

Best management systems for intensifying a maize – soybean rotation:
integrating field production, plant physiology, and modeling

by

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B.S., Rio Cuarto National University, 2008
M.S., Rio Cuarto National University, 2014

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Abstract

Potential yield (PY) is defined by the yield limited by temperature, radiation, and genetics – under no limitation on nutrients or water. The difference between PY and actual yield (AY) is defined as yield gap (YG). Management practices such as planting date, row spacing, seeding rate, fertilization program, pest, and disease control can help producers to intensify the productivity of the farming systems and consequently, close the YGs. To evaluate the impact of different management system (MS, specific combination of management practices) on closing the YG the following objectives were established: i) conduct a historical synthesis analysis to characterize shifts in soybean yields, biomass and nutrient uptake and partitioning dissecting the main physiological component related to nutrient use efficiency, seed nutrient composition and nutrient stoichiometry; ii) study the contribution of five MS for intensifying maize-soybean production systems; iii) quantify the nitrogen (N) contribution from the biological N fixation (BNF) process for soybeans under two contrasting MSs (low vs. high inputs); and iv) utilize the same contrasting input treatments to calibrate the Agricultural Production System Simulator (APSIM) for modeling a maize – soybean rotation and apply the parametrized model to estimate a long-term (1980-2016) simulation. For the first objective, main findings indicate that soybean yield increase over time was driven by an increase in biomass with a relatively small variation in harvest index, and with modern varieties producing more yield per unit of N uptake. For the second objective, field experiments demonstrated that intensification practices (narrow row spacing, increasing seeding rate and implementation of a balanced nutrition program) increased yields in both soybeans and maize under rainfed and irrigated conditions. For the third objective, to better understand the soybean N status, BNF measurements were collected during the 2015 growing season and investigated in a greenhouse setting. The B value, N fixation when plants are

fully relying on atmospheric N, changed among varieties, growth stages and plant fractions. Overall B value at R₇ (beginning of maturity) was -1.97 contrasting with the -1.70 value reported as mode according to a literature review. For the range of fixation measured in this research (average of 45-57%), utilization of a B value obtained from the scientific literature or measured in field conditions will have a reduced impact on BNF estimations. Lastly, for the last and fourth objective, the APSIM performed well in estimating yield, biomass production and total N uptake with a high model efficiency and low relative root mean square error (RRMSE). The long-term simulation helped characterize the YG for each crop and MS according to different weather patterns. The modeling approach increased the value of data collected in field experiments. Overall, this research project provided an approach to quantifying and understanding YGs in a maize-soybean rotation and the impact of different MSs on intensifying productivity. Future work can be conducted to model specific MSs to advise producers on the best management systems (BMSs) for sustainably intensifying productivity while minimizing the environmental footprint of current farming systems.

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Dedication

To my late father Ricardo, who unfortunately didn't stay in this world long enough to see his son become a doctor. To my mother Marta, my sisters Cecilia, Laura and Marta and my brother Leandro for their unconditional love, support, and encouragement.

Preface

Potential yield (PY) is defined by the yield limited by temperature, radiation, and genetics – under no limitation on nutrients or water. The difference between PY and actual yield (AY) is defined as yield gap (YG). Management practices such as planting date, row spacing, seeding rate, fertilization program, pest, and disease control can help producers to intensify the productivity of the farming systems and consequently, closing the YGs. The overall dissertation objective was to evaluate the impact of different management systems (MS, specific combination of management practices) on closing the YG on a maize – soybean rotation.

The Primary objectives for each chapter are as follows:

1. to conduct a historical synthesis analysis to characterize shifts in soybean yields, biomass and nutrient uptake and partitioning dissecting the main physiological components related to nutrient use efficiency, seed nutrient composition and nutrient stoichiometry (Chapter 1);
2. to study the contribution of five MS for intensifying maize-soybean production systems (Chapter 2);
3. to quantify the nitrogen (N) contribution from the biological N fixation (BNF) process for soybeans under two contrasting MSs (low vs. high inputs) (Chapter 3);
4. to utilize the same contrasting input treatments to calibrate the Agricultural Production System Simulator (APSIM) for modeling a maize – soybean rotation and apply the parametrized model to estimate a long-term (1980-2016) yield gap (Chapter 4).

Chapter 1 - Shifts in Soybean Yield, Nutrient Uptake, and Nutrient Stoichiometry: A Historical Synthesis-Analysis

Balboa, G. R., Sadras, V. O., and Ciampitti, I. A. (2018). Shifts in soybean yield, nutrient uptake, and nutrient stoichiometry: A historical synthesis-analysis. *Crop Sci.* 58. doi:10.2135/cropsci2017.06.0349

ABSTRACT

Few studies have investigated changes over time in nutrient uptake and yield, in addition to the study of nutrient stoichiometry as a metric of nutrient limitations in soybean [*Glycine max* (L.) Merr.]. A comprehensive synthesis-analysis was performed by compiling a global historical soybean database of yield, total biomass, and nutrient (N, P, and K) content and concentration in studies published from 1921 to 2016. This period was divided in three eras based on genetically modified soybean events: Era I (1921–1996), Era II (1997–2006), and Era III (2006–2015). The main findings of this review are: (i) seed yield improved from 1.3 Mg ha⁻¹ in the 1930s to 3.2 Mg ha⁻¹ in the 2010s; (ii) yield increase was primarily driven by increase in biomass rather than harvest index (HI); (iii) both N and P HIs increased over time; (iv) seed nutrient concentration remained stable for N and declined for both P (18%) and K (13%); (v) stover nutrient concentration remained stable for N, diminished for P, and increased for K; (vi) nutrient ratios portray different trends for N/P (Era I and III > II), N/K (Era I > II and III), and K/P (Era II and III > I); (vii) yield per unit of nutrient uptake (internal efficiency) increased for N (33%) and P (44%) and decreased for K (11%); and (viii) variations in nutrient internal efficiency were primarily explained by increase in nutrient HI for N and K, but equally explained by both HI for P and seed P concentration. These findings have implications for soybean production and integrated nutrient management to improve yield, nutrient use efficiency, and seed nutrient composition.

Keywords: nutrient uptake; nutrient internal efficiency; nutrient ratio; soybean.

INTRODUCTION

Globally, soybean [*Glycine max* (L.) Merr.] is the largest source of both animal protein feed and vegetable oil, providing a variety of nutrients and essential elements important for human health (FAO, 2002). Soybean meal, produced in the crushing and oil extraction process, accounts for 65% of protein feed worldwide. Between 1961 and 2014, global soybean production rose 10-fold to reach >306 million Mg, with an average yield increase from 1.1 to 2.6 Mg ha⁻¹ (FAO, 2017). For the United States, soybean yield gain from 1922 to 2007 was 25 to 30 kg ha⁻¹ yr⁻¹ (Specht and Williams, 1984; De Bruin and Pedersen, 2009; Specht et al., 2014). Global yield increases of 1.3% (current for 2013) will not be sufficient to meet the required production by 2050 (Ray et al., 2013).

The causes of soybean yield improvement included changes in environmental conditions, genetic improvement, management practices, and the interactions among these factors. Increases in atmospheric CO₂ and O₃ concentration, air temperature, and climate variability affected yields in the previous decades (Curry et al., 1995; Grashoff et al., 1995; Southworth et al., 2002). Under elevated CO₂, total biomass in soybean increased proportionally more than yield, with positive changes in the photosynthetic rate and leaf area, and negative changes for harvest index (HI), stomata conductance, and Rubisco activity (Ainsworth et al., 2002). Kucharik and Serbin (2008) showed that increasing summer temperature could potentially decrease soybean yield in the United States by 16%, whereas increased precipitation might produce a counter effect, improving yield by 5 to 10%.

Traits that contribute to improved soybean yield include longer reproductive or seed-filling periods (Gay et al., 1980; Kumudini et al., 2001; Shen and Liu, 2015), decreased lodging (Specht and Williams, 1984), and improved disease resistance (Foulkes et al., 2009). Changes in

management practices that increased soybean yields are related to narrow rows (Heatherly and Elmore, 2004), improvement of weed control (Pike et al., 1991; Osteen, 1993; Bradley and Sweets, 2008; Foulkes et al., 2009), conservation tillage and reduction in harvest losses (Heatherly and Elmore, 2004), and early sowing (Wilcox and Frankenberger, 1987; Conley and Santini, 2007; Bastidas et al., 2008; Sacks and Kucharik, 2011). Early sowing can increase yield by lengthening both the vegetative phase and seed-filling phases (Egli and Cornelius, 2009).

After historical changes in crop yield, variations in nutrient uptake and related nutrient efficiencies have implications for crop and soil management, breeding, and seed quality, as illustrated for sorghum [*Sorghum bicolor* (L.) Moench; Ciampitti and Prasad, 2016], maize (*Zea mays* L.; Ciampitti and Vyn, 2014), wheat (*Triticum aestivum* L.; Guttieri et al., 2017), and legumes and oil seed crops (Sadras, 2006). From a historical perspective, few studies analyzed nutrient uptake, partitioning, and remobilization for soybean; those available took place in the 1930s (Borst and Thatcher, 1931), 1950s (Hammond et al., 1951), 1970s (Hanway and Weber, 1971; Harper, 1971), and 2010s (Bender et al., 2015). The latter study reported maximum yields of 3.5 Mg ha⁻¹ in modern soybean varieties without exploring high yielding levels (>6 Mg ha⁻¹). Gaspar et al. (2017) evaluated nutrient uptake for soybean in the US Midwest in a range of yields (3.6–5.4 Mg ha⁻¹). Neither of the abovementioned studies characterized historical trends of nutrient uptake nor nutrient stoichiometry. Furthermore, published studies on soybean plant nutrition have largely focused on individual nutrients, mostly on N (Fabre and Planchon, 2000; Salvagiotti et al., 2008, 2009; Jin et al., 2011; Rotundo et al., 2014; Van Roekel and Purcell, 2014; Divito et al., 2016; Cafaro La Menza et al., 2017), less on P (Pan et al., 2008; Wang et al., 2009, 2010; van de Wiel et al., 2016), and a few on K (Pettigrew, 2008; Parvej et al., 2015). Nutrient stoichiometry is useful to understand crop nutrient status, and nutrient ratios can be

useful to comprehend nutrient supplies (Mo et al., 2015). Some recent studies have focused on nutrient stoichiometry in soybean (Mallarino et al., 2011; Salvagiotti et al., 2012; Divito and Sadras, 2014; Divito et al., 2016; Tamagno et al., 2017). For example, S/N ratio is a better indicator of soybean S status than S concentration alone (Divito and Sadras, 2014). To the best of our knowledge, there is the gap of an historical analysis that identifies possible shifts in yield and nutrient-related traits that can help to understand soybean plant nutrition. A historical database of soybean yield and uptake and partitioning of N, P, and K was compiled to characterize historical shifts in these traits and dissect the main physiological components related to nutrient use efficiency, seed nutrient composition, and nutrient (N, P, and K) stoichiometry.

MATERIALS AND METHODS

Approach

We pursued a synthesis-analysis, as in previous works (Ciampitti and Vyn, 2012, 2013, 2014; Ciampitti and Prasad, 2016; Tamagno et al., 2017). There is a tradeoff in pursuing a synthesis analysis rather than a meta-analysis. A synthesis-analysis aggregates a large amount of data and summarizes trends but does not provide a quantitative measure of the effect size (Ciampitti and Vyn, 2012). A meta-analysis can calculate the effect size but requires measures of variation (individual replications, standard deviation) that is not reported by treatment in most of the references collected for this study (Curtis and Wang, 1998). Usually this information is not available, restricting the number of datasets that can be included in an analysis.

Data Search Criteria

We focused on time trends in soybean yield, nutrient uptake and nutrient partitioning, nutrient internal efficiency (i.e., yield per unit nutrient uptake), and nutrient stoichiometry. Papers were retrieved from CABI, Web of Science Core Collection, Scopus, Springer Link, Agricola, and Google Scholar using the keywords: “soybean,” “nitrogen,” phosphorous,” “potassium,” “seed yield,” “nutrient uptake,” “nutrient ratio,” “harvest index,” and “internal efficiency.” In addition, unpublished data available to us were included. Only field experiments were included in the database, including those that studied (i) seed yield; (ii) aboveground biomass (total biomass) at maturity (end of the season); (iii) seed N, P, and K uptake or tissue concentration at maturity; (iv) stover (leaf + stem + petiole + pod wall) N, P, and K uptake or tissue concentration at maturity;(v) dry mass and nutrient partitioning HIs; (vi) nutrient internal efficiency; and (vii) N/P, N/K and P/K ratios derived from total nutrient uptake at harvest. The majority of data were retrieved from tables, some from equations, and a small proportion from

digitized figures. Seed yield, aboveground biomass, and plant nutrient uptake are all expressed in dry basis. Units were standardized to megagrams per hectare for seed yield and stover biomass, kilograms per hectare for nutrient uptake, and grams per 100 grams for nutrient concentration.

Data Description

Relevant experimental details of country, design, and year of experimentation were retrieved from the publications. Crops grew under contrasting soil, weather, and management conditions (Table 1.1), causing large variation in all traits under study (Fig. 1.1). Our analysis involved three steps. First, we tested for time trends in each of the traits. Second, the database was divided into three temporal groups: 1921 to 1996 (Era I, $n = 43$), 1997 to 2006 (Era II, $n = 110$), and 2007 to 2016 (Era III, $n = 216$); the increase in data points (means indicated in parenthesis) with time reflects the increased research effort. The criteria to define the eras were: Era I spans from the first published study (Borst and Thatcher, 1931) to the commercial release of the first transgenic soybean in 1996 (Fernandez-Cornejo and McBride, 2002), Era II spans from the first transgenic soybean to >50% of the global soybean crop represented by transgenic soybean in 2006 (Ainsworth et al., 2012), and Era III spans from this point to the present time. Average study year for each era was 1979, 2003, and 2011 (Supplemental Fig. S1.1). Therefore, our era-based evaluation was primarily focused in the 1980s, 2000s, and 2010s. Third, we assessed changes over time of all traits of study. The departure of each trait relative to the mean of each decade was calculated and used to graphically synthesize the main findings.

Descriptive and Statistical Analysis

Traits were plotted against time to quantify trends (Fig. 1.1). For each variable, a least squares linear regression was conducted, and the slope was tested for significance (Motulsky and

Christopoulos, 2003). Histograms were constructed for seed yield and boxplots for N, P, and K uptake (Fig. 1.2).

Zero-intercept linear regression was used to derive nutrient ratios (Fig. 1.3A–1.3C) and to relate seed yield and total N, P, and K uptake (Fig. 1.4A–1.4C). An *F*-test was performed to compare slopes among eras using $p = 0.05$ threshold in all cases. Allometric analyses were performed to quantify the changes, primarily in the slope, of the nutrient internal efficiencies (IEs) and nutrient ratios among eras. To account for error in both x and y , a Model II regression was used (reduced major axis; Niklas, 1994) in Fig. 3A to 3C and 4A to 4C. The SMATR package (Warton et al., 2015) from the R program (R Core Team, 2017) was used to test for common slopes. Envelopes portraying maximum and minimum boundaries (0.99 and 0.01 quantile lines, respectively) for yield-to-nutrient content and nutrient ratios were calculated using the “quantreg” package in R software (Koenker, 2005, 2017). The upper boundary of the envelope represents the maximum nutrient dilution, and the lower boundary represents the maximum nutrient accumulation (Janssen et al., 1990; Witt et al., 1999). Residuals of the adjusted functions were plotted against stover, seed nutrient concentration, and nutrient HI to account for physiologically important sources of variation in these traits. The proportion of variance (R^2) was determined between residuals of the seed yield to plant traits such as plant nutrient uptake, nutrient concentration, and nutrient HI. The same procedure was implemented by Sadras (2006), Ciampitti and Vyn (2013), and Tamagno et al. (2017).

To summarize the changes in yield, nutrient uptake, and partitioning, we used the framework developed by Sadras et al. (2016). First, we compared the rate of change in seed yield and the rate of change in nutrient uptake in relation to the $y = x$ line, which corresponds to IE (Fig. 1.5A). The variation of the trait (seed yield or nutrient uptake) was calculated as a coefficient relative to

the observations within an era. For example, the change in yield for the first era was determined by dividing each observation within an era by the average yield for the last year of the era, and then all observations were averaged and the deviation from a unit was calculated (average minus one). The rationale is that IE would increase if the rate of gain in seed yield is larger than the rate of increase in nutrient uptake (points above $y = x$) whereas the reverse would indicate decreased IE (i.e., points below $y = x$). If the value is different than zero, then the plant trait evaluated for that era presented a change compared with the last year within the era. The slope of the relation of yield to nutrient uptake represents the IE that can be mathematical expressed, as proposed by Sadras (2006), as:

$$IE = \frac{Y}{Nutrient} = \frac{Y}{Nutrient_{seed}} \times \frac{Nutrient_{seed}}{Nutrient_{seed} + Nutrient_{stover}} \quad [1]$$

where Y is grain yield, “Nutrient” is plant nutrient uptake, “Nutrient_{seed}” and “Nutrient_{stover}” are nutrient concentration in seed and stover. The second term of Eq. [1] represents nutrient harvest index:

$$Nutrient\ HI = \frac{Seed\ nutrient}{Seed\ nutrient + Stover\ nutrient} \quad [2]$$

where “Seed nutrient” refers to seed nutrient uptake and “Stover nutrient” to stover nutrient uptake.

Second, to analyze the contribution of nutrient seed concentration and nutrient harvest index to the variation in IE, the rate of change in nutrient seed concentration was plotted against the rate of change in yield per unit nutrient uptake using the $y = -x$ line as reference (Fig. 1.5B). Points aligned around $y = -x$ indicate that a given increase (reduction) in yield per unit nutrient uptake would result in a proportional reduction (increase) in seed nutrient concentration unless nutrient HI changes.

