source of nutrient pollution in surface waters (Davenport et al., 2005; Mueller and Dennis, 1996; Sharpley et al., 2003). It has recently been projected that rock phosphate resources will be depleted within the next 50-100 years (Marschner 2012; Cordell et al., 2009; Gilbert, 2009). P as a nutrient pollutant is transported through the environment predominately attached to soil particles. Soil erosion carries P into stream ways and ultimately coastal waters. Once in water ways, P combines with nitrogen (also a nutrient pollutant in more abundance in freshwater systems). Together these nutrients create environmental conditions for eutrophication, or growth of algae and other water plants. As these water plants use up much of the oxygen in the water, a phenomenon called hypoxia occurs. The concentrations of oxygen within the water are so low that it can no longer sustain life of aquatic fishes and invertebrates (Pierzynski, 2005). Under these conditions, ecological diversity is reduced, putting a major strain on aquatic and related ecosystems. Improving PUE could dramatically reduce eutrophication of most freshwaters where biological productivity is limited by P, not nitrogen (Pierzynski, 2005). Although important, environmental reasons alone have not been the sole drivers to improving PUE. Narrow profit margins and high fertilizer costs in the agricultural industry are also major economic incentives (Hopkins, 2008). Studies have furthermore shown antagonistic effects on plants when fertilized with excessive amounts of P. The excess P has, at times, interacted with soil micronutrients by negatively affecting yields (Barben, 2010; Marschner, 2012; Mengel and Kirkby, 2001). Finding better ways to efficiently
increase plant P uptake without overloading the soil system with excess P will be vital to the long term sustainability of agriculture.

A number of rate, timing, and placement options can be used to improve PUE (Hopkins and Ellsworth, 2005; Hopkins et al., 2010a, b, c). In addition to the cultural practices that may enhance P uptake and utilization, fertilizer manufacturers have sought to engineer materials to enhance plant P uptake. One such approach has been to supply organic acids to P fertilizer as an additive. Several acids have been used including humic and fulvic which have attracted a lot of attention. The literature is full of mixed reviews on the efficiency of such products and will be reviewed later in this paper. Although the use of organics has proven promising in many cases, several products have been dubbed “snake oils” because the product performs with a high level of variability. The science may be real, but issues with manufacturing, heterogeneity in the composition of organics used, as well as the inability to precisely characterize dominant organic substances such as humic and fulvic acids have turned soil scientists at large away from this field of study.

**Carbond® P**

Land View Fertilizer Inc. (Rupert, ID) has developed a P fertilizer (Carbond® P or CBP) that incorporates the use of organic acids through a sophisticated manufacturing process. The product is not an additive that goes on with a fertilizer product, but is a premixed solution that holds the P in a soluble form within the product. Their claim is that they are able to get more P into plants using less total applied P. They believe they have incorporated a variety of organic molecules to help preserve the P in a soluble state across a variety of soil types longer than traditional fertilizers such as ammonium polyphosphate (10-34-0). Preliminary studies showed highly significant increases in P solubility and mobility up to 365 days across several different
soil textures, mainly high in pH. The apparent longevity of soil soluble P found with CBP over established fertilizers gave rise to our curiosity with this product. Because of proprietary restrictions, the exact organic components utilized by CBP technology cannot be divulged, except that humic and fulvic acids are incorporated. However, organic acids commonly studied in relationship to P fertilization perform many overlapping functions in keeping the P more soluble in the soil solution.

Based on this information about Carbond P, the major knowledge gap we face is what makes Carbond P unique to other traditional and organic-based fertilizers? Discussion of the theory behind the use of organic acids in P fertilization, as well as revealing where this field of science is today will create the foundation needed to present experimentation that will help disprove or validate Land View’s claims. Additionally, it will assist in discovering possible underlying mechanisms for the unique job Carbond P plays in P fertilization.