RESULTS

Time Trends for Yield and Nutrient Traits

Seed yield, biomass, and HI showed positive trends across years (Fig. 1.1A–1.1C). The rate of increase in biomass, seed yield, and HI was $0.025 \text{ Mg ha}^{-1} \text{ yr}^{-1}$, $0.058 \text{ Mg ha}^{-1} \text{ yr}^{-1}$, and 0.0008 yr^{-1} , respectively. Seed yield ranged from 1.3 Mg ha^{-1} in the 1930s to 3.2 Mg ha^{-1} in the 2010s, with a minimum of 0.83 Mg ha^{-1} and a maximum of 7.88 Mg ha^{-1} (Fig. 1.1A). Total N, P, and K uptake showed a positive time trend for the pooled data (Fig. 1.1F, 1.1K, and 1.1P). The rate of increase in nutrient uptake was $1.57 \text{ kg N yr}^{-1}$, $0.076 \text{ kg P yr}^{-1}$, and $0.51 \text{ kg K yr}^{-1}$ (Table A.1).

Seed N (N_{seed}) and K (K_{seed}) concentrations were stable across years (Fig. 1.1D and 1.1N). The N_{seed} displayed the smallest variation across years, with a CV of 14% (Supplemental Table A.2). Phosphorous seed concentration declined with time (Fig. 1.1I), with an average rate of $-0.0027 \text{ g } 100 \text{ g}^{-1} \text{ yr}^{-1}$. Stover nutrient concentration remained stable for N (Fig. 1.1E), decreased for P at $-0.0013 \text{ g } 100 \text{ g}^{-1} \text{ yr}^{-1}$ (Fig. 1.1J), and increased for K at $0.009 \text{ g } 100 \text{ g}^{-1} \text{ yr}^{-1}$ (Fig. 1.1O). For all three nutrients, concentration varied more in stover than in seed.

Nitrogen HI increased at a rate of 0.0014 yr^{-1} (Fig. 1.1G), starting from as low as 0.66 in 1930 and ending with an average of 0.72 in 2010. Similarly, HI for P (PHI) increased with time (Fig. 1.1L) at 0.0015 yr^{-1} , from 0.68 to 0.73 between 1930 and 2010. Potassium HI (KHI) declined over time (Fig. 1.1Q) at -0.018 yr^{-1} , from 0.59 to 0.43 between 1930 and 2010. Nutrient internal efficiency increased over time for both N at $0.026 \text{ kg seed kg}^{-1} \text{ N yr}^{-1}$ and P at $0.767 \text{ kg seed kg}^{-1} \text{ P yr}^{-1}$ (Fig. 1.1H and 1.1M); IE for K (KIE) declined at $-0.085 \text{ kg seed kg}^{-1} \text{ K yr}^{-1}$ (Fig. 1.1R).

The largest proportion of the change in IE for N (NIE) was related to improvement in yield, followed by improvements in HI for N (NHI) and N uptake with stable N_{seed} and stover N concentration (N_{stover}) (Fig. 1.1). The change in IE for P (PIE) was related to an increase in yield, followed by improvement in PHI and a decrease of seed P concentration (P_{seed}) (Fig. 1.1I–1.1M). The largest proportion of change in KIE was related to a larger increase in yield relative to total plant K uptake. Potassium HI also decreased with a slight improvement in K_{stover} over time (Fig. 1.1O and 1.1Q).

Trends by Eras

Yield and total uptake of N, P, and K for each era are presented in Fig. 1.2. As documented in Fig. 1.1, average seed yield and nutrient uptake increased across historical eras (Supplemental Table A.2).

Nutrient Stoichiometry

Figures 1.3A to 1.3C show the change in nutrient ratio with era. Nitrogen/phosphorus ratio ranged from 4.9 to 19.0, with greater values for Eras I and III than for Era II. The slope for Eras I and III was larger (11.5) than for Era II (9.0). This is related to the smaller total nutrient content (N, P, and K) for Era II compared with the other eras. The histogram shows higher frequency of greater N/P ratio for Eras I and III than for Era II (Fig. 3A1). Overall, P_{seed} and stover P concentration (P_{stover}) accounted for >50% of the N/P variation (Fig. 1.3A2).

Nitrogen/potassium ratio ranged from 1 to 4 (Fig. 1.3B). Eras II and III clustered and have smaller ratios than Era I. Era I showed frequency of higher N/K ratio, followed by Eras III and II (Fig. 1.3B1). For Era I, N/K averaged 3.1 and decreased to 1.9 for Eras II and III. Seed and stover K concentrations accounted for 5 and 10% of the variation in the residuals of the N/K ratio.

Potassium/phosphorus ratio ranged from 2 to 11 (Fig. 1.3C). Era I showed greater frequency of smaller K/P ratios (Fig. 1.3C1). Analysis of slopes clustered Eras II and III, with smaller ratios for Era I. The P_{stover} explained 20% of the variation in the K/P ratio and P_{seed} only contributed to 5% of the variation (Fig. 1.3C2). Stover P concentration accounted for a larger proportion of the variation in K/P relative to P_{seed} , in agreement with the larger decrease in P_{stover} (36%) relative to P_{seed} (19%) among eras (Table A.2).

Nutrient Internal Efficiency

For both N and P, IE clustered for Eras II and III and was superior to the IE for Era I. For K, IE clustered for Eras II and III and was inferior to Era I. Average NIE increased 33% (Fig. 1.4A) from Era I (9 kg seed kg⁻¹ N) to Eras II and III (12 kg seed kg⁻¹ N); average PIE (Fig. 1.4B) increased by 44% from Era I to Eras II and III (90 to 130 kg seed kg⁻¹ P ha⁻¹); KIE decreased from 27 to 23 kg seed kg⁻¹ K from Era I to Eras II and III (Fig. 1.4C). Variation in seed yield per unit of nutrient uptake was portrayed with two boundaries (dotted lines in Fig. 1.4A–1.4C).

Residuals for the relationship between seed yield and nutrient uptake are presented in Fig. 1.4A1 to 1.4C1. The residuals were regressed against nutrient concentrations in seed and in stover to further dissect the nutrient IEs. The proportion of variance (R^2) explained by each trait is presented in Fig. 1.4A2 to 1.4C2.

Nitrogen HI accounted for 37% of the variation in NIE, whereas N_{seed} accounted for 12% of the NIE variation. Phosphorous HI accounted for 36% of the variation in PIE, and 28% was explained by P_{seed} . Potassium HI accounted for 67% of the variation in KIE, with a smaller contribution from K_{seed} (19%). For all three nutrients, nutrient HI accounted for a large proportion of the variation in the IE, also reflected in the lesser variation relative to NHI. In contrast, N_{stover} and P_{stover} decreased 9.5 and 15.5% for Eras II and III relative to Era I,

respectively, and K_{stover} increased by 37% comparing Era I with the average of Eras II and III (Fig. 1.1E, 1.1J, and 1.1O; Table A.2).

Relative Traits Change

Nitrogen IE improved between Eras I and III, as improvements in yield were unmatched by increased N uptake (Fig. 1.5A). The relative variation in N_{seed} (-1%, *y*-axis, Fig. 1.5B) was small compared with variation in NIE (+30%, slope, Fig. 1.5A) for Eras I and III. Changes in nutrient seed concentration and nutrient HI drive changes in IE (Eq. [1]). Since N_{seed} remained approximately stable (Fig. 1.5B), variations in IE (Fig. 1.5A) were driven by nutrient HI. The increase in NIE was partially a consequence of the increase in NHI (+19%, Fig. 1.1G). Similar to N, PIE increased between Eras I and III, as yield gain was not matched by increase in P uptake (Fig. 1.5A). The increase in PIE (+39%) was more closely related to increase in PHI (+14%, Fig. 1.1L) compared with P_{seed} (-19%, Fig. 1.1I and 1.5B).

For K, the relative improvement in uptake was larger (+35%, Fig. 1.5A) than the increase in K_{seed} (Fig. 1.5B), evidenced as a major distance between triangles in the *x*-axis compared with variations in *y*-axis. Reduction in KIE (-4%) was related to a time trend increase in K_{stover} (+28%, Fig. 1.1O) with a concomitant decrease in KHI (-11%, Fig. 1.1Q).

In summary, improvement in nutrient IE (Fig. 1.5A) was more proportionally explained by changes in nutrient HIs, with more dispersion from the 1:1 line, whereas nutrient seed concentrations remained stable, primarily for N and K (Fig. 1.5B). Therefore, as previously stated, maintenance of nutrient seed concentrations for N and K were obtained by compensatory increases in nutrient HI, with an insufficient change for P. Phosphorus seed concentration declined over time, more proportionally than the increase in PHI (Fig. 1.1I and 1.1L).

DISCUSSION

Yield, Biomass, and Harvest Index

Our database, primarily from small-plot experiments, showed similar yield gain per year to the 25.8 kg ha⁻¹ yr⁻¹ reported by FAO between 1960 and 2014 (FAO, 2017). Historical time trends for soybean yield average increase have been documented in United States (23 kg ha⁻¹ yr⁻¹; Long, 2013; Rowntree et al., 2013; Specht et al., 2014; Wilson et al., 2014), China (11 kg ha⁻¹ yr⁻¹; Wu et al., 2015), Argentina (44.33 kg ha⁻¹ yr⁻¹; de Felipe et al., 2016), and Brazil (41 kg ha⁻¹ yr⁻¹; de Toledo, 1990). Our database indicates that yield improvement was mostly associated with increased biomass and little or no change in HI, in agreement with previous studies (Schapaugh and Wilcox, 1980; Spaeth et al., 1984; Johnson and Major, 1986).

Nutrient Ratios

Nutrient ratios can help to predict nutrient limitations better than individual nutrient concentrations (Güsewell, 2004; Sadras, 2006; Malingreau et al., 2012; Divito and Sadras, 2014; Koerselman and Meuleman, 2017). Nutrient ratios are sensitive to changes in biomass, with ratios decreasing as the proportion of the storage tissues increase (Greenwood et al., 2008; Ciampitti and Vyn, 2014). Nutrient availability and nutrient uptake for immediate use and storage are major sources of variation in nutrient ratio (Bollons and Barraclough, 1999; Ågren, 2008). Liebig's law inadequately assumes a single limiting factor at a given time (Sinclair and Park, 1993), whereas in reality, crop growth is often colimited by multiple factors (Sadras, 2005b; Ågren et al., 2012; Mooney et al., 2012).

In this review, average N/P ratio for modern soybean varieties was 11.5, as compared with the range of 12 to 13 that is typical across terrestrial plants (Güsewell and Koerselman, 2002;

Güsewell, 2004; Knecht and Göransson, 2004). Nitrogen/phosphorous ratio reported was 4.5 for oilseed crops, 5.6 for cereals, and 8.7 for legumes (Sadras, 2006). Nitrogen/phosphorous ratio has been used mainly to assess whether N or P is more limiting for biomass production.

Tamagno et al. (2017) reported a N/P ratio of 11.4 for soybean with experiments in Argentina and United States. In legumes, Sadras (2006) found that differences in P rather than N concentration better explain the N/P ratio variation, since N is more closely regulated by the plant compared with P. Nitrogen concentration in seeds is a conservative trait—small variation (Sinclair and de Wit, 1976; Jin et al., 2011) with a neutral trend over time (Long, 2013).

Variations between N/K and K/P reported in this review were greatly influenced by the increase in K uptake in Era III.

Nutrient Internal Efficiency

In previous decades, scientific research improved the understanding of nutrient uptake and underlying genetics, but less effort was directed towards comprehension of nutrient IE (Santa-María et al., 2015). In agreement with our findings, analysis of modern soybean varieties showed larger changes in IE than in total nutrient uptake (Bender et al., 2015; Gaspar et al., 2017; Yang et al., 2017). Selection for yield increased NIE and PIE but reduced KIE. Soybean seed requires large amounts of N per unit of C relative to other crops (Sinclair and de Wit, 1975), and superior N demand is accompanied by superior P and K requirements (Bender et al., 2015; Gaspar et al., 2017; Yang et al., 2017). Recently, Cafaro La Menza et al. (2017), reporting fertilizer N response in soybeans grown in high-yielding environments ($>6 \text{ Mg ha}^{-1}$), found that indigenous N supply was insufficient to fulfill N requirement with lower N_{seed} when no N was applied. Similar findings were also reported by Tamagno and Ciampitti (2017) but in lower-yield environments

(average 3.5 Mg ha^{-1}), with superior soybean yield to N fertilization explained by longer duration of the seed-filling period, even when biological N fixation was partially inhibited.

The conserved N_{seed} documented in this review is in contrast with other studies reporting negative time trends (Cregan and Yaklich, 1986; Wilcox, 2001; Long, 2013). Nonetheless, our dataset provides a robust explanation, whereby N_{seed} was conserved because both N uptake and NHI increased to fully compensate for the increase in yield.

Phosphorous IE increased over time as a consequence of a reduction in P_{seed} (with reduction in the nutrient partitioning to the seed), and also in P_{stover} , with similar total P uptake across eras. In general, there are two mechanisms to modify PIE within the plant: (i) increasing remobilization from storage, or (ii) reducing the partitioning to developing reproductive organs. For field crops, P_{seed} decreased over time with breeding in association with improvement in HI (Veneklaas et al., 2012).

Variation in KIE can be achieved by (i) substitution of K with other cations, (ii) K translocation between organelles, cells, and organs to regulate K concentration in the cytoplasm, and (iii) K translocation to seed (Gerloff, 1987; Sattelmacher et al., 1994; Rengel and Damon, 2008). In this review, K_{stover} was larger in Eras II and III than in Era I, which can be related to the increase in K fertilization since 1960 (USDA-ERS, 2016). Several authors have reported increases in K_{stover} with K fertilization and without yield response in maize and soybean (Heckman and Kamprath, 1995; Randall et al., 1997; Clover and Mallarino, 2013). Luxury nutrient uptake occurs when uptake is above the crop requirement for a target yield level. As presented by Ciampitti and Vyn (2012), luxury nutrient consumption also occurs if soil nutrient status is high and crop nutrient removal is greater than normal, promoting unbalanced plant nutrient ratios.

Changes in Nutrient Internal Efficiency: Dissecting Critical Components

Changes in nutrient IEs were attained differently for each nutrient. Nitrogen HI increased over time (1921–2016), as reported by others (Jin et al., 2011; Long, 2013), but with N_{stover} remaining stable (Long, 2013). Superior PIE was achieved by a decrease in both P_{seed} and P_{stover} over time. Larger yields were associated with improvement in K uptake, reductions in K_{seed} and KHI, and a substantial increment in K_{stover} , leading to a reduction in KIE over time. Increases in K_{stover} were also reported by others (Heckman and Kamprath, 1995; Randall et al., 1997; Clover and Mallarino, 2013).

Several studies (Vitousek et al., 2009; Peñuelas et al., 2013; Bouwman et al., 2017) reported nutrient imbalances in croplands around the world, but improving nutrient IEs may contribute to reversing this situation.

CONCLUSIONS

Historical analysis of soybean for the period 1921 to 2016 showed that (i) seed yield improved from 1.3 Mg ha⁻¹ in 1930 to 3.2 Mg ha⁻¹ in 2010; (ii) seed yield increase was primarily driven by higher biomass rather than HI; (iii) NHI and PHI both increased over time; (iv) seed nutrient concentration remained stable for N but declined for both P (18%) and K (13%); (v) stover nutrient concentration remained stable for N, declined for P, and increased for K; (vi) nutrient ratios show different trends for N/P (Era I and III > II), N/K (Era I > II and III), and K/P (Era II and III > I); (vii) nutrient IE increased for N (33%) and P (44%) but decreased for K (11%); and (viii) variations in nutrient IEs were primarily explained by changes in nutrient HIs for N and K but were equally explained by both PHI and P_{seed}. A focus on plant nutrient ratios and their relations to crop growth rate is likely to return better tools for nutrient management.

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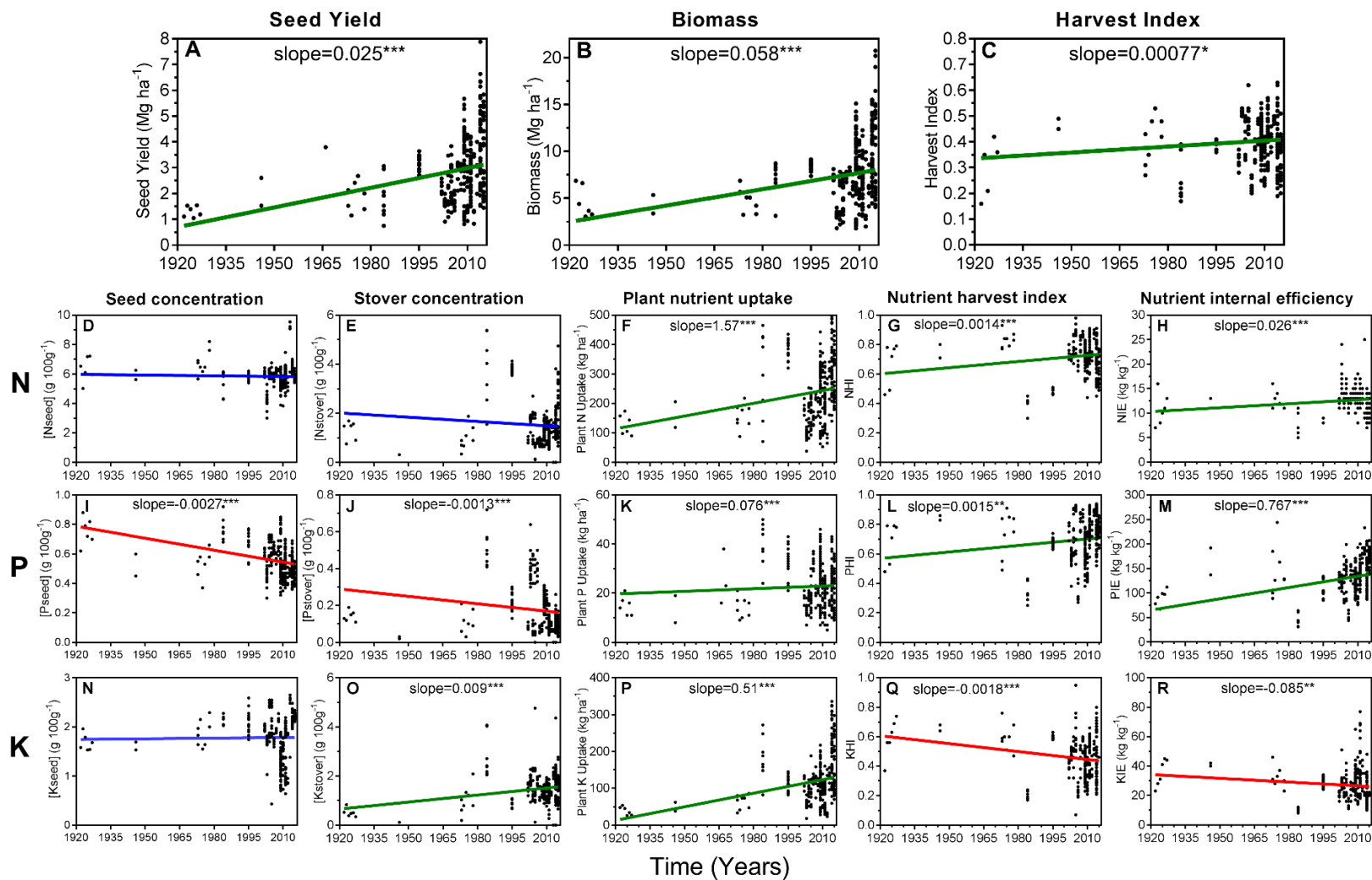


Figure 1.1 Seed yield (A), Biomass (B), Harvest Index (C), N, P, K seed concentration (D, I, N), N, P, K stover concentration (E, J, O); N, P, K plant nutrient uptake (F, K, P); N, P, K nutrient harvest index (G, L, K) and nutrient internal efficiency (H, M, R) among time for the pool data (n=322). Solid line represents linear regression. Green color positive slope, red line negative slope (***) $p < 0.001$ and blue line slope not different from zero. N, Nitrogen; P, phosphorous; K, potassium. NHI, nitrogen harvest index; PHI, phosphorous harvest index; KHI, potassium harvest index; NIE, nitrogen internal efficiency, PIE, phosphorous internal efficiency; KIE, potassium internal efficiency.

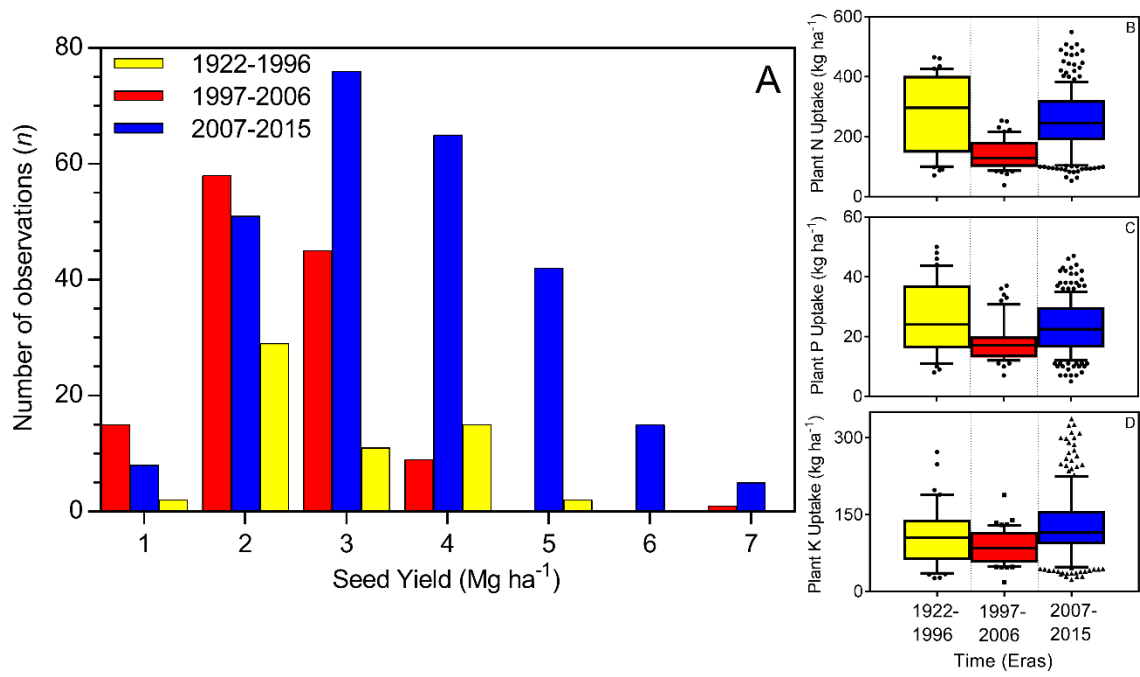


Figure 1.2 (A) Soybean seed yield by era and plant N, P, and K uptake (B, C, and D, respectively) by era.

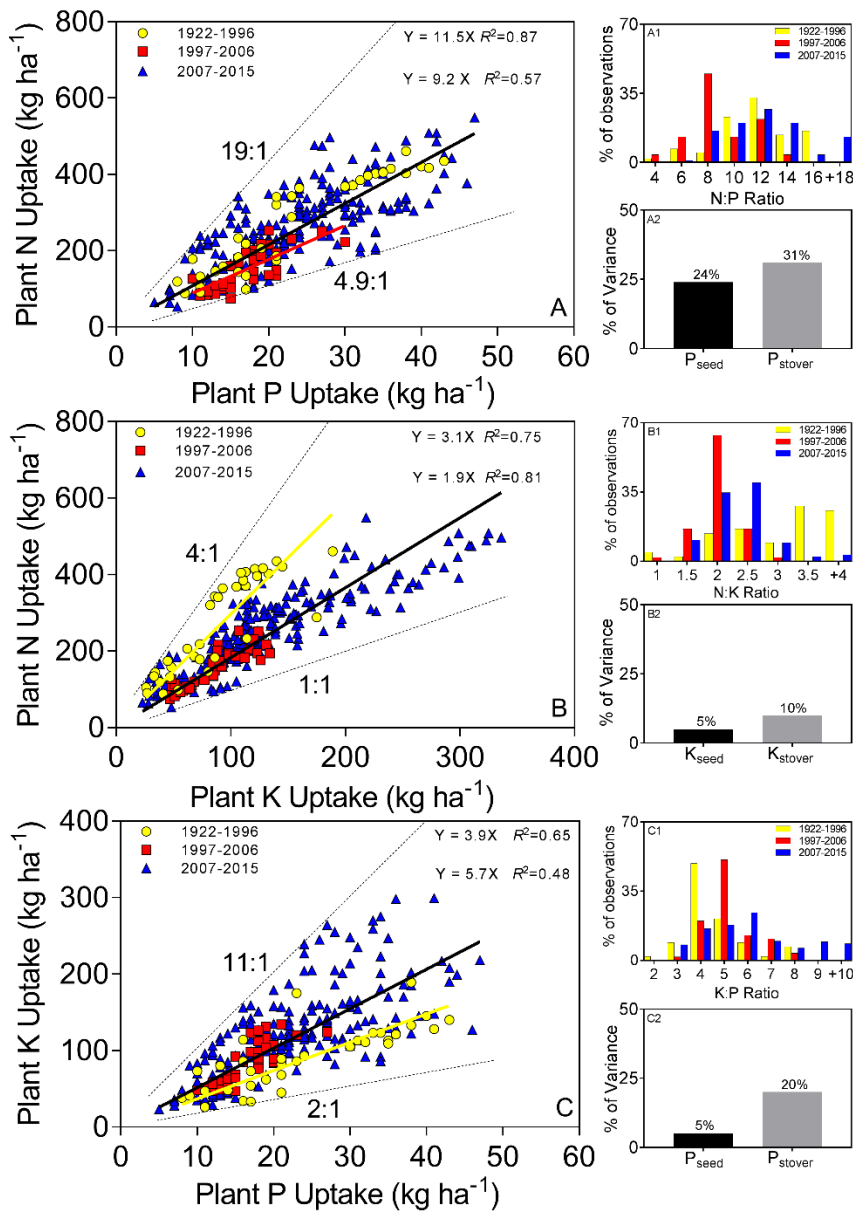


Figure 1.3 (A) Plant N uptake as a function of plant P uptake, (B) plant N uptake as a function of K uptake, and (C) plant K uptake as a function of plant P uptake. N/P, N/K, and P/K ratio by era (A1, B1, and C1, respectively). Percentage of variance (R^2) explained by the linear regression between (A2) residuals of Fig. 3A as a function of seed and stover P concentration, (B2) residuals of Fig. 3B as a function of seed and stover K concentration, and (C2) residuals of Fig. 3C as a function of seed and stover P concentration. Dotted lines indicate boundaries for maximum and minimum ratio for each dataset. Due to lack of significant difference ($p > 0.05$) in slopes, Eras I and II were pooled in Fig. 3A (black line); Eras II and III were pooled in Fig. 3B and 3C (black line). Linear function for pooled dataset: (A) $Y = 11.5 X$ ($R^2 = 0.87$), (B) $Y = 1.9X$ ($R^2 = 0.81$), (C) $Y = 5.7X$ ($R^2 = 0.48$).

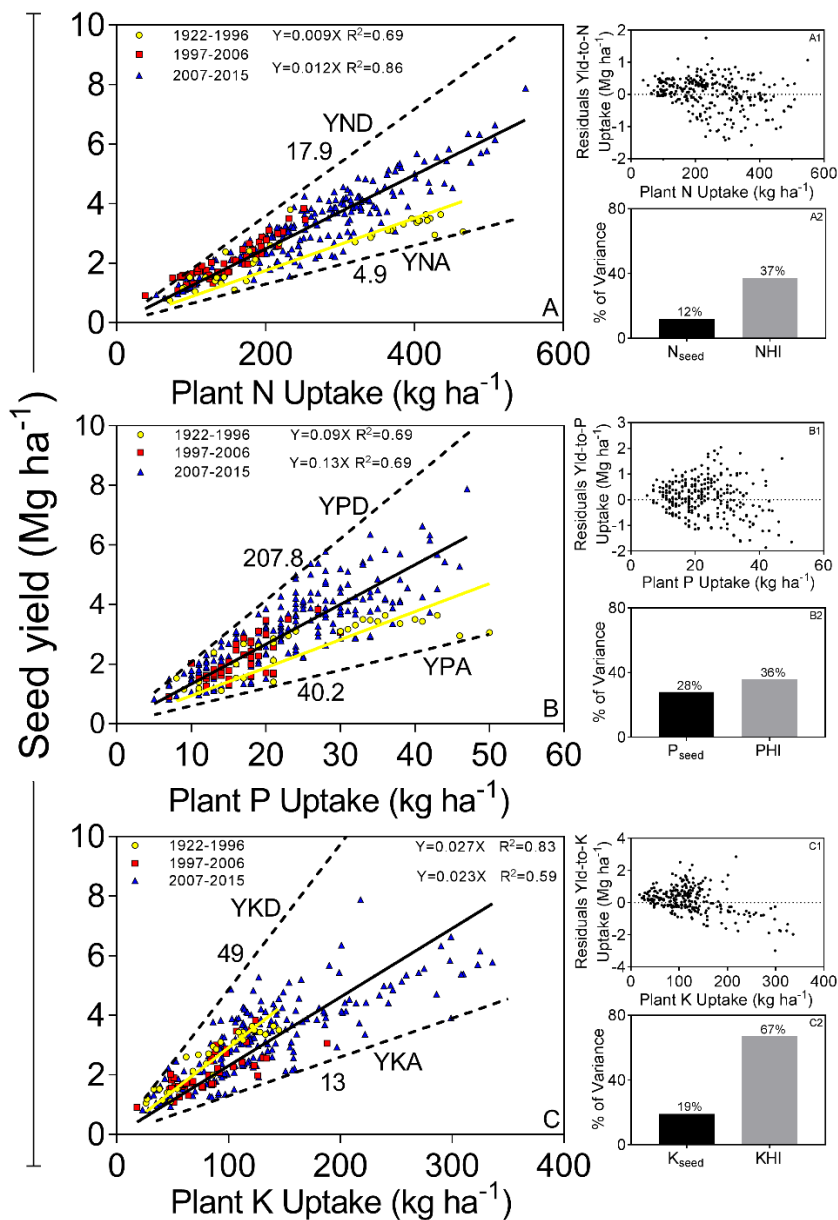


Figure 1.4 Seed yield as a function of (A) plant N uptake, (B) plant P uptake, and (C) plant K uptake. (A1), (B1), and (C1) show residuals for the fitted functions in (A), (B), and (C), respectively. (A2) Proportion of variation (R^2) provided by the linear regression between residuals of Fig. 4A as a function of seed N concentration and N harvest index (NHI); (B2) Residuals of Fig. 4B as a function of seed P concentration and P harvest index (PHI); and (A3) residuals of Fig. 4C as a function of seed K concentration and K harvest index (KHI). YNA, yield N accumulation; YND, yield N dilution; YKA, yield K accumulation; YKD, yield K dilution; YPA, yield P accumulation; YPD, yield P dilution. . Due to lack of significant difference ($p > 0.05$) in slopes, Eras I and II where pooled in Fig. 3A to 3C (black line) Linear function for pooled dataset (A) $Y = 0.012X$ ($R^2 = 0.86$), (B) $Y = 0.13X$ ($R^2 = 0.69$), (C) $Y = 0.023X$ ($R^2 = 0.59$).

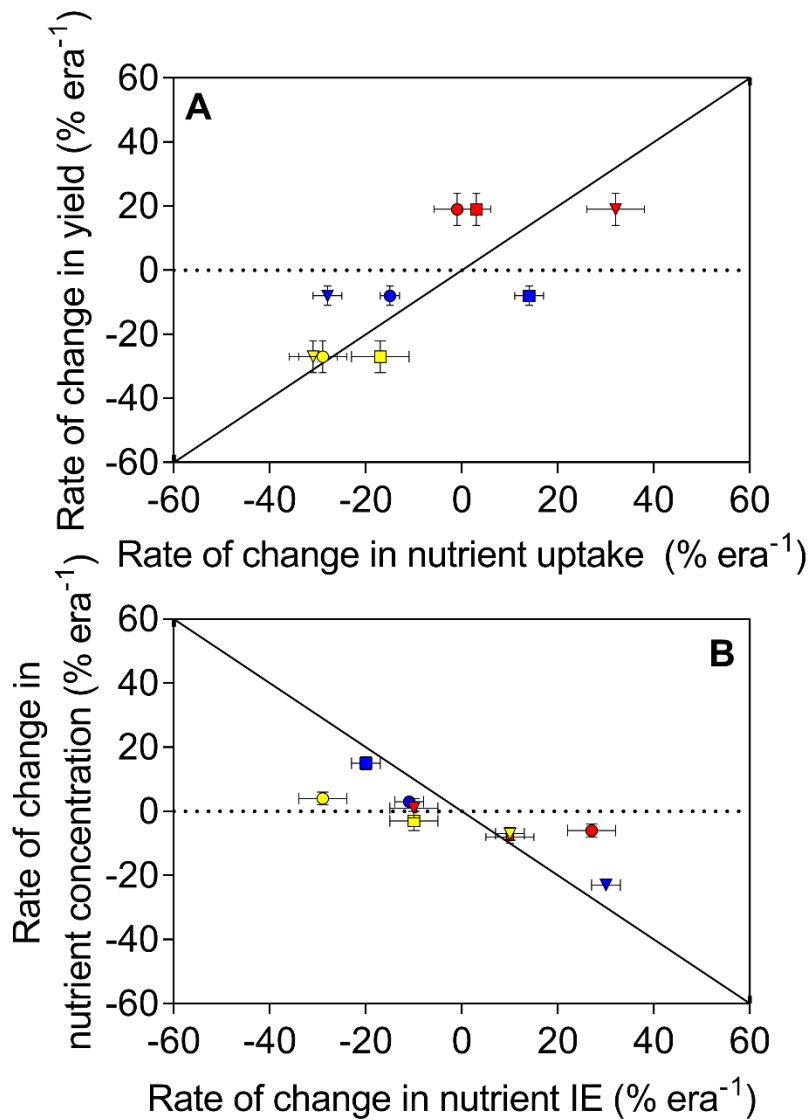


Figure 1.5 (A) Rate of change of soybean seed yield as a function of rate of change of nutrient uptake, (B) rate of change of nutrient concentration. Symbols indicate nutrients: circle = N, square = P, inverted triangle = K. Each point has standard error bars for X and Y variables. Rate of change for a trait was calculated for each era considering the average value for the trait during the last year of the era as a reference. Color indicates era: yellow = Era I, red = Era II, blue = Era III.

Table 1.1 Global soybean database information, including author, year of publication, state or country, year(s) of the study, number of observations collected, and main characteristics evaluated.