**Organic Acids**

Plants deficient in P have been shown to up-regulate root exudation of organic acids into the soil (Grierson, 1992; Zhang, 1997). It is well documented that various organic acids help to mobilize poorly soluble mineral nutrients (with citrate, malate and oxalate that appear to be the most common and effective at mobilizing P) (Oburger, 2009; Hoffland, 1992). Their ability to reduce P precipitation (Grossl and Inskeep, 1991) and even solubilize poorly soluble phosphates (Singh, 1998) is potentially valuable in obtaining sufficient plant P demands. However, the effectiveness of each organic acid in keeping P accessible to plants is based on: 1) functional-group content and geometry (Grossl and Inskeep, 1991), 2) pH (Kirk, 1999), 3) affinity with the adsorption sites (Strom et al., 2002), and 4) ability to withstand degradation (Andrade, 2007). As a variety of organic acids are incorporated into CBP technology, all are theoretically working in
concert to achieve the previously mentioned benefits, but only vary in their degree of effectiveness based on the parameters previously given. Humic and fulvic acids comprise a considerable fraction of the organic acids included in CBP, and since they are the only known organic components, much attention will be given to their specific roles in keeping P readily available for plant uptake.

**Humic Matter and Terminology**

Humic substances (primarily macromolecules of humic and fulvic acids) exert a number of important functions such as slow release of plant nutrients, pH buffering, and interaction with micronutrients, toxic metal ions and xenobiotic organic molecules (Brunetti 2007; Stevenson 1994; Senesi and Loffredo, 1999; Hays and Clapp, 2001; Clapp et al., 2005). They are the most chemically active compounds in soils, with electrical charges and exchange capacities exceeding those of the clay minerals (Tan, 2003). They can sorb to the surfaces of clays and oxides, blocking P sorption sites, leaving more P in the soil solution (Pierzynski, 2005; Grossl and Inskeep, 1991; Larsen, 1959). Andrade (2007) mentions that one of the major criticisms for using organic acids to help keep P free in the soil solution is that most organic acids are transitory in nature and mineralize quickly. Many of the experiments are, therefore, carried out only for a few weeks. However, Andrade (2007) has shown that humic acids can withstand degradation over longer periods of time and maintain their beneficial functions in keeping P free in the soil solution compared to smaller, less stable organic acids, such as citric acid and acetic acid. There appears to be benefits, but the discussion of humic matter often sends scientists running for cover.

**Obstacles Working With Organic Acids**
The perplexing nature of humic matter structure, chemistry, formation theory (humification) as well as disagreements in terminology has turned soil scientists at large, away from the study, thus slowing work on humic substances and soil interactions among soil scientists (Tan 2003). The subject continues to stay quite controversial. Nonetheless, humic matter chemistry is making great progress in other disciplines and much is understood. For the purposes of this paper, the terminology of Tan (2003) will be used. The following terms are often used interchangeably in scientific literature: organic matter, humus, humic matter, humic substances and humic acids. However, the overlapping usage of such terms has created ambiguity among scientists when trying to pinpoint exactly what the other is talking about. Each term has a distinctive definition from the other, and should be used accordingly to avoid further confusion. Figure 1 displays a flow chart of these terms and their relationships.

Soil organic matter comprises the living and dead organisms or parts of organisms. Humus is essentially the dead organic matter that can no longer be identified visually from its original organic source. The non-humified fraction of humus contains molecules synthesized by living organisms such as carbohydrates, amino acids, lipids, etc. Humic substances (those gone through the humification process, of which many theories exist) do not contain peptides, sugars, nucleic acids and the like (Tan, 2003). These substances are comprised of humic acids, fulvic acids, hymatomelanic acids and humin to name the basic contributors. Humic matter is composed of high molecular weight organic colloids comprised of humic substances and bears no morphological resemblances to the original plant and animal debris which provided the source materials (Clapp et al. 1993). Generally speaking, high quality sources of humic and fulvic acids come as humic matter. They are found together as a result of the humification process in fairly consistent ratios which will be discussed later.
Origin of Humic Matter

Humic matter is assumed to be the most widely distributed organic carbon containing material on the earth’s surface, being present in: soils, water of streams, lakes and oceans, and in their foam and sediments, from the tropics to the arctic regions (Tan 2003). Although there are many sources, the focus here will be upon the geological deposits of leonardite since it is common and the source used for CBP. High quality leonardite (lignite) tends to be 80-90% humic matter, with a prevalence of humic acid and minimal fulvic acid content (Tan 2003). The formation of leonardite occurs under the metamorphism process where high pressure and temperature induce the compaction and drastic chemical changes of ancient bog, peat and muck sites (Miller and Gardiner, 1998; Tan 2003).