Author(s)	Year of publication	State or country	Year(s) of the study	<i>n</i>	Factors evaluated
Borst et al.	1931	Ohio, USA	1922–1927	6	Variety, seeding rate, and planting date.
Hammond et al.	1951	Iowa, USA	1946	2	Soil fertility levels
Bataglia et al.	1976	Goiás, Brazil	1973–1976	1	P, K, and S fertilization
Larcher et al.	1984	Casamace, Senegal	1978–1979	2	Fertilization
Terman	1977	Alabama, USA	1973–1974	2	N and K fertilization
Desoky	1996	Assiut, Egypt	1995–1996	16	Variety, plant density, and N, P, and K fertilization
Vasilas	1984	Minnesota, USA	Not reported	8	N fertilization
Henderson et al.	1970	North Carolina, USA	1966–1968	3	Nutrient uptake
Bataglia et al.	1977	Sao Paulo, Brazil	1974–1976	3	Nutrient uptake
Heard	2006	Manitoba, Canada	2005	1	Nutrient uptake
Rao and Lakshmi	2009	Telangana, India	2003	18	Compost
Rathore et al.	2008	Uttarakhand, India	2006	7	Seaweed fertilizer
Najar et al.	2011	Maharashtra, India	2004	10	Fertilization
Rao et al.	2012	Karnataka, India	2005–2007	2	Fertilization
Chandel	2011	Maharashtra, India	2009	8	Fertilization
Meena and Biswas	2013	New Delhi, India	2010	8	Fertilization
Patel	2011	Madhya Pradesh, India	2009–2010	12	Fertilization
Kurihara et al.	2013	Mato Grosso do Sul, Brazil	2001	1	High yield management
Fageria et al.	2013	Tocantins, Brazil		5	Liming
Bender et al.	2015	Illinois, USA	2012–2013	2	Nutrient uptake
		Santa Fe, Argentina	2009–2013	68	Survey of soybean under rainfed conditions without nutrient limitations across several fields
Tamagno et al.	2017	Kansas, USA	2014	45	Row spacing, inoculation, plant density, and fertilization strategy
		Indiana, USA	2011–2012	54	Variety, biomass, and nutrient uptake and partitioning
Balboa and Ciampitti	Unpublished	Kansas, USA	2015	30	Row spacing, inoculation, plant density, and fertilization strategy

Chapter 2 - Management systems to increase yields on a Maize –

Soybean rotation

ABSTRACT

Different combinations of management practices define distinctive management systems for intensifying crop productivity. Intensification of cropping systems, defined as the increase in productivity per unit of land, is one of the strategies available to achieve the increasing global food demand. The aim of this research was to evaluate the impact, at a farming system scale, of management systems in a maize-soybean rotation, and thereby improve the understanding of the concept of Ecological Intensification. Following this rationale, a maize-soybean rotation was established in Scandia, Kansas, with the goal of evaluating five management systems under contrasting water scenarios (rainfed vs. irrigated). The management systems included different combinations of seeding rate, row spacing, fertilization, inoculation, and pest and disease control. Overall intensified management systems, high input use, based on seeding rate increase, narrow row spacing and a balanced nutrition program increased yields compared to common practices, low input use. For soybeans, utilization of high inputs increased yields relative to the low input use by +1.3 and 2.0 Mg ha⁻¹ for rainfed and irrigated environment, respectively; and for maize, by +1.0 and 2.0 Mg ha⁻¹ for irrigated and rainfed conditions, respectively. In soybean, response to intensified practices with a balanced nutrition program was observed in yield environments above 4.5 Mg ha⁻¹. Soybean yields presented a wider variation throughout all growing seasons relative to maize yields. Each unit of fertilizer applied to soybean produced more yield for high- vs. low-input systems. Grain N concentration in maize varied four-fold (107%), while for soybeans, seed N concentration remained quite stable (20%), with narrower

variation. Further research should be focused on evaluating the impact of management systems to find better strategies to increase productivity in a sustainable manner.

Keywords:

Crop rotation – fertilizer – crop nutrition – crop management – management practices

INTRODUCTION

World population will grow over a third between 2009 and 2050. The growing population is demanding more food and fiber; thus, more efficient and sustainable production systems are needed to face that demand. Reduction of food waste can also greatly contribute to feeding the growing population. There are two options to increase overall production: i) to increase productivity per unit of land or ii) to incorporate new land in production. The option to increase productivity per unit of land can be achieved by increasing crop yields or by increasing crop intensity, number of crops per year (Matson et al. 1997; Cassman 1999; Caviglia et al. 2004; Sadras and Roget 2004; Lobell et al. 2008; Fischer et al. 2014; FAO 2017).

The potential yield of a crop is defined by temperature, radiation, genetic material under conditions of no water and nutrient limitation. The difference between the potential yield and the actual yield (on-farm yield) is called the yield gap (Evans and Fischer 1999; Van Ittersum et al. 2013). Selecting the best crop and nutrient management practices (e.g., genotype selection, row spacing, planting date, and nutrient 4Rs—right fertilizer source, rate, time, and place), and considering their interactions with each other and with the environment (soil plus weather), will directly impact the size of the yield gap. Identifying the best combination of management practices applied to a specific production situation is herein referred to as the selection of the best management systems (BMSs).

Crop intensification can be defined as the yield improvement per unit of land area and time (Cassman 1999; Gregory et al. 2002; Sadras and Roget 2004). Increasing productivity (more yield per unit of area) and/or increasing crop intensity (more crops per year) are two strategies for meeting the increasing global demand for food. Achieving the mentioned goal of intensifying cropping systems in a sustainable manner requires an integrated approach for evaluating the BMSs for each environment. In recent years, several studies have evaluated the effect of

different management practices individually without considering the interaction with other practices or with crops in the rotation.

For maize crop, modern hybrids (Duvick and Cassman 1999; Duvick 2005), increasing plant population (Duvick 2005), higher rates of fertilizers (Randall et al. 1997; Duvick and Cassman 1999; Dobermann and Cassman 2002), and row spacing (Farnham 2001; Lambert and Lowenberg-DeBoer 2003), among other management practices are commonly reported in the literature. For soybean crop, selection of the best varieties, seeding rate (Board and Maricherla 2008), row spacing (Holshouser and Whittaker 2002; Holshouser et al. 2006; Hanna et al. 2008), fertilization (Gutiérrez-Boem et al. 2004; Salvagiotti et al. 2008; Wang et al. 2008), fungicide application (Hanna et al. 2008; Swoboda and Pedersen 2009; Gaspar et al. 2015), and inoculation (Bergersen et al. 1989; Suri and Choudhary 2013; Zimmer et al. 2016; Moretti et al. 2017), are among the best production practices recommended for improving yields.

Scientific reports presenting information on the impact of management systems at rotation- or cropping system-level are scarce. Sustainable intensification practices has been reported to increase soil organic carbon (SOC) levels with improvement in soil fertility, biological activity and water holding capacity (Seybold et al. 1998). Agricultural practices will impact the ability of ecosystems to provide good services. Intensification of fertilizer application, for example can increase nutrient concentration in water reservoirs with an environmental consequence (Tilman et al. 2002) .

There is need for research from a global perspective to develop a framework to identify cropping areas where intensification will produce a positive impact in production while maintaining sustainability, as opposed to other fragile areas that should be managed under less pressure. Sustainable agriculture must assure sufficient, secure, stable and equitable supply of

food and services for the growing population (Lynam and Herdt 1989; Tilman et al. 2002).

Selection of the BMSs at a cropping system level can contribute to this goal. Thus, the aim of this research was to evaluate the impact, at a farming system scale, of management systems in a maize-soybean rotation, and thereby improve the understanding of cropping systems based on the concept of Ecological Intensification (Cassman, 1999).

MATERIALS AND METHODS

A maize – soybean rotation was established in 2014 at the Northcentral Kansas Experiment Field near Scandia, Kansas, US (39°49'54"N; 97°50'21"W), on a Crete silt loam soil (fine, montmorillonitic, mesic Panchic Argiustoll). The climate at the site is classified as warm-humid continental (Peel et al., 2007), with a mean annual temperature of 11.8 °C and precipitation of 713 mm. About 75% of the rain occurs during the growing season (April-October). The rotation was established under both rainfed and irrigated conditions. Both phases of the rotation were present each year. Five treatments were evaluated: common practices, CP (low seeding rate, wide-row spacing, no seed inoculation, and without P, K, S application); comprehensive fertilization, CF (low seeding rate, wide-row spacing, inoculation, and P, K, S application); production intensification, PI (high seeding rate, narrow-row spacing, inoculation, and without P, K, S application); ecological intensification, EI (high seeding rate, narrow-row spacing, seed inoculation, balanced nutrition N, P, K, S application, fungicides and insecticide application); and advanced plus, AP (high seeding rate, narrow-row spacing, seed inoculation, balanced nutrition N, P, K, S application, double application of fungicides and insecticides). The details of management practices are presented on Table 2.1. For treatments (CF, EI and AP) receiving fertilizer application, nutrients were broadcasted at planting using two granular sources, 12-40-0-10S-1Zn (MicroEssentials® SZTM) - Mosaic Co.) and 0-0-58-0.5B (Aspire® - Mosaic Co.). The criteria to decide P, K and S fertilizer rate was replacement by providing all P, K, and S according to grain removal driven by yield target. The criterion for N fertilization for maize was to follow a balanced approach, calculated as the difference of N provided by soil N (soil test) and crop N demand according to a yield target. Rates under irrigation and rainfed conditions were different since maximum attainable yield for the irrigated environment was higher. Field experiments under irrigation during 2014-2017 growing season received between 160 to 190 mm

of water. Plot size was 15 m length and 6 m width replicated 5 times in a complete randomized block design (CRBD). In-season measurements were conducted in three out of five replications. Phenological data was recorded for maize (Ritchie et al. 1986) and soybean (Fehr et al. 1971). All fertilizer rates applied in maize and soybean are presented in Table 2.1. Maize plots in 2015 were placed in the same location as the soybean plots for the 2014 season with the goal of documenting residual effects from the management tested. In-season measurements were conducted in three out of five replications. Grain samples were ground to determine N concentration via combustion method (AOAC 2000). Yield moisture was adjusted to 13 g kg⁻¹ for soybean and 15.5 g kg⁻¹ for maize.

The partial factor productivity of the fertilizer (PFPf) was calculated as the ratio between grain yield and amount of fertilizer applied (N, P, K, S) per unit of area (Cassman et al. 1996) for maize and soybean under rainfed and irrigated water condition.

When studying how to intensify a cropping system, interactions between crops and environment need to be taken into account to better understand the complex interactions taking place in the system. To evaluate the interaction between crop species and environment maize to soybean yield ratios were plotted against soybean yields following the approach proposed by Egli (2018).

Statistical analysis

Yields and PFPf were analyzed by ANOVA and means were separated by LSD Fisher ($p=0.05$). Descriptive statistics (mean, min, max, standard deviation, and coefficient of variation) were performed for the maize to soybean ratio and to characterize N concentration in soybean seed and maize grains.

RESULTS

Soybean and maize yields

Management systems implemented in the maize and soybean rotation impacted yields in both crops for the 2014-2017 period. For the maize phase, yield differences were found between treatments for both water scenarios. Across seasons (2014-2017), average yield under rainfed conditions was 11.7 Mg ha⁻¹, while under irrigation average yield increased to 12.9 Mg ha⁻¹ (Fig. 2.1). A balanced nutrition program (2, CF) or intensification with a balanced nutrition program (4, EI; 5, AP) resulted in the maximum yield with 13.5 Mg ha⁻¹. The common practices treatment (1, CP) and intensification without a balanced nutrition program (3, PI) yielded the minimum with an average of 12 Mg ha⁻¹. No differences were found between treatments for 2014 and 2017 growing seasons for both phases rainfed and irrigated and no differences on the rainfed phase for 2015. Under irrigation, for 2015 and 2016 growing seasons, maximum yields were above 14 Mg ha⁻¹ and statistical differences were found between treatments with a balance nutrition program applied. In those scenarios treatment 1 (CP) and 3 (PI) yielded significantly less (-2.1 and 3.1 Mg ha⁻¹ for 2015 and 2016) that the average of treatments with a balance nutrition program applied (Fig. 2.1).

Among seasons (2014-2017), average soybean yield under rainfed was 3.7 Mg ha⁻¹, while under irrigation average yield was 4.6 Mg ha⁻¹ (Fig. 2.2). Treatment PI (3), EI (4) and AP (5) produced more yield (4.2 Mg ha⁻¹) compared to CP (1) and CF (2), which averaged 3.9 Mg ha⁻¹. On the irrigated side, greater yield differences were found. Maximum yield was attained by treatment 4 and 5 (EI, AP) with 5.5 Mg ha⁻¹ average across seasons, followed by treatment 3 (PI) with 4.8 Mg ha⁻¹. In all rainfed environments, addition of a balance nutrition program to a common practice system or intensification with addition of a balanced nutrition program did not

increased yields. Soil nutrients in those environments provided the requirements for the 3.7 Mg ha⁻¹ yield achieved on average. When taking a look to the irrigated scenarios in seasons 2016 and 2017 a balanced nutrition program was producing more yield over common practices and production intensification. The 4.7 Mg ha⁻¹ of yield achieved under irrigation required a balanced nutrition program to increase yields. Treatment 4 and 4 where the ones showing the highest yields in all environments, while treatment 1 was always achieving the lowest yield. In high yielding environments, a balanced nutrition program enhanced productivity.

Intensification benefit

To account for the impact of a balanced nutrition program, treatment PI (3) and EI (4) were compared under rainfed and irrigated scenarios for both maize and soybean (Fig. 2.3). In soybean, the first year in both rotations did not show yield differences between treatments with an average of 2.5 and 5.2 Mg ha⁻¹ under rainfed and irrigated conditions, respectively. On the same plots maize was planted in 2015 and soybean came back to the same plots by 2016. Under rainfed conditions, 2015 soybean showed an increase in yield for treatment 4 (EI), but without presenting any statistical difference related to treatment 3 (PI). Under irrigated conditions, treatment 4 (EI) yielded 0.8 Mg ha⁻¹ more than treatment 3 (PI). Soybean planted in 2015 and 2017 seasons under irrigation shows that there was a difference of 0.8 Mg ha⁻¹ between treatments consistently in both years. In 2015, 2016 and 2017 growing seasons, statistically significant differences between treatments 3 (PI) and 4 (EI) were observed with an increase in yields of 0.8 Mg ha⁻¹ for treatment 4 compared with treatment 3. For the level of yield reported, a balance nutrition program consistently increased yield in most of the scenarios under irrigated

conditions for soybean. These results support the need of a balance nutrition program when intensifying production systems.

As was showed in Fig. 2.1 the experiment was located in an environment with suitable soil conditions for maize production. Average yields under rainfed conditions were above 10.5 Mg ha⁻¹ in all seasons, while under irrigation, yields ranged from 11.8 to 13.6 Mg ha⁻¹. Treatment 4 (EI) produced more yield in all seasons (Fig. 2.3), but yield differences between treatment 3 and 4 were only statistically significant in 2016 for both water scenarios.

Rotation benefit

The experimental design and the rotation setup where treatments were placed in the same location the following season, but switching crops enabled evaluation of residual effects of treatments in the following crop of the rotation. This approach aims to start looking at the results with a cropping system point of view. The average yield of soybean for 2014-2015 and for maize 2015-2016 by treatment were plotted in Fig. 2.4. Analyzing treatments as management systems, no yield response to balance nutrition program was identified for soybeans since treatment 2 (CF) was not statistically different from treatment 1 (CP); and with intensification treatment 3 (PI), 4 (EI) and 5 (AP) not differing in yield. Taking a look at maize yields coming after the soybean crop, results shows that a balanced nutrition program applied to treatments 2, 4, and 5 yielded more than treatments 1 and 3, which lacked balanced nutrition. Soybeans did not response to a balanced nutrition program, but the lack of application of nutrients might be affecting crop production in the following season, particularly for maize.

Partial factor productivity of the fertilizer (PFPf)

The effect of a balanced nutrition program was already analyzed in terms of the impact on production. Now, focusing on treatments where nutrients were added, the efficiency of those nutrients to produce yield can be quantified. The partial factor productivity of the fertilizer (PFPf) indicates how many units of grain/seed are produced per each unit of fertilizer applied to the system. For soybeans, intensification increased PFPf by 11 and 15% under rainfed and irrigated environments, respectively (Table 2.2). In the case of maize, intensification with a balanced nutrition program did increase yields, and since treatment 4 (EI) received an additional 56 kg N ha⁻¹, the PFPf was lower compared with the treatment 2 (CF). Common practices plus a balanced nutrition program (CF) produced 35% more yield per unit of fertilizer applied compared with the intensification, EI, (narrow rows, higher seeding rate, and a balanced nutrition).

Maize and soybean yield ratio stability

From a cropping system point of view, a better understanding of crop species interactions with the environment is needed. Maize and soybean yield ratio can be implemented to characterize this interaction (Fig. 2.5). The relation between the maize-soybean ratio and soybean yields shows that soybean explored a wider range of yields compared with maize. In conditions when soybean yields are above 3.5 Mg ha⁻¹, maize yield remained more stable, while soybean yields increase in response to environment and treatment characteristics. Average ratio under rainfed conditions was 3.5, while for irrigated conditions the ratio decreased to 2.9. Under rainfed conditions soybean yields varied 35% compared with a 17% variation on maize yields. Soybean yields increased 26% in response to irrigation while yield increase in maize was 15%

both crops compared to the rainfed environment (Table 2.3). As previously mentioned, maize yields were above 10 Mg ha⁻¹ in all cases, while soybean yields were less stable and more responsive to the management systems evaluated.

Maize and soybean N removal: implications for N balance

When intensifying production systems, special attention is required to the nutrient status of the seeds/grains produced with the goal of quantifying nutrient removal from the field. Soybean yields ranged on average from 2.9 to 5.5 Mg ha⁻¹ in response to the management systems evaluated. In the case of maize, the range of yield was narrowed going from 10.5 to 13.6 Mg ha⁻¹. Mean N seed concentration for soybean was 5.85 g kg⁻¹ with a minimum of 5.32 and a maximum of 6.38 g kg⁻¹, and 5.71 – 5.95 g 100 g⁻¹ for the 25% and 75% percentile respectively. For maize, mean grain N concentration was 1.26 g kg⁻¹, ranging from 0.74 to 1.53 g kg⁻¹ with 1.17 -1.40 g 100 g⁻¹ for the 25% and 75% percentile respectively. (Table 2.4). The analysis of seed N concentration for both crops showed that soybean seed N concentration was four times less variable compared with maize. This points out the ability of soybean to maintain stable seed concentrations through a wide variety of yields.

DISCUSSION

In this research, a maize-soybean rotation was evaluated for two complete cycles of the rotation (2014-2017) looking to the performance of five management systems under rainfed and irrigated conditions. This integrated approach provides a better understanding of the complex interactions that takes place in cropping systems. Several authors have demonstrated the effect of intensification on closing yield gaps for field crops (Cassman 1999; Wood et al. 2000; Mueller et al. 2012; Fischer et al. 2014). For major crops, climate, fertilizer application and irrigation can explain 60 to 80% of yield variability (Mueller et al. 2012). However, concern over the long-term sustainability and environmental consequences of intensification of agricultural systems has developed, especially in fragile environments (Matson et al. 1997; Gregory et al. 2002; Cassman et al. 2003).

Foy soybeans, intensification based on narrowing row spacing, increasing seeding rate, and pest and disease control increased seed yield, with response to a balanced nutrition program observed in environments yielding more than 4.5 Mg ha⁻¹. Soybean has an extraordinary ability to compensate with seeding rate from 80 to 900 thousand seeds per ha⁻¹ (Coulter et al., 2011; Grichar, 2007). In this research, narrowing row spacing from 76 to 38 cm impacted yields positively as was previously reported by other authors (Ethredge et al. 1989; Lambert and Lowenberg-DeBoer 2003; Kratochvil et al. 2004; Conley et al. 2008; De Bruin and Pedersen 2008; Hanna et al. 2008; Chen and Wiatrak 2011b; Cox and Cherney 2011).

For maize, intensification without a balanced nutrition program negatively impacted yields, reflecting the sensitivity of this crop to lack of N and imbalanced nutrition. Increasing seeding rate consistently impacted yields (Jones et al. 1977; Andrade et al. 2002; Barbieri et al. 2012) but narrowing row spacing presented mixed results, with positive (Jones et al. 1977; Andrade et al. 2002; Johnson and Hoverstad 2002; Lambert and Lowenberg-DeBoer 2003; Shapiro and

Wortmann 2006; Fawcett et al. 2015), neutral (Farnham 2001) or negative effects (Maddonni and Martínez-Bercovich 2014).

Beside the impact of management systems for each particular crop rotation, important improvements in productivity can be achieved with innovations at the cropping or farming system levels (Rodriguez and Sadras 2011). Increasing cropping system diversity can allow to produce the same or more yield than traditional rotations with reductions in agricultural inputs (Davis et al. 2012).

The 4R (right source, right rate, right time, and right place) nutrient stewardship approach provides a synthesis to proper management of fertilizers in a cropping systems with consideration for economic, social and environmental benefits (Roberts 2007). Even when no responses were observed to nutrient application, we can assume a negative nutrient balance for the soil that will lead to decrease soil fertility. Fertilizer efficiency in cropping systems can be a metric to compare management systems. The PFPf showed that, in addition to yields increase, intensification for soybeans produced more seed yield per unit of fertilizer applied. The PFPf can be utilized to measure the long-term sustainability of agricultural systems. Sustainable systems are characterized by a non-negative trend in the PFPf over a period of time (Cassman et al. 1996, 2003; Hobbs and Morris 2011).

Analyzing the ratio of yields of two crops growing at the same location for a certain number of growing seasons allow the identification of interactions between crops and the environment (Egli 2018). In the preset research, management systems positively impacted soybean yields, generating a wider yield variation compared to maize yields. A historical analysis of maize/soybean ratios in US showed an increase after 1950 based on the higher rate of

improvement in maize relatively to soybean yields, and a stabilization between 2.8 and 3.2 in 1980 and thereafter (Randall et al. 1997; Specht et al. 1999; Egli 2008).

The mentioned increase in maize and soybean yield in the last fifty years has lead us to think about the impact on grain nutrient concentration. Actual crop simulation models require accuracy to estimate nutrient content for crops. An understanding of nutrient concentration variation is needed to develop relatively simple methods to predict with a higher level of accuracy the nutrient balance in our farming systems. In this research N seed content was characterized for maize and soybean with a database that included 4 years of data on a varying range of yields (n=120 observations per crop). For N concentration, soybean presented a four-fold variation relative to maize. A review in maize (1940-1980) reported an optimum grain N concentration of 1.58 g 100 g⁻¹ with a variability of 132% (range 0.90-2.09) (Jones 1983). For soybean, a historical review from 1922 to 2015 reported an average of 5.84 g 100 g⁻¹ with value of 5.43 and 6.28 g 100 g⁻¹ for the 25% and 75% percentile (Balboa et al. 2018) while a study with experiments from 2009 to 2014 reported a mean of 5.92 g 100 g⁻¹ with values of 5.45 -6.34 g 100 g⁻¹ for the 25% and 75% percentile (Tamagno et al. 2017) . The greater variability observed in soybean can be accounted for by the wide range of environments and genetic materials that were included in those publications.

Increasing grain production and maintaining quality is one of the most critical challenges of agriculture. Intensification of cropping systems will not be possible in all environments. Inclusion of best management systems (BMSs), including new crops to the rotation, increasing the number of crops per year (harvesting more radiation), adopting new technologies to account for the within field variability (precision agriculture) are some of the options that will enable

increased food production with sustainability (Lynam and Herdt 1989; Gregory et al. 2002; Caviglia et al. 2004; Sadras and Roget 2004; Rodriguez and Sadras 2011).

CONCLUSIONS

Overall, intensified management systems based on seeding rate increase, narrow row spacing and a balanced nutrition program increased yields compared to common practices. For soybeans, utilization of high inputs increased yields relative to the low input use by $+1.3 - 2.0 \text{ Mg ha}^{-1}$ for rainfed and irrigated environment, respectively; and for maize, by $+1.0 - 2.0 \text{ Mg ha}^{-1}$ for irrigated and rainfed conditions, respectively. In soybean, response to intensified practices with a balance nutrition program was observed in yield environments above 4.5 Mg ha^{-1} . Soybean yields presented a wider variation throughout all growing seasons relative to maize yields. Each unit of fertilizer applied to soybean produced more yield for high- vs. low-input systems. Grain N concentration in maize varied four-fold (107%), while for soybeans, seed N concentration remained quite stable (20%), with narrow variation. Further research should be focused on evaluating the impact of management systems to find better strategies to increase productivity in a sustainable manner.

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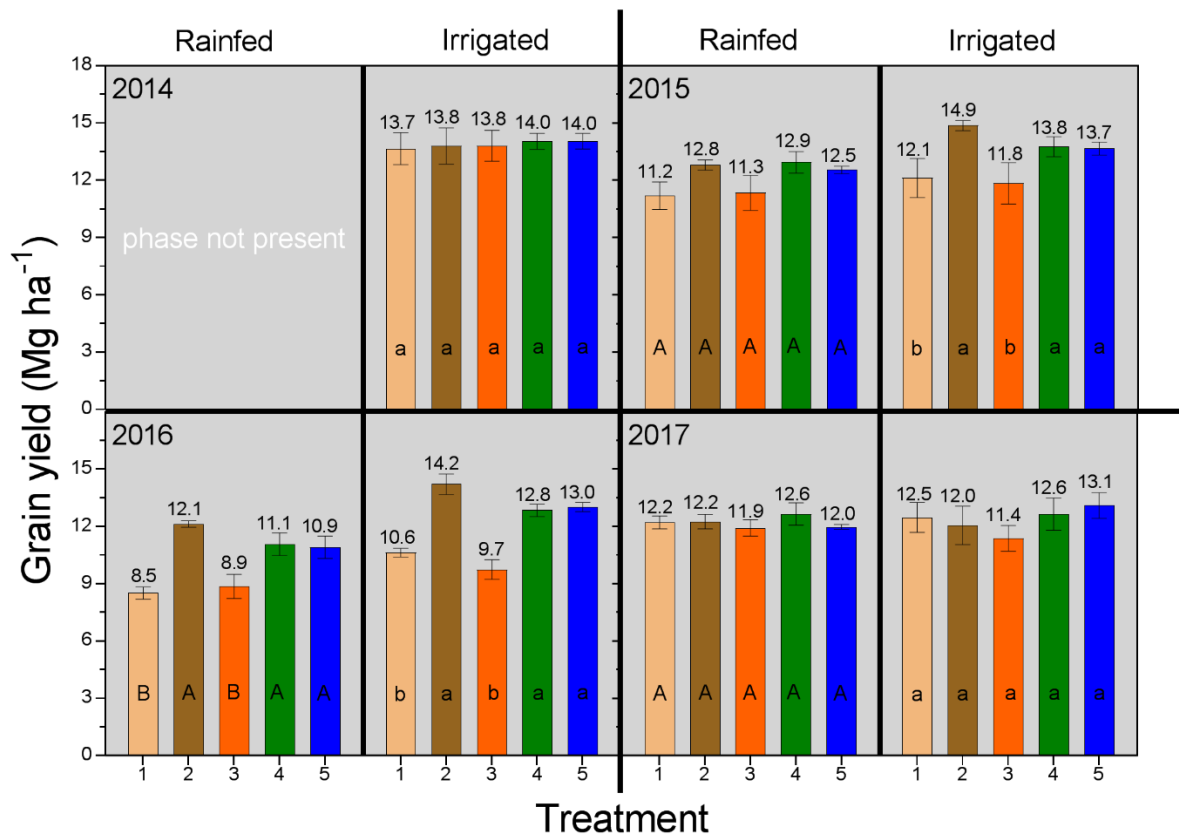


Figure 2.1 Maize yield under rainfed and irrigated scenarios for 2014, 2015, 2016, and 2017 growing season, Scandia, KS. Average for 2014-2017 growing seasons. Different letters within water scenario and year indicate statistical differences between treatments ($p < 0.05$). Treatments names: 1, CP common practices; 2 CF comprehensive fertilization; 3, PI production intensification; 4, EI ecological intensification; 5 AP advance plus.

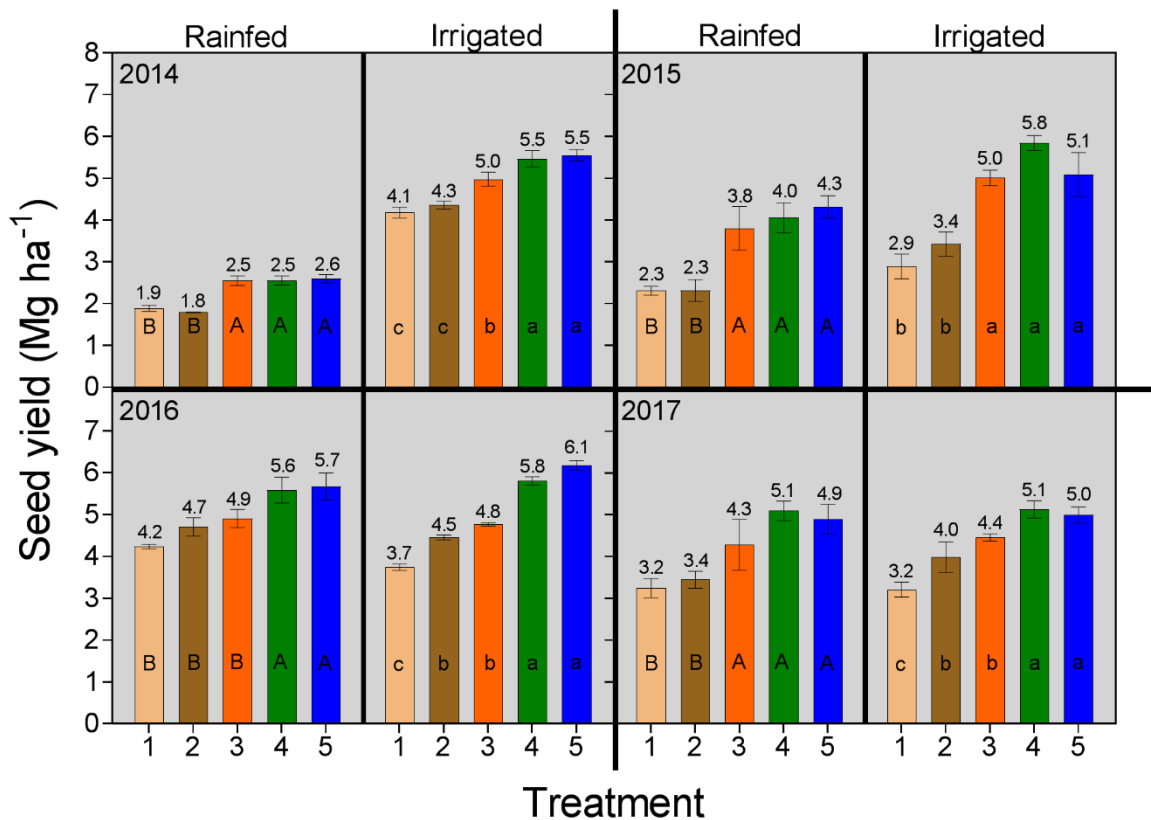


Figure 2.2 Soybean yield under rainfed and irrigated scenarios for 2014, 2015, 2016 and 2017 growing season, Scandia, KS. Different letters within water scenario and years indicate statistical differences between treatments ($p < 0.05$). Treatments names: 1, CP common practices; 2 CF comprehensive fertilization; 3, PI production intensification; 4, EI ecological intensification; 5 AP advance plus.

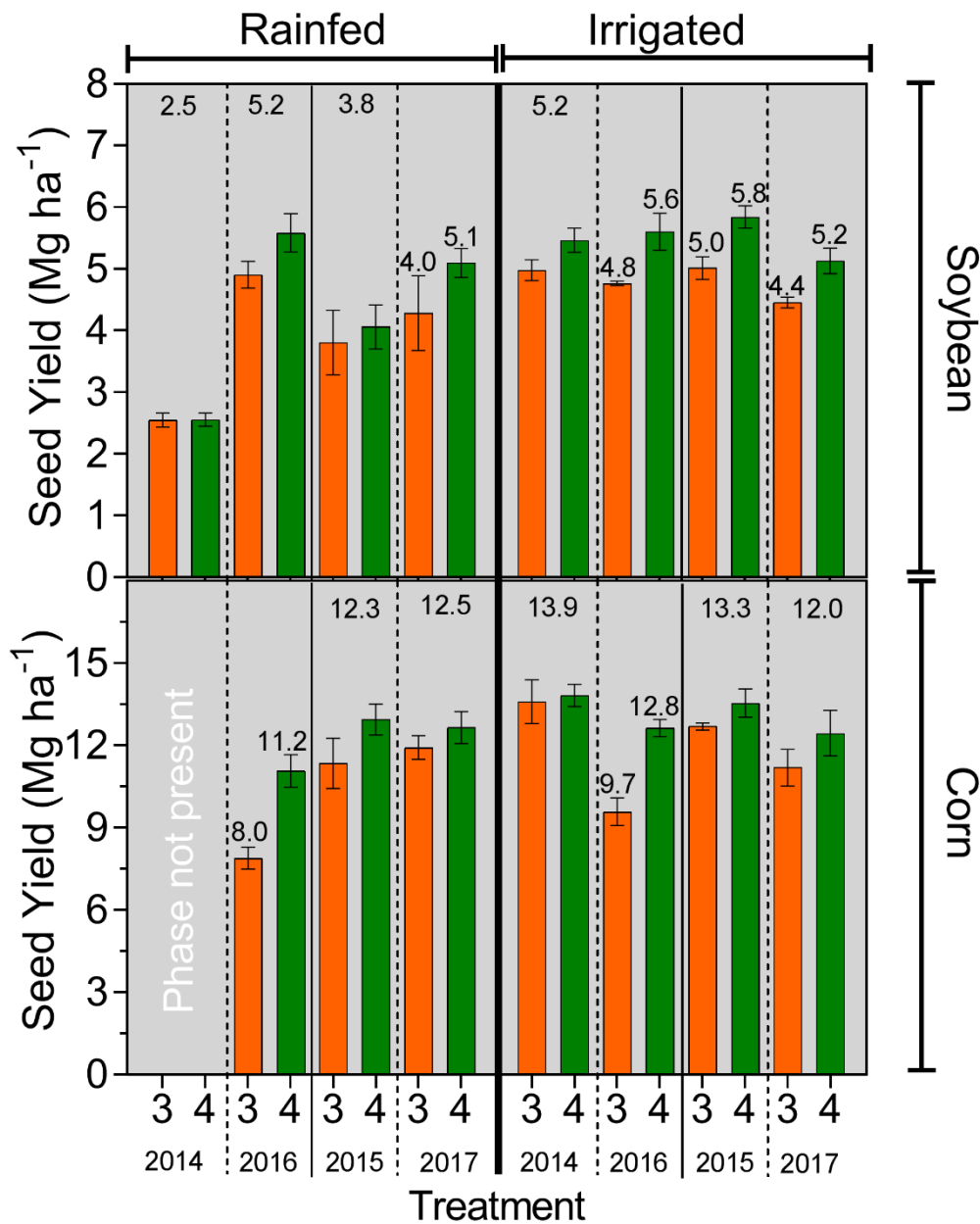


Figure 2.3 Soybean and maize yield under rainfed and irrigated conditions for treatments 3 PI (production intensification), and 4 EI (ecological intensification) for growing seasons 2014, 2015, 2016, and 2017. Single yield value within a crop-year means no statistical difference on yields for treatments 3 PI (production intensification) and 4 EI (ecological intensification), two values indicates mean for each treatment when statistical difference were found ($p < 0.05$).