The poorly understood process by which humic matter is formed is called humification. The principal discrepancies between the common theories differ in the way the sources of original or raw materials are utilized in the synthesis of humic substances, namely: humic substances formed as a result of a decomposition process or as a gathering of decomposed monomers. It is agreed that the materials for formation originate mostly from plant material (or animal residue) including lignin, cellulose and hemicellulose, proteins, phenols and amino sugars synthesized by microorganisms (Tan 2003).

Structural Components

There are varying models that attempt to show the structural composition of humic or fulvic acids. Most of the humic fractions of soil organic matter are defined on an operational basis (Felbeck 1965) and therefore can be argued that these acids are created in the extraction processes. Some leading scientists believe that there is no consistent chemical structure (Felbeck, 1965) nor can there be since these substances are not biologically predestined to carry out a
specific biochemical action and therefore cannot be defined in functional terms (Aiken et al. 1985). They are most frequently described as being exceedingly complex and of unknown composition (Stevenson 1994). Clapp et al. 1997 explains them as macromolecules with varying chemical structures depending on the origin of the humic acid.

The major elements involved in humic/fulvic acids are C, H, O, N, and S and the percent composition of each element (56.2, 4.7, 35.5, 3.2, and 0.8 respectively for humic acid) remains within relatively fixed limits (Tan, 2003). Tan 2003 refers to differences in variation in the elemental composition of extracted humic/fulvic acids as affected by: variability in soils, variability of humic substances in time and space, different isolation techniques, and errors in sampling and analyses. As technology advances, this debate of whether or not humic substances actually exist as replicable molecules or not is becoming more clear. For example, Schulten (2001 2002) has presented a replicable molecular structure of a humic substance.

**Functional Groups**

The functional groups that make up fractions of humic substances are very important in determining their structural properties and the chemical reactions they will be involved in as a result. The main functional groups in humic and fulvic acids are carboxyl, carbonyl, aliphatic hydroxyls, phenolic hydroxyls, and amine groups (Stevenson 1994). The protonation of amino groups will create positive charges (Tan 2000), making them amphoteric compounds (carry both positive and negative charges) but this reaction is expressed mainly in extremely low pH soils of less than 4. Humic and fulvic acids produce much of their negative charges through deprotonated carboxylic groups in normal pH conditions, as well as deprotonated phenolic hydroxyl groups at high pH (Mengal and Kirkby 2001), which permit them to act as weak bases (Stevenson 1994). These are also the most important functional groups with regards to inhibiting P precipitation in...
the soil. The carboxyl and phenolic alcohol groups can complex or chelate P fixers like Fe, Al, and Ca. They can also occupy clays and oxides through water and metal bridging. The same reaction can occur to hold phosphate in a weak bridging bond for short term phosphate storage.

These functional groups are present in both humic and fulvic acids, but the percent composition of each group and relative size of molecules differs between humic and fulvic acids. The distinction of each acid is made primarily in the processes that isolate the two from the original humic matter source (i.e. lignite). Humic acid is the fraction of humic substances that is not soluble in water under acid conditions (below pH 2) but increases in solubility at greater pH. Fulvic acid is the fraction that is soluble under all pH conditions while humin is not soluble in water at any pH value (Aiken et al. 1985).

The rough molecular weights of soil humic substances range from 2,000 or 3,000 for alkali-soluble, acid-soluble fractions to well over 300,000 for the fractions insoluble in both alkali and acid (Felbeck 1965). Tan 2003 has compiled a comprehensive review of humic and fulvic acids extraction protocols, and describes the major differences between humic and fulvic acids which primarily pertain to their size and functional group composition. Again, there are major discrepancies in the literature but Tan 2003 depicts the current understanding of these issues well.

**Application to Carbond® P**

It is apparent that there are many uncertainties that exist when discussing the commercial use of humic/fulvic acids. This becomes a very important detail when considering the consistency of a product like CBP. Not only could there be variability in the sources from where these materials are derived, but the ratios of both acids, filtration of the impurities that are mined alongside of the humic and fulvic acids, and even the structural composition of one humic acid to
another may be variable. This could be a major factor in why we do not see humic/fulvic acid based fertilizers going mainstream in the agricultural industry. With that said, it could simply be a matter of the humic and fulvic acids acting alone are not enough to keep P free in the soil solution over long periods of time under current farming practices. The unique capacities of CBP found in preliminary studies as well as field observations have led us to believe there is some exclusive job CBP is performing that other fertilizers are not. One possible explanation revolves around the variety of organic acids used in CBP. The fact that they use different size molecules with a variety of functional groups blended in some correct ratio in a sophisticated manufacturing process may be the key.