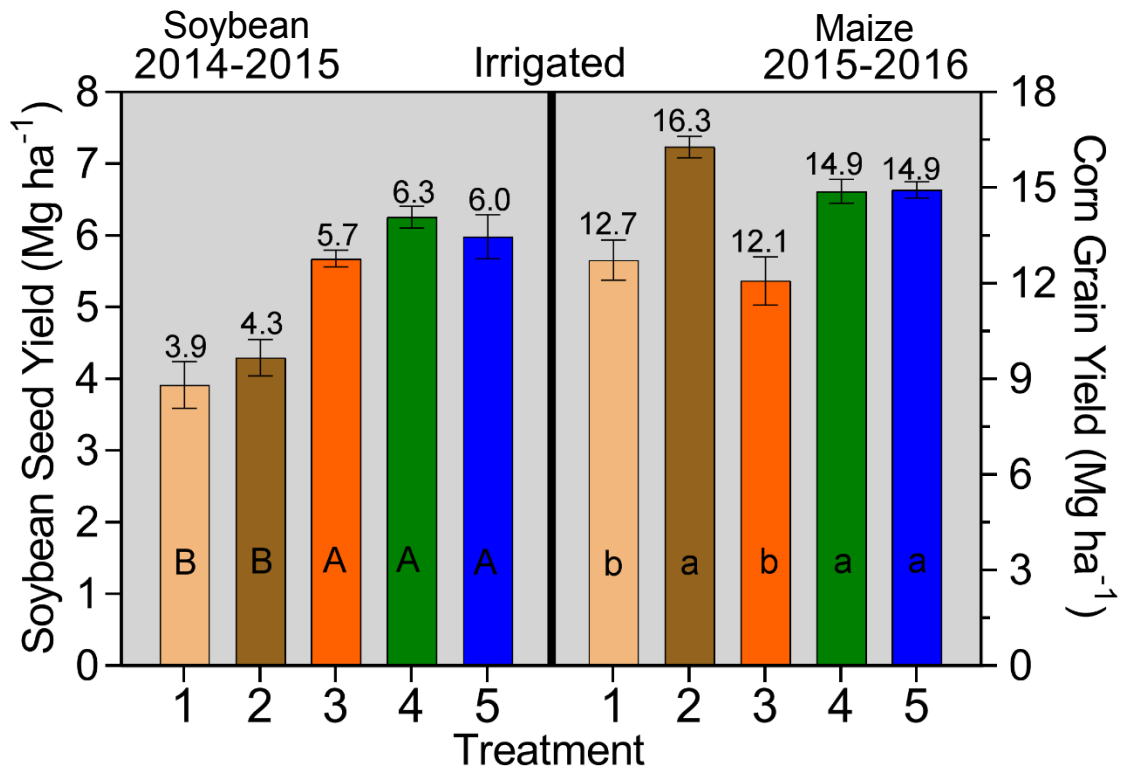


Figure 2.4 Soybean yield for 2014-2015 growing season (averaged) and maize yield for 2015-2016 growing season (averaged), both crops under irrigation. Different letters within crop indicate statistical differences between treatments ($p < 0.05$). Treatments names: 1, CP common practices; 2 CF comprehensive fertilization; 3, PI production intensification; 4, EI ecological intensification; 5 AP advance plus.

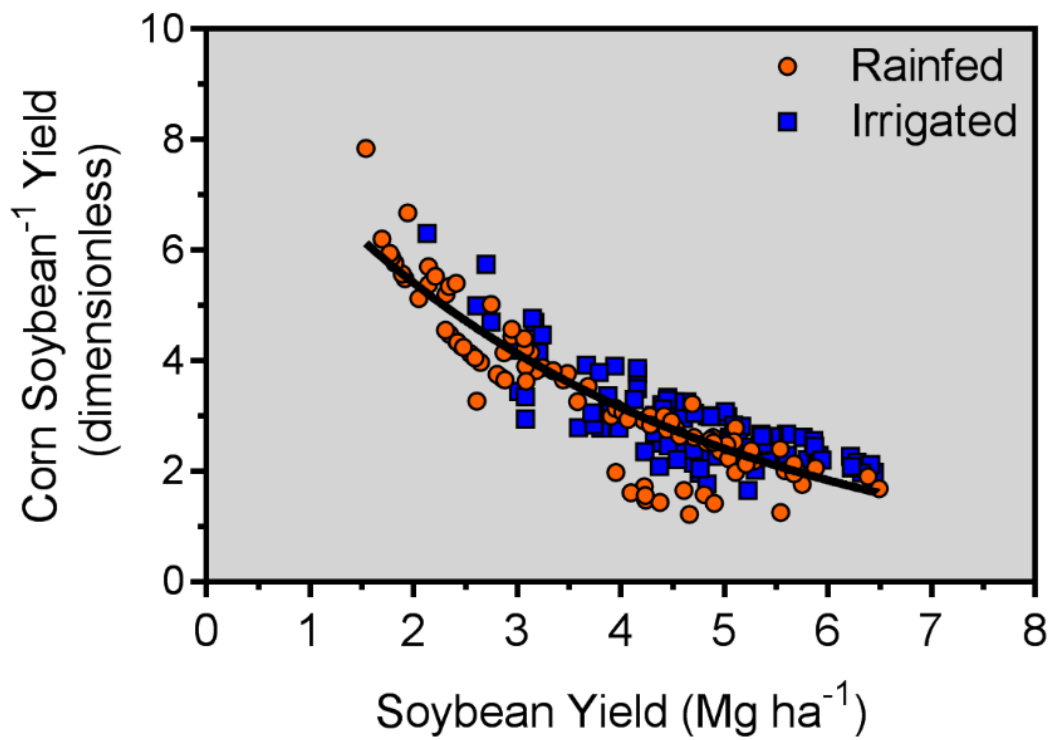


Figure 2.5 Maize and soybean yield ratio against soybean yield. Orange circles represent rainfed scenario blue squares represents irrigated scenario data points. All treatments were pulled together.

Table 2.1 Treatment description for soybean experiment at Scandia, Kansas

Treatments	CP	CF	PI	EI	AP
Seeding rate S/M (Seeds m ⁻²)	27/7.5	27/7.5	43/9	43/9	43/9
Row spacing (m)	0.76	0.76	0.38	0.38	0.38
Fertilization	No	(P-K-S)	No	(N*-P-K-S)	(N*P-K-S)
Micronutrients	No	No	No	1x (Fe, Zn, B)*	2x (Fe, Zn, B)**
Fungicide/Insecticide	No	No	No	1x**	2x**

CP=common practices, CF= comprehensive fertilization, PI= production intensity, EI= ecological intensification (CF+PI), AD= advanced plus.

*Applied at R3. **Applied at R1 and R3. S, soybean; M, maize.

Fertilizer rates in kg N-P₂O₅-K₂O-S ha⁻¹: (63-10-35-9) and (63-15-48-12) for rainfed and irrigated. Treatment CF did not receive any N application.

Table 2.2 Partial factor productivity of the fertilizer for maize and soybean under rainfed and irrigated conditions, Scandia, KS. (2015-2017).

<i>Treatment</i>	<i>Soybean</i>		<i>Maize</i>	
	Rainfed	Irrigated	Rainfed	Irrigated
2 (CF)	16.7 b	14.3 b	65.4 a	56.6 b
4 (EI)	18.6 a	16.5 a	51.6 b	45.2 a
<i>p value</i>	0.04	0.01	0.006	0.005

CF, comprehensive fertilization; EI, ecological intensification. Different letter within column indicates statistical differences (P<0.05).

Table 2.3 Yield descriptive statistics (mean, minimum, maximum, standard deviation and coefficient of variation) for maize, soybean, and maize-soybean ratio under rainfed and irrigated scenario.

	Maize		Soybean		Maize/Soybean	
	Rainfed	Irrigated	Rainfed	Irrigated	Rainfed	Irrigated
Average	11.23	12.86	3.71	4.67	3.71	4.67
Min	5.68	8.57	1.54	2.13	1.54	2.13
Max	15.12	16.09	6.50	6.45	6.50	6.45
SD	1.88	1.77	1.30	0.95	1.30	0.95
CV	17%	14%	35%	20%	35%	20%

Min, minimum; Max, maximum; SD, standard deviation; CV, coefficient of variation.

Table 2.4 Descriptive statistics for maize and soybean seed N content

	Soybean	Maize
Mean	5.85	1.26
25% percentile	5.71	1.17
75% percentile	5.95	1.4
Min	5.32	0.74
Max	6.38	1.53
CV	3.71	14.2
Variation	20%	107%
n	120	120

Min, minimum; max, maximum; CV, coefficient of variation; n, number of observations

Chapter 3 - Soybean B value characterization and the impact on biological nitrogen fixation estimation

ABSTRACT

Soybean plants are able to fix nitrogen from the atmosphere. On average that fixation was estimated to be 60%. Quantification of biological nitrogen fixation (BNF) becomes critical to develop a nitrogen balance for the crop. The ^{15}N Natural Abundance (NA) technique can estimate BNF in soybean. The technique requires the estimation of the relative ^{15}N concentration from a N fixing soybean plant growing in a N free environment, the so-called B value. The aim of this study was to determine the B value throughout the growing cycle and plant fractions for four soybean (*Glycine max* L.) varieties, and to quantify the impact of the B value on the BNF process with the natural abundance technique for two contrasting management systems (low- vs. high-inputs). The BNF was assessed by the ^{15}N natural abundance technique. A greenhouse study was conducted to determine the B value by plant fraction and by phenological stage for four soybean varieties. In a field setting, two contrasting management systems were conducted and BNF was estimated with the B value obtained from the greenhouse study. The BNF contribution among stages was calculated with three different B values: the mode from the literature, the B value measured from the greenhouse for each sampling time, and the B value at the peak of the biomass accumulation measured in greenhouse. Nitrogen fixation rate was estimated for both treatments (low- vs. high-inputs) under rainfed and irrigated conditions. Yield and total N uptake statistically differed for both treatments under analysis, with EI out yielding CP by 0.7 and 1.2 Mg ha⁻¹ for rainfed and irrigated conditions, respectively. The B value measured in the greenhouse study varied among phenological stages, plant fractions, and

soybean varieties. Average B value at the peak of biomass accumulation was -1.97 (R₇), while the mode value reported in the scientific literature was -1.70. Intensification significantly increased N rate fixation from 1.4 to 2.9 and 3.7 to 6.1 kg N ha⁻¹ from the low- to the high-input treatments under rainfed and irrigated conditions, respectively. The peak of N fixation was at phenological stages R1-R3. The B value determinations will impact BNF estimation; therefore, measuring B value for each variety and rhizobia strain will help to reduce errors in BNF estimations. Fixation percentage ranged from 45 to 56% of the total N demand. For the level of BNF measured, the source of B value had a relatively low impact on BNF estimation.

Keywords: fixation rate, Natural abundance, intensification, fixation estimation, greenhouse.

INTRODUCTION

It was estimated that soybean fixes close to 25% of total N around the world (Herridge et al. 2008). On average, biological N fixation (BNF) in soybean (*Glycine max* L.) provides 50-60% of the crop demand, and in most of the cases (80%), N balance in production systems is negative (Salvagiotti et al. 2008). Increasing BNF in soybean can be achieved by selection and engineering of *bradyrhizobia* and soybean genotypes, improving inoculation techniques, matching the best fitted genotypes to the right environment, and management of other inputs (Keyser and Li 1992). However, accurate estimation of BNF and identification of factors impacting this process are needed to guide further crop improvement. Soybean has two sources of N available for growth, mineral N from soil or fertilizer sources, and fixed atmospheric N₂. It is not a simple matter of accurately quantifying the proportion and amount of N fixed by field-grown soybeans. To estimate BNF in legumes such as soybean several methods have been summarized by Unkovich et al. (2008). The natural abundance (NA) technique (Shearer and Kohl 1986) is one of the most accurate and inexpensive methods at the field-scale, providing an overall estimate of the contribution of BNF up to the time of sampling (Unkovich et al. 2008). Atmospheric N₂ and plant-available soil N differ in isotopic composition (Amarger et al. 1979), and the amount of N coming from fixation (%Ndfa) can be estimated using a two-source mixing model (Unkovich et al. 1994a). The ¹⁵N NA of atmospheric N is by definition 0.00‰ when plants are growing entirely relying on BNF for N supply, the resulting δ¹⁵N value -denominated the B value- of the plants is usually negative. The B value is defined as the difference between the ¹⁵N NA of the legume plant grown entirely on BNF and that of the air (Okito et al. 2004). There are references in the literature comparing ¹⁵N NA with other methods such as ureide or ¹⁵N dilution (Herridge et al. 1990; Oberson et al. 2007).

The NA technique requires the determination of the isotopic ^{15}N of three fractions: i) the legume of interest, ii) a non- N_2 -fixing reference plant growing within the same field as the mentioned legume, and iii) the isotopic ^{15}N fraction of soybean growing in a N free media (relying only on BNF) to produce the so-called B value. The magnitude of the ^{15}N abundance value for each component involved in the technique will impact BNF estimation errors. Significant errors could be experienced in the estimates of %Ndfa if the ^{15}N abundance of reference plant is close to zero and/or when the rhizobial strain differs then changing the B value. When %Ndfa is low, ^{15}N abundance from the reference plant have more impact on the accuracy of the determination; on the other hand, changes in the B value will impact when plant reliance on N_2 fixation is high (Boddey et al. 2000; Pauferro et al. 2010).

Selection of reference plant is critical for accuracy of isotope-based methods (Danso et al. 1993, Pate et al. 1994). In agricultural ecosystems, relatively minor changes in reference plants for ^{15}N NA were reported in the literature (Unkovich et al. 1994a; Boddey et al. 2000; Okito et al. 2004). Examples of reference plants include weeds (Schwenke et al. 1998, Schweiger et al. 2014), non- N_2 -fixing mono- or dicotyledonous crops (McNeill et al. 1998; Reiter et al. 2002; Collino et al. 2015), and no- N_2 -fixing mutants of the legume (Okito et al. 2004). The assumption is that the reference plant will have access to the same available N pool as the soybean plant.

The B value is determined in a greenhouse experiment by measuring the ^{15}N abundance of fixing plants growing in an N free substrate (Shearer and Kohl 1986; Boddey et al. 2000). The B value has been reported to vary between legume species (Unkovich et al. 1994b, 2008), varieties within the same species (Danso et al. 1987; Boddey et al. 2000; Nebiyu et al. 2014; Zimmer et al. 2016), rhizobium strains (Danso et al. 1987; Guimarães et al. 2008; Pauferro et al. 2010; Zimmer et al. 2016), and phenological stages (Kyei-Boahen et al. 2002; López-Bellido et al. 2010). Few

studies have focused on BNF estimation by NA technique through different growth stages in soybean (Oberson et al. 2007; López-Bellido et al. 2010; Schweiger et al. 2014) but without looking to genotype effect. Therefore, the objectives of this research were to: 1) determine the B value among cycle and plant fractions for four soybean varieties, 2) quantify the impact of the B value on BNF estimation for two contrasting soybean management systems (low- vs. high-input) with the NA technique.

MATERIALS AND METHODS

Natural abundance technique

With the purpose of quantifying the percentage of N coming from BNF in two contrasting management systems (low- vs. high-input) in soybean, the ^{15}N NA technique was implemented to estimate the %Ndfa by accounting for the natural variation in ^{15}N . The percentage of N coming from the air was calculated by the following equation (Shearer and Kohl 1986):

$$\%Ndfa = \frac{\Delta^{15}\text{N reference plant} - \Delta^{15}\text{N legume}}{\Delta^{15}\text{N reference plant} - B}$$

Where %Ndfa is the percentage of N coming from the atmosphere through BNF; reference plant refers to a no- N_2 fixing plant to be placed with the fixing soybean to account for the soil mineral N uptake; the factor B refers to the $\Delta^{15}\text{N}$ NA of the N derived from the air and it is expressed in parts per thousand relative to the atmospheric N_2 . In this research, the B value was calculated for four varieties and five sampling times in a greenhouse study. In biological materials, the B value range for $\Delta^{15}\text{N}$ range from -5 to +10 ‰. Reference plant was non-nodulating soybean isolate (William 82). Soybean was selected as reference plant to ensure the closest possible match in temporal and spatial root development and activity patterns between the N_2 -fixing and non- N_2 -fixing plants (Herridge et al. 1990).

Three approaches were followed to assess the impact of different B values on BNF estimations. The first one was to use the mode B value summarized from the scientific literature for soybeans (-1.75 ‰); the second one was to use the greenhouse measured B value by plant fraction and by stage for the soybean variety planted in the field; and the last approach was to use the B value for the maximum biomass point (-1.93 ‰) measured in the greenhouse.

B value review

With the objective of characterizing the B values reported in the literature, an assessment of scientific publications in peer-reviewed scientific journals was conducted. Papers were retrieved from CABI, Web of Science Core Collection, Scopus, Springer Link, Agricola, and Google Scholar using the keywords: “soybean,” “B value,” ^{15}N natural abundance. Papers that measured the soybean shoot b value in a greenhouse were included.

B value determination in greenhouse

The B value was estimated in a greenhouse experiment (Amarger et al. 1979; Bergersen and Turner 1983) designed to evaluate four soybean varieties. Soybean genotypes were 93B82 (non-RR), P39T67R (RR1), 93Y92 (RR1) and P34R43R2 (RR2). Varieties were selected representing the ones used in field experiments to estimate BNF. Soybean plants were grown in a 3.8 l plastic pot with sterilized sand in complete absence of mineral N with six replications. Seeds were sterilized in 70% (v v⁻¹) ethanol for 3 minutes and 3% (v v⁻¹) bleach solution for 2 minutes, followed by 3 minutes rinse in deionized water (Schipanski et al. 2010). Seeds were inoculated with a multi-strain *Bradyrhizobium japonicum* (Vault® HP from BASF, a minimum count of 1×10^{10} CFU mL⁻¹) at a rate of 1.3 mL kg seed⁻¹. Six seeds were sown per pot and thinned to 4 plants per pot at phenological stage V₁ (Fehr et al. 1971). Plants were regularly irrigated with tap water and with N-free Hoagland’s nutrient solution. Aboveground biomass was collected at V₄, R₁, R₃, R₅, and R₇ phenological stages (Fehr et al. 1971). Samples were partitioned into stem, leaves, and pods (when present); and dried in an oven at 65 °C until constant weight. Dry weight was recorded, samples were ground, and analyzed for $\Delta^{15}\text{N}$ and N concentration in a mass spectrophotometer.

Field Experiments

Two soybean experiments were established at the Northcentral Kansas Experiment Field near Scandia, Kansas, US (39°49'54"N; 97°50'21"W) with the purpose of quantifying the %Ndfa of two contrasting soybean management systems. Soil type was a Crete silt loam soil (fine, montmorillonitic, mesic Panchic Argiustoll), with soybeans planted during 2015 growing season. One experiment was under irrigated conditions and the other under rainfed. Two treatments were evaluated: low input or common practices, CP [low seeding rate (274 thousands seeds per ha), wide-row spacing (0.76 m), and no P, K, S application]; and high input or ecological intensification, EI [high seeding rate (429 thousand seeds per ha), narrow-row spacing (0.38 m), balanced nutrition (N, P, K, S application), fungicides and insecticide application]. For the Ecological Intensification treatment receiving fertilizer application, nutrients were broadcasted at planting using two granular sources-40-0-10S-1Zn (MicroEssentials[®] SZ^(TM) - Mosaic Co.) and 0-0-58-0.5B (Aspire[®] - Mosaic Co.). The criteria to decide P, K and S fertilizer rate was replacement by providing all P, K, and S according to seed nutrient removal driven by yield target approach. Rates under irrigation and rainfed conditions were different since maximum attainable yield for the irrigated environment was greater. Plot size was 15 m length, and 6 m width replicated five times in a complete randomized block design (CRBD). In-season measurements were conducted in three replications out of five. Yield moisture was adjusted to 13 g kg⁻¹. Aboveground biomass and nutrient concentration were expressed on dry matter basis. The total amount of irrigation during 2015 growing season was 190 mm, equally split in five irrigation times.