The purpose of the following experiments will be to explore the underlying chemistry that could explain the previously mentioned functionality claims of CBP technology. Also, to examine the efficiency claims of increased P uptake with less P applied, giving rise to the validity, or error of these claims. Finally, monitor physiological responses throughout the experiments as well as yield potential which are major signs of PUE. These studies will be conducted through a series of laboratory, greenhouse and field experiments.

**STUDY OBJECTIVES**

1) Suggest possible mechanisms to explain the underlying chemistry of Carbond® P. Determine if there are chemical bonds being formed between the organic molecule components in the Carbond® P formulation and P.

2) Ascertain mobility of Carbond® P and water soluble P concentrations through the soil profile.

3) Investigate plant availability and the efficiency factor of applied Carbond® P fertilizer.
4) Explore Carbond® P as a means to ameliorate rapid precipitation of P fertilizer with the soil medium and increase phosphorus use efficiency.

**HYPOTHESES AND EXPERIMENTS**

Hypothesis 1: Based on the functional groups of humic and fulvic acids and the nature of phosphoric acid, there is a chemical bond being formed between the P and organic acids.

Hypothesis 2: Mobility of P will be enhanced with Carbond® P since the P bonding with organic molecules will prevent precipitation reactions with the surrounding soil environment.

Hypothesis 3: Carbond® P will reach maximum fertilizer performance at lower application rates than ammonium polyphosphate.

Hypothesis 4: We will see increased PUE with Carbond® P through increased percent tissue P, in-season growth measurements as well as final crop yields.

**Experiment 1: Confirmation of chemical bonding of P to organic molecules.**

Details of the materials, methods and results for this experiment are kept confidential for proprietary purposes. The use of nuclear magnetic resonance has confirmed that there is chemical bonding between organic molecule components within the Carbond product.

**Experiment 2: Water Soluble P and Mobility Trial**

Ammonium polyphosphate (APP; 10-34-0), monoammonium phosphate (MAP; 11-52-0), and Carbond® P (CBP; 7-24-0) sources will be evaluated in a laboratory soil column mobility study. The fertilizers will be applied as either a banded or mixed application to three different soil types (310 g alkaline sand, 310 g calcareous sand, and 245 g calcareous loam) at 20 or 30 kg P ha⁻¹ and compared to an untreated control. Soil will be placed in 0.18 m by 0.05 cm diameter soil columns and packed in uniform bulk density similar to native soils. Soil will then be retained
in soil columns through use of DeWitt weed-barrier landscape fabric (Sikeston Missouri) fastened securely to the bottom. The banded treatments are to be injected 5 cm below the soil surface and mixed treatments applied to the bulk quantity of soil and mixed thoroughly before packing into columns. Three replicates of each treatment will be established in a complete randomized block design. After fertilizer placement or mixing, each soil column will be initially wetted to field capacity with minimal leaching and then cycled between this moisture level and a nearly constant mass air-dry state throughout the trial—avoiding loss of moisture from the bottom of the column (120ml water). Mobility of P will be evaluated at 24, 48, and 110 daf by applying two pore volumes of water to each of the soil columns and collecting the leachate after passing through a Whatman #2 filter. After 110 d, each soil will be removed from the column and mixed thoroughly to simulate a tillage operation and then repacked into each column. Soil columns are to be stored in a building without heat or insulation to simulate natural field conditions with freezing and thawing over the duration of the winter season. As with the initial extraction conditions, soils will continue to be periodically wetted thus simulating normal cycles of wetting and drying. After 365 d of treatment (original placement or mixing), soils will be leached for a final time. Following this final leaching, soils will be removed from soil columns and mixed thoroughly before being extracted with sodium bicarbonate (Olsen et al., 1954). The P and other nutrient concentrations from each collected leachate and the P from the soil extraction was analyzed by inductively coupled plasma spectroscopy (Johnson and Ulrich, 1959; ICP, Thermo Electron Corporation, Franklin, Maryland, USA) Statistical analyses will be performed using Statistical Analysis System (Version 9.1, SAS, SAS Institute, Cary, North Carolina, USA). Data will be analyzed using analysis of variance (ANOVA) with mean separation using Duncan Waller Multiple Range Test at the 0.05 level of significance.
Experiment 3: Corn Glasshouse P Use Efficiency Factor Study