Statistical analysis

The B value from soybeans varieties and % of BNF in field experiments were compared with analysis of variance (R Core Team 2016). Separation of means was evaluated by implementing Fisher LSD test with a significance level of 0.05.

RESULTS

Soybean B value in the literature

A complete list of the values retrieved from the scientific literature can be observed in Table 3. 1. Average B value reported was -1.69 with a mode of -1.70, a minimum of -3.86 and a maximum of 0.20. All the values reported refer to aboveground biomass at peak accumulation. Most of the studies reviewed do not measure the B value for the variety involved in the field experiments. Instead, they use previously measured B values from the literature to estimate BNF. The reviewed references did not have in-season estimations of BNF, and only one B value was applied for all cases.

B value determination

To accurately determine seasonal BNF under field conditions and contrasting management, a B value was measured in a greenhouse experiment. The greenhouse study was conducted to characterize the B value per phenological stage for each plant fraction for four soybean varieties, which included one of the soybean varieties planted under field conditions. The B value tends to become more positive throughout the growing season, while for the leaf fraction the opposite is true. For the first sampling time (V4), a large variability between varieties was observed (Fig. 3.1).

Statistical analysis for the B value showed differences between varieties, phenological stages, and plant fractions. No interaction was detected between these factors ($p > 0.05$) (Table 2). Average across sampling times and plant fractions, the range of B value between varieties was -2.19 to -1.96 (Table 3. 3). Within-plant fractions, leaves and stems showed a similar B value (-2.12 and -2.18, respectively) but statistically differing from pods (-1.78) plant fraction.

Statistical analysis of biomass accumulation showed differences between varieties, plant fractions and stages. There were no significant differences in N content across soybean varieties in the greenhouse study ($p>0.05$). Average N concentration was 2.9 mg kg^{-1} (Table 3. 3).

BNF estimation at the field scale

The field experiment evaluated the performance of two contrasting management systems, common practices (CP, low input) and ecological intensification (EI, high input) under both rainfed and irrigated conditions. Seed yield was significantly affected by treatments under both water scenarios. Under rainfed conditions, EI resulted in 2.5 Mg ha^{-1} while CP yielded 1.8 Mg ha^{-1} . Under irrigation, EI yielded 5.4 Mg ha^{-1} , 29% more than CP (4.2 Mg ha^{-1}) (Fig. 3.S1).

Biomass accumulation, plant N content, and percent of N coming from fixation by plant fraction are presented in Table 3. 4. The BNF values, calculated with the B value measured from the greenhouse study, were estimated for each sampling time and plant fraction for a field-grown soybean.

For BNF trait, there were statistical differences between environments (rainfed-irrigated) and phenological stage. For the 2015 growing conditions, the selection of the B value did not produce statistical differences in BNF estimations. At phenological stage R₅, a larger proportion of the N content came from BNF (70%). Under irrigated conditions, the EI treatment presented more N coming from fixation relative to CP. Under rainfed conditions, the proportion coming from N fixation averaged 51% and was not different between treatments (Fig. 3.3).

Total N content coming from BNF was 130 vs. 190 kg ha^{-1} for CP and EI for the rainfed environment, respectively; while under irrigation, total N fixed at R₇ stage went from 124 and 285 kg ha^{-1} from CP and EI treatments.

Minor differences were found within treatment and weather scenarios depending on the B value implemented in the estimation. For CP, 56, 51 and 53% total N fixed for the B value from literature, per growing stage, and at the peak of the biomass growth under rainfed conditions; similar trend for the same treatment was recorded for total N fixed from 49, 46 and 46% but under irrigation. For EI, N fixation values averaged across the season were 58, 54 and 54% for the rainfed, and 50, 46 and 45% under irrigation.

N Fixation Rate

The ^{15}N natural abundance technique allowed determination of N fixation at five phenological stages in soybean. By combining this data with aboveground plant N content and biomass, a N fixation rate curve was estimated throughout the entire crop season for both treatments under rainfed and irrigated scenarios (Fig. 3.4). At the early stage (V_6), N fixation rate was similar for all treatments and water scenarios and ranged from 0.09 to 0.19 kg N ha⁻¹ d⁻¹. Maximum N fixation rate for the CP treatment was 1.4 and 2.9 kg N ha⁻¹ d⁻¹ for rainfed and irrigated scenarios, respectively. The peak for CP was at the R_1 growth stage and remained approximately stable until the R_5 stage. For EI, maximum N fixation rate was 3.7 and 6.1 kg N ha⁻¹ d⁻¹ for rainfed and irrigated scenarios (R_3 , R_1), respectively. In most cases, the N fixation rate remained high and stable between R_1 and R_5 stages. After the R_5 growth stage, all treatments and water scenarios showed a declining phase until R_7 stage, with a final N fixation rate ranging from 0.15 to 1.19 kg N ha⁻¹ d⁻¹(Fig. 3.4).

DISCUSSION

The present research provided a new insight into N fixation estimation utilizing the ^{15}N NA technique. The impact of B value determination on BNF estimation and its variation as related to variety, plant fraction, and phenological stage was evaluated in this study. Ecological Intensification (EI) significantly increased yields, N uptake and its rate compared with the CP treatment. Seasonal N fixation rate followed a similar pattern as that documented by Patterson and LaRue (1983), rapidly increasing after R1, peaking at R3, and decaying at R5 growth stage. Overall, BNF contributed from 11 to 87% of total N uptake, while Salvagiotti et al. (2008) reported an average value of 50-60% with a range from 0 to 97%. Changes observed in this study in the N isotopic composition of plant parts during the development of nodulated soybeans followed the previously described pattern of decreasing ^{15}N of soybean shoots (Shearer et al. 1980, Bergersen et al. 1988).

Implementation of the NA technique to estimate N fixation was reported on many years ago by Amarger et al. (1979) and Kohl et al. (1980). Reviews on the ^{15}N natural abundance technique have also been published (Shearer and Kohl 1986; Boddey et al. 2000). Several authors consider the ^{15}N NA a feasible technique to be applied in field studies (Oberson et al. 2007; Unkovich et al. 2008; Pauferro et al. 2010), subject to a lower error arising from spatial and temporal variations in the ^{15}N abundance of the N available to plants.

Overall N fixation estimation utilizing the proposed B values ranged from 45 to 56%, which is a relatively narrow variation and relatively moderate to low percentage of N fixation. Unkovich et al. (2008) demonstrated that with a range of B value from -1.5 to -2.1 ‰ small impacts (4-6% units) can be observed in BNF when overall fixation is low (<50%). When $\Delta^{15}\text{N}$ of N_2 fixing legumes is close or below zero, %Ndfa is high, and the impact of the B value on BNF estimation is higher. At low %Ndfa, the B value becomes relatively less important, but with

high N fixation, the impact of B value becomes more critical to BNF estimation (Unkovich et al. 2008).

Typically, biological materials range from ^{15}N value of -5 to +10‰; these values can be easily measured. The ^{15}N distribution between non – nodular tissue is reasonably uniform, variations between plant parts are generally within about 2‰ of each other, except for nodules that are substantially enriched (Kohl et al. 1980; Bergersen and Turner 1983; Shearer and Kohl 1986). The B value for shoots usually ranges from 0 to -2‰, but when the whole plant value is calculated it is close to zero since the roots are slightly positive and nodules are strongly positive (Shearer et al. 1984; Boddey et al. 2000; Okito et al. 2004). Soil N is usually more abundant in ^{15}N than atmospheric N_2 (Shearer and Kohl 1986). Non- N_2 fixing plants would also be expected to be more abundant in ^{15}N than atmospheric N_2 .

In the present research, B value was determined in the greenhouse. Okito et al. (2004) followed the method implemented by Doughton et al. (1992) to determine the B value at the field scale. The technique is based on the comparison of soils artificially enriched with ^{15}N with those derived from the use of the NA technique. They suggested that this B value is more appropriate to estimate BNF since it involves growth in field conditions.

The B value should be determined at the same growth stage that the legume and reference plant are sampled in the field including the specific rhizobia strain (Unkovich et al. 1994c, 2008)

A non-nodulating soybean isolate was selected as the reference plant for this study. Reference plants should not fix nitrogen and should be of similar phenology and growth form, with similar rooting pattern, and be affected in the same fashion by changes in the environment (Boutton 1991). Many factors might contribute to variations among reference plants; horizontal or vertical heterogeneity in the ^{15}N abundance of soils is one of them (Houngnandan et al. 2008).

The lack of consistent estimates of the ^{15}N NA technique to the appraisal of the symbiotic dependence of N_2 – fixing plants can be related to asynchrony of mineral N uptake by legumes, the insufficient difference in delta value between the atmosphere and soil available N, and reference plant and errors in the estimation of the B value (Chalk et al. 2016). The B value corrects for any isotopic discrimination during the uptake and redistribution of symbiotically fixed N (Bergersen et al. 1985; Shearer and Kohl 1986). At high N fixation rates, the methodology becomes insensitive to changes in reference plant $\Delta^{15}\text{N}$. At low N fixation rates (high legume delta ^{15}N values), appropriate selection of a reference plant that closely matches the legume will produce more accurate estimations. Agricultural soils tend to be enriched (4-17%) in ^{15}N relative to atmospheric N_2 (Unkovich et al. 1994a). Differences between cultivars of common bean (*Phaseolus vulgaris* L) in B value throughout the growing season and with seed size have been identified (Pacheco et al. 2017). For field pea (*Pisum sativum*) and lupin (*Lupinus angustifolius*) shoot $\Delta^{15}\text{N}$ decreased with plant age with more slope for field pea.

This research evaluated N fixation rates for a modern soybean variety grown under field conditions and with two contrasting management systems. Few references can be found in the literature measuring N fixation rates (Harper 1974; Thibodeau and Jaworski 1975; Patterson and LaRue 1983). Most of the previous studies agree that maximum N fixation rates occur during the pod filling stage. In a newer study, Fabre and Planchon (2000) reported a maximum N fixation rate at the R₅- stage. Gaspar et al. (2017) reported N uptake rates in soybean yields ranging from 3.6 to 5.5 Mg ha⁻¹ without determining the contribution of N fixation to plant N demand. There is a need to accurately estimate N fixation to provide better estimations of plant N demand and overall N balance (N fixed minus removed in seeds) for soybean systems.

CONCLUSIONS

The ^{15}N NA technique was implemented to estimate BNF throughout the soybean growth cycle. Differences between the B value of soybean varieties, growth stages, and plant fractions were documented in this study. Overall, for the entire plant, the B value at the peak of biomass accumulation was of -1.97. The mode reported in the literature was -1.70, with a minimum of -3.86 and a maximum of 0.20 for soybeans. When the B value was utilized to estimate BNF in field-grown soybeans, intensification (high input) significantly increased fixation N rate from 1.4 to 2.9, and 3.7 to 6.1 kg N ha⁻¹ under and irrigated conditions, respectively, relative to the control (CP, low input). The peak of N fixation was approximately at the onset of the pod formation process. Across all factors, N fixation percentage ranged from 45 to 56%. For the level of fixation measured in this experiment, the source of B value had a small impact on BNF estimation. Future research should be focused on more accurately quantifying N fixation for high-yielding soybean systems (> 7 Mg ha⁻¹) for improving N balance estimations.

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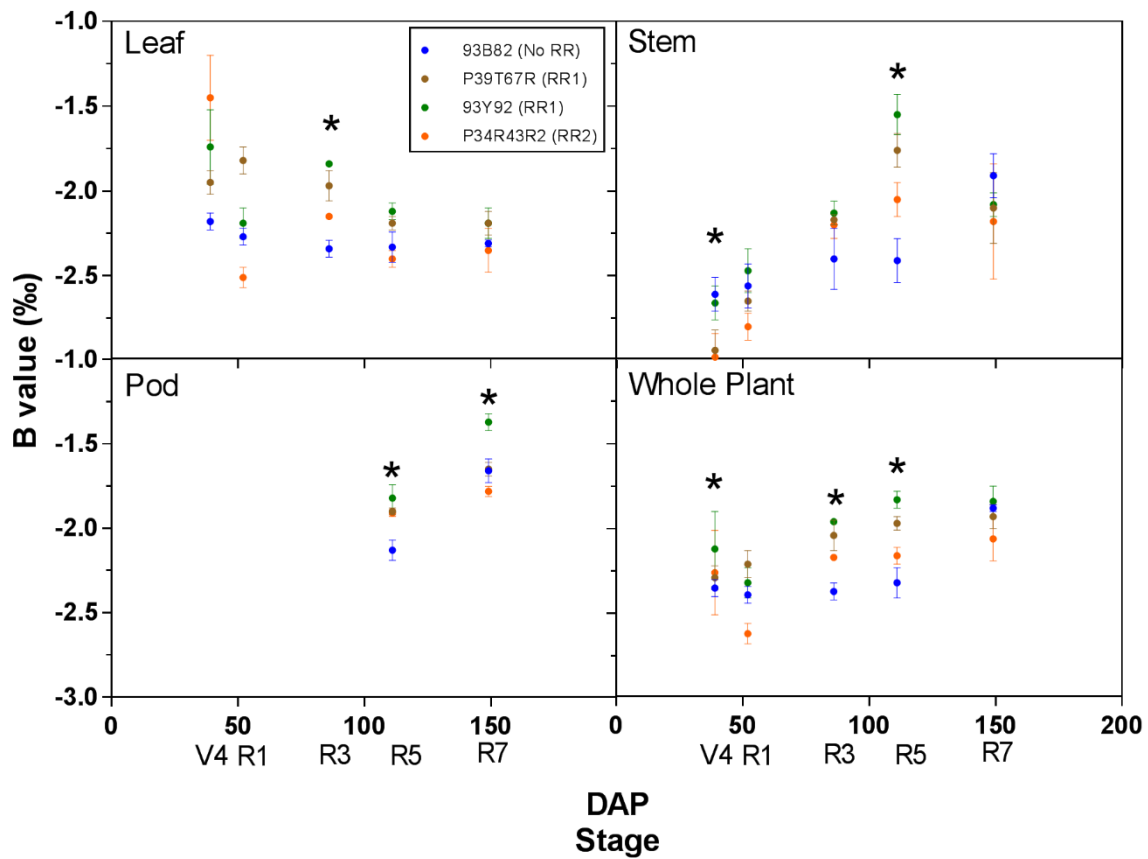


Figure 3.1 B value (%) for leaf (a), stem (b), pod (c) and whole plant (d) for four soybean varieties by phenological stage growth in greenhouse experiment. Asterisk indicates statistical differences for each specific plant part and stage ($p < 0.05$).

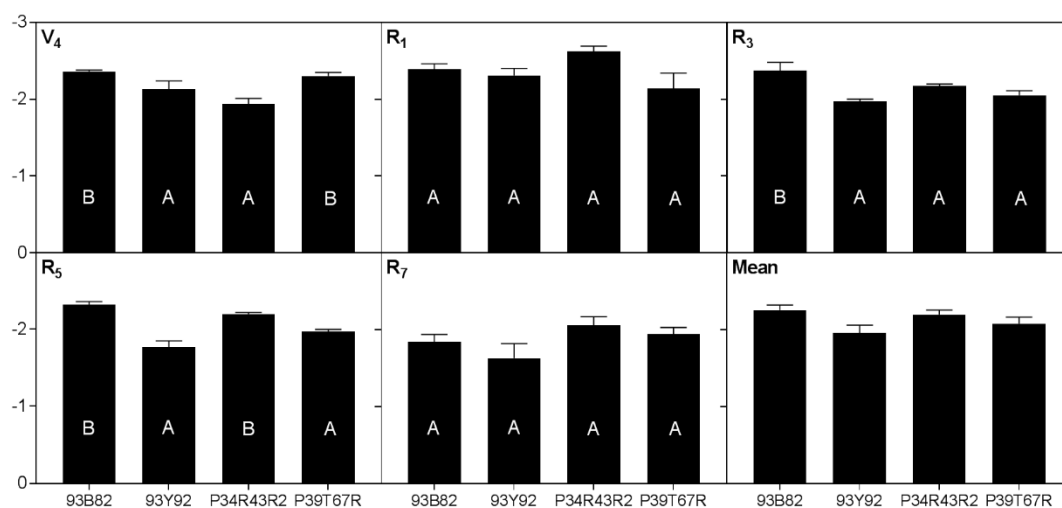


Figure 3.2 B value (%) for variety and phenological stage in soybean. Different letters within phenological stage indicate statistical differences between varieties ($p < 0.05$).