Three early season growth studies on maize will be conducted simultaneously in a glasshouse. In each study, six replications of each treatment will be arranged in randomized complete block designs. The objective is to evaluate differences in early season PUE of APP and CBP fertilizers. The trial will be performed on three soils low in bioavailable P concentration and ranging from moderately to highly calcareous. Nitrogen will be balanced across all rates with urea. Equal amounts of other nutrients will be applied based on soil test results. Best management practices for plant growth in a glasshouse will be observed.

Targeted daytime and nighttime air temperatures will be 25°C and 15°C, respectively. Natural light will be supplemented with artificial light (high pressure sodium lamps) to maintain a 14/8-hour light/dark cycle.

Soils (1.5 kg) will be placed in elongated square pots (10 cm wide x 25 cm depth) lined with landscape fabric to prevent soil loss yet allow drainage. The containerized soil will be tapped repeatedly to achieve uniform soil depths (soil filled to approximately 5 cm from the top of the pot). Soil (7.5 cm) will then be displaced and P fertilizer treatments were applied in a concentrated band. Seven rates of each fertilizer source will be applied based on 50 cm row spacing (0, 5, 10, 20, 40, 80, and 160 kg P ha⁻¹). Maize hybrid seeds N27B-3111 (presoaked in water 4 h prior to planting) will be planted three seeds pot⁻¹, 2.5 cm deep to achieve a banded placement 2 cm below and 2 cm to the side of the seed.

Plants in each pot will be thinned to two of the healthiest plants after 15 days. Upon final harvest, plants will be measured for height, stem width, chlorophyll content and total shoot biomass. Chlorophyll content will be measured with a Chlorophyll Meter (CCM-200, Opti-Sciences, Tyngsboro, Massachusetts). Total shoot biomass will be determined by destructive
harvest followed by drying to the point of constant weight at 65° C. Shoot tissues will be ground uniformly (Wiley mill, 1-mm sieve), digested in nitric-perchloric acid, and analyzed for nutrient content by inductively coupled plasma spectroscopy (Johnson and Ulrich, 1959; ICP, Thermo Electron Corporation, Franklin, Maryland).

**Experiment 4: Field Studies**

Two field trials comparing CBP and APP on g maize (hybrid, P8107) will be conducted in commercial fields in south-central Idaho in 2011 to confirm/extend the findings of the glasshouse trial. The first trial will be conducted on a 12-ha irrigated in Gooding, Idaho. Plot sizes will be 24-twin rows wide (56-cm row widths) and approximately 600 m long. Control plots (0 kg P ha⁻¹) will be randomly embedded within the treatments with the same width but only 25 m long. Treatments will be placed in a randomized block design replicated three times. Four rates (0, 10, 15 and 20 kg P ha⁻¹) of both fertilizer sources will be applied at planting in a concentrated band 5 cm below and 5 cm to the side of the seed. Same measurements will be taken as in the greenhouse trials at the 12 leaf vegetative growth stage, the second reproductive growth stage, and then again at final harvest (which will include grain yield).

A second field study will be conducted as a small plot study on a fraction of a commercial field in Rupert, Idaho. Seven rates of APP and CBP will be applied (0, 1, 3, 8, 18, 40, and 90 kg P ha⁻¹). Each treatment will be applied to four 7-cm wide twin rows and 9 m long. Treatments will be organized in a randomized complete block design replicated six times.

Fertilizer will be applied by hand in a concentrated band 5 cm below and 5 cm to the side of the seed shortly after plant emergence. The N in the P fertilizer sources used in this trial will be balanced across all treatments with urea ammonium nitrate solution (UAN; 32-0-0) for a total of 62 kg N ha⁻¹. Additional fertilizers (not P) will be applied based on soil test to insure optimum
plant growth for commercial production. Best management practices will be generally followed by the grower. The same measurements will be taken in this field as in the previously mentioned field.
REFERENCES


Figure 1. Flow Chart of humic matter constituents according to Tan 2003.