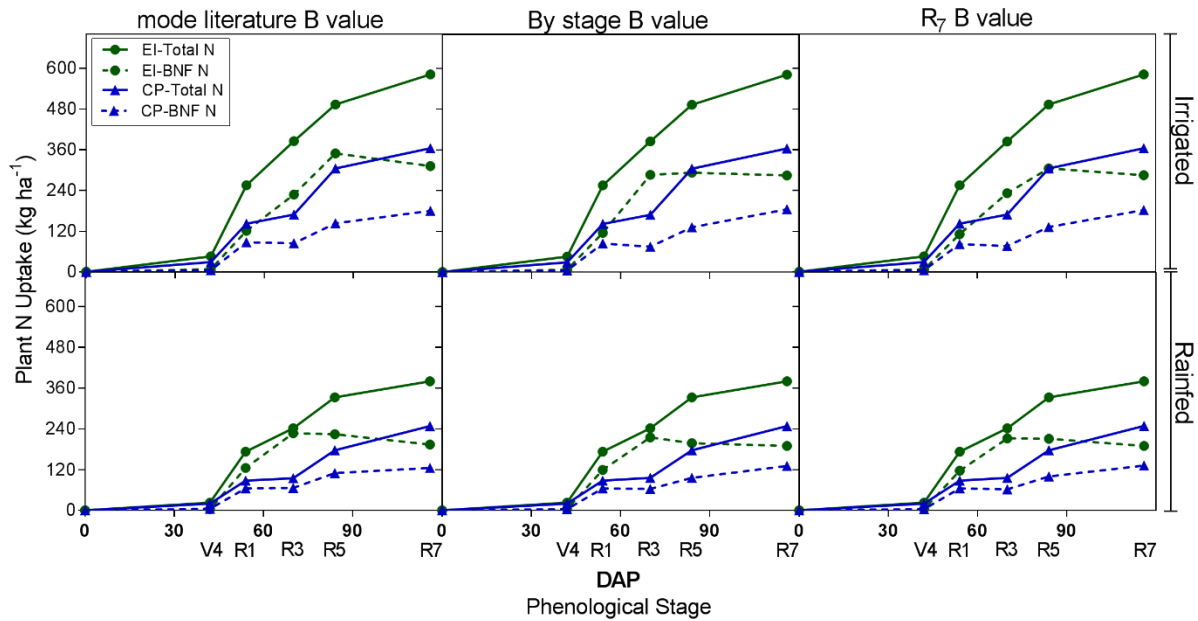


Figure 3.3 Plant N uptake (green and solid blue line) and proportion of the N coming from BNF (green and blue dash lines) by days after planting and phenological stage under irrigated (upper panels) and rainfed (lower panels) scenario. For BNF estimation the mode B value from literature (left panels), B value estimated in the greenhouse by stage (central panels) and R₇ (biomass peak) B value (right panels) was retrieved; and then multiplied by stage uptake to estimate the contribution of fixation.

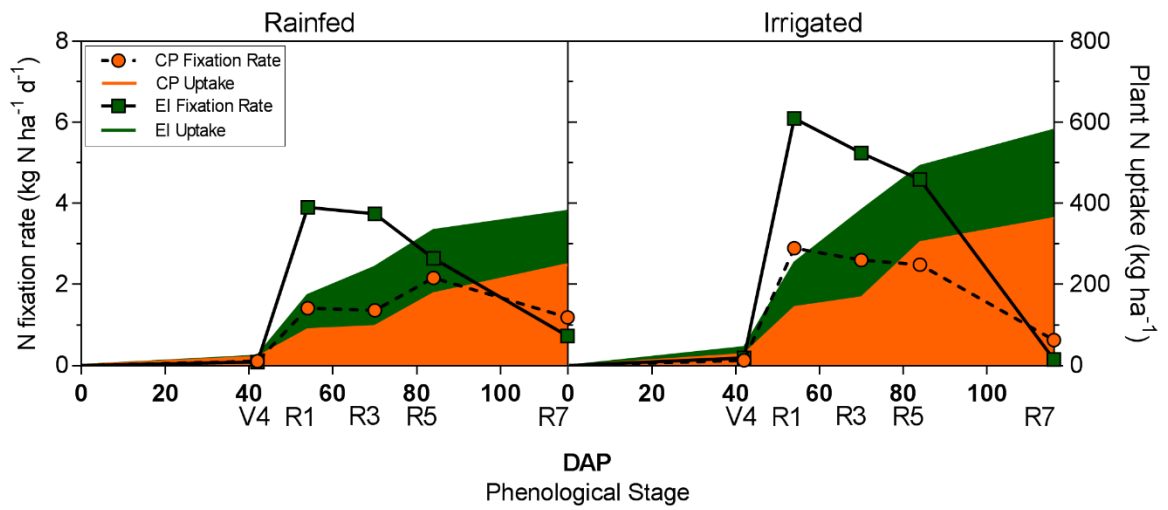


Figure 3.4 Calculated soybean nitrogen fixation rate (green square continuous line, EI; orange circle dash line, CP) and plant N uptake (green, EI; orange, CP) among season (days after planting and phenological stage) under rainfed (left panel), and irrigated (right panel) scenario.

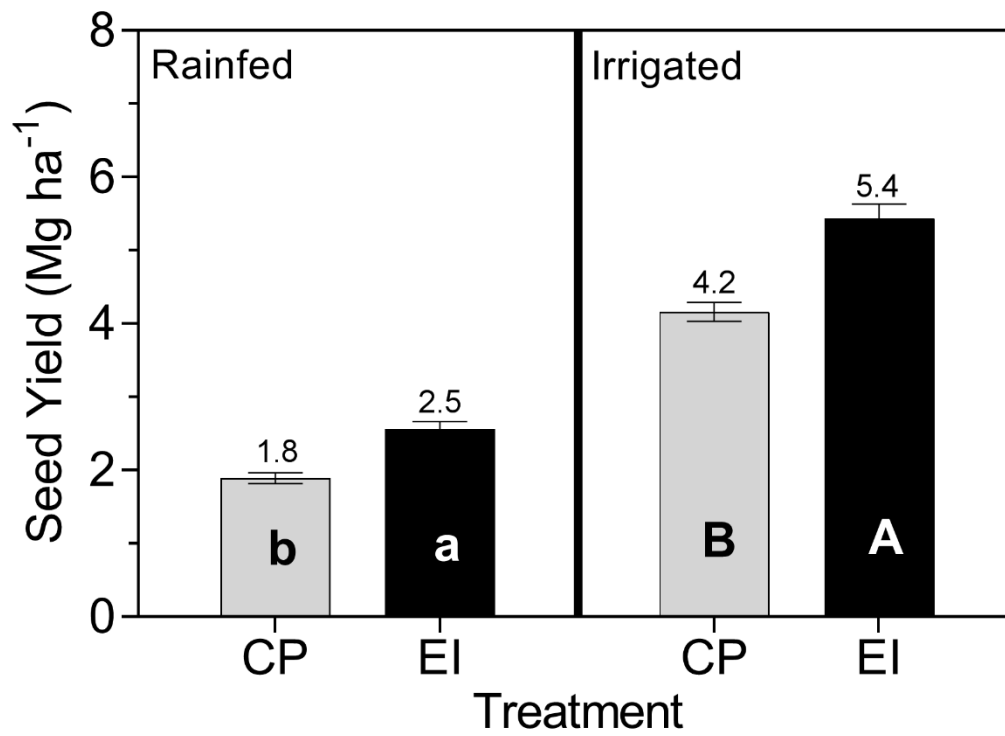


Fig. S1. Seed yield for common practices (CP) and ecological intensification (EI) treatment, under rainfed and irrigated conditions for the BNF estimation experiment at Scandia, KS. Different letters within water environment (rainfed/irrigated) indicates statistical differences between treatments ($p < 0.05$).

Table 3.1 B value for soybean reported retrieved from references.

Author	Publication	'B' values	Author	Publication	'B' values
Kohl et al.	1980	6.50	Houngnandan et al.	2008	-0.78
Kohl and Shearer	1980	0.98	Houngnandan et al.	2008	-0.51
Mariotti et al.	1980	-1.83	Houngnandan et al.	2008	-1.53
Shearer et al.	1980	-0.90	Houngnandan et al.	2008	0.20
Steele et al.	1983	-2.00	Houngnandan et al.	2008	-2.77
Turner and Bergersen	1983	1.00	Houngnandan et al.	2008	-0.01
Yoneyama et al.	1986	-1.54	Guimaraes	2008	-1.99
Bergersen et al.	1989	-1.30	Schipanski et al.	2010	-1.98
Herridge et al.	1990	-0.59	Schipanski et al.	2010	-2.26
Peoples and Herridge	1990	-1.30	Pauffero et al.	2010	-3.38
Peoples et al.	1997	-2.50	Pauffero et al.	2010	-2.69
Okito et al.	2004	-1.83	Pauffero et al.	2010	-3.46
Okito et al.	2004	-3.61	Pauffero et al.	2010	-3.20
Ojiem et al.	2007	-2.00	Pauffero et al.	2010	-3.56
Oberson et al.	2007	-0.88	Pauffero et al.	2010	-3.26
Oberson et al.	2007	-0.49	Pauffero et al.	2010	-1.75
Guimaraes et al.	2008	-1.84	Pauffero et al.	2010	-1.53
Guimaraes et al.	2008	-3.67	Pauffero et al.	2010	-1.35
Houngnandan et al.	2008	-1.32	Pauffero et al.	2010	-2.02
Houngnandan et al.	2008	-1.56	Pauffero et al.	2010	-1.91
Houngnandan et al.	2008	-0.47	Pule-Meulenberg et al.	2011	-1.00
Houngnandan et al.	2008	-1.88	Pule-Meulenberg et al.	2011	-0.72
Houngnandan et al.	2008	-1.24	Schweiger et al.	2014	-1.30
Houngnandan et al.	2008	-2.07	Collino et al.	2015	-1.03
Houngnandan et al.	2008	-0.48	Peoples	Unpublished	-1.70
Houngnandan et al.	2008	0.10	Peoples	Unpublished	-1.10
Houngnandan et al.	2008	-1.71			
Houngnandan et al.	2008	-0.01			
Houngnandan et al.	2008	-0.65			

Table 3.2 B value, biomass per plant and N content by soybean variety, stage, and plant fraction for the greenhouse study to determine B value.

	B value	Biomass g ⁻¹	N Cont.
	P value		
Variety	0.0001	0.0001	0.5
Stage	0.0001	0.0001	0.0001
Fraction	0.0001	0.004	0.0012
Fraction*Variety	0.93	0.36	0.14
Fraction*Stage	0.07	0.21	0.18
Variety*Stage	0.21	0.12	0.10
Fraction*Variety*Stage	0.29	0.36	0.65
Variety			
93B82	-2.19a	2.28a	2.83
P34R43R2	-2.11ab	2.07a	3.00
P39T67R	-2.02b	2.77b	2.87
93Y92	-1.96b	2.21a	2.87
Fraction			
Lv	-2.12a	1.70b	3.12b
St	-2.18a	1.60b	1.46a
Pd	-1.78b	2.07a	4.47a
Stage			
V4	-1.98b	0.37a	2.21a
R1	-2.35a	1.06b	3.06b
R3	-2.11b	1.69c	2.98b
R5	-2.05b	2.74d	2.82b
R7	-1.97b	4.85e	3.20b

Lv, leaf; St, stem; Pd, pod.

Table 3.3 B value, biomass per plant and N content by plant fraction, phenological stage, and soybean variety. Greenhouse study.

Soybean Variety	Fraction.	Phenological Stage														
		V ₄			R ₁			R ₃			R ₅			R ₇		
		B value‰	Biomass (g pl ⁻¹)	N cont. (mg kg ⁻¹)	B value‰	Biomass (g pl ⁻¹)	N cont. (mg kg ⁻¹)	B value‰	Biomass (g pl ⁻¹)	N cont. (mg kg ⁻¹)	B value‰	Biomass (g pl ⁻¹)	N cont. (mg kg ⁻¹)	B value‰	Biomass (g pl ⁻¹)	N cont. (mg 100g ⁻¹)
93B82	Lv	-2.18	0.55	2.82	-2.27	1.32	3.90	-2.34	2.88	3.34	-2.33	1.97	3.24	-2.31	3.30	2.41
	St	-2.61	0.38	1.05	-2.56	0.99	1.60	-2.40	2.51	1.87	-2.41	2.17	1.65	-1.91	2.28	1.06
	Pd	-	-	-	-	-	-	-	-	-	-2.13	1.43	3.37	-1.66	6.40	5.38
	Pl	-2.35	0.92	2.09	-2.39	2.31	2.94	-2.37	5.39	2.65	-2.32	5.56	2.64	-2.03	14.25	3.60
P39T67R	Lv	-1.95	0.41	2.69	-1.82	1.21	3.29	-1.97	4.38	3.51	-2.19	3.18	3.63	-2.19	4.25	2.45
	St	-2.94	0.22	1.04	-2.65	1.06	1.46	-2.17	2.99	1.63	-1.76	2.75	1.88	-2.10	5.02	1.27
	Pd	-	-	-	-	-	-	-	-	-	-1.90	1.00	3.28	-1.65	6.73	5.40
	Pl	-2.30	0.64	2.11	-2.19	2.27	2.46	-2.05	7.37	2.75	-1.97	6.93	2.87	-1.93	16.00	3.32
93Y92	Lv	-1.74	0.41	2.60	-2.19	1.06	3.60	-1.84	2.60	3.07	-2.12	1.10	3.19	-2.19	4.15	3.24
	St	-2.66	0.29	0.75	-2.47	1.05	1.75	-2.13	1.95	1.36	-1.55	2.00	1.74	-2.08	4.97	1.32
	Pd	-	-	-	-	-	-	-	-	-	-1.82	1.26	3.32	-1.37	5.70	5.24
	Pl	-2.12	0.70	1.82	-2.33	2.12	2.66	-1.96	4.55	2.34	-2.00	4.36	2.88	-1.84	14.82	3.36
P34R43R2	Lv	-1.45	0.46	2.74	-2.51	1.05	3.42	-2.15	2.42	3.44	-2.40	1.78	3.24	-2.35	4.05	2.55
	St	-1.95	0.23	1.87	-2.80	0.73	1.77	-2.20	2.22	1.83	-2.05	1.09	1.76	-2.18	4.23	1.80
	Pd	-	-	-	-	-	-	-	-	-	-1.91	0.58	3.52	-1.78	5.95	6.27
	Pl	-1.62	0.69	2.44	-2.63	1.78	2.74	-2.18	4.63	2.67	-2.20	3.43	2.82	-2.06	14.23	3.89

Cont., content; Lv., leaves; St., stem; Pd., pod; Plt., plant.

Table 3.4 Soybean stem, leaf and seed biomass and N concentration measured, and biological nitrogen fixation estimation by treatment at V₄, R₁, R₃, R₅ and R₇ growth stages for 2015 growing season. Scandia KS.

Trt.	WC	Stem					Leaf					Seed	
		V ₄	R ₁	R ₃	R ₅	R ₇	V ₄	R ₁	R ₃	R ₅	R ₇	R ₅	R ₇
Biomass (g m ⁻²)													
CP	Rainfed	28(4)	145(17)	116(20)	492(16)	382(21)	32(4)	140(16)	169(16)	338(18)	157(7)	104(6)	187(5)
	Irrigated	42(1)	307(55)	264(8)	290(6)	532(13)	47(2)	244(32)	197(8)	221(20)	250(28)	317(8)	415(12)
EI	Rainfed	34(7)	272(18)	723(14)	525(17)	411(17)	40(8)	268(26)	498(12)	455(12)	245(26)	215(20)	260(7)
	Irrigated	69(9)	486(29)	758(12)	807(18)	829(25)	76(10)	373(25)	485(55)	594(31)	389(51)	485(19)	543(19)
Nitrogen Concentration (g 100g ⁻¹)													
CP	Rainfed	1.87(0.29)	1.7(0.05)	1.3(0.36)	1.15(0.23)	1(0.12)	4.69(0.55)	4.52(0.11)	4.73(0.16)	4.03(0.2)	2.13(0.12)	5.31(0.32)	5.6(0.25)
	Irrigated	1.73(0.06)	1.5(0.13)	1.67(0.18)	1.22(0.17)	1.18(0.32)	4.46(0.27)	5.01(0.23)	5.1(0.35)	4.39(0.16)	2.32(0.18)	5.42(0.17)	5.83(0.42)
EI	Rainfed	1.7(0.7)	1.65(0.18)	1.34(0.06)	1.32(0.22)	1.15(0.2)	4.35(0.46)	4.77(0.22)	4.72(0.2)	4.26(0.28)	1.85(0.08)	5.39(0.29)	5.74(0.28)
	Irrigated	1.72(0.25)	1.49(0.13)	1.74(0.4)	1.55(0.35)	1.14(0.17)	4.51(0.32)	4.89(0.26)	5.21(0.22)	3.14(1.48)	2.13(0.24)	5.56(0.23)	5.8(0.5)
Estimated Biological Nitrogen Fixation (%)													
CP	Rainfed	18(9.3)	64(1.3)	39(1.4)	37(1.8)	59(5.8)	23(9.1)	78(1.2)	71(2.3)	30(2.9)	79(8.9)	61(4.6)	78(3.7)
	Irrigated	13(5.2)	28(5.3)	38(1.1)	38(1.3)	73(2.8)	18(3.7)	58(3.2)	58(0.3)	25(3.1)	73(3.4)	50(6.7)	73(4.9)
EI	Rainfed	11(0.4)	57(2.1)	45(2.8)	39(2.5)	51(6.2)	18(4.7)	73(1.4)	73(0.4)	42(7.8)	79(5.2)	77(4.5)	87(1.19)
	Irrigated	16(3.6)	36(9.2)	66(6.1)	36(0.3)	39(6.7)	16(9.4)	49(4.6)	79(3.8)	62(6.3)	59(8.2)	49(4.9)	63(3.5)

CP. Common Practices. EI Ecological intensification. WC, water condition. Number in parenthesis indicate standard error. Trt. Treatment.

Chapter 4 - Quantifying yield gaps in a maize-soybean rotation system under high and low management inputs in the Western US Corn Belt using APSIM

ABSTRACT

Quantifying yield gaps (potential yield minus actual yield) and identifying practices to close those gaps is critical for sustaining high-yielding production systems. The objectives of this study were to 1) calibrate APSIM (the Agricultural Production Systems Simulator) for modeling a maize-soybean rotation under two contrasting input levels, and 2) to apply the parametrized model to estimate the yield gap in different weather years in the western US Corn Belt. The APSIM model was calibrated for in-season crop growth data (four site-years per crop), and the parameterized model was utilized to estimate the yield gap as a function of management (high- vs. low-input) and weather conditions (wet-warm, wet-cold, dry-warm and dry-cold years). Experimental data collected in Scandia, Kansas, US over two seasons (2014-2015) was used to train the model. Experimental data included two management systems: : 1) common practices (CP, low input), wide row spacing, lower seeding rate, and lack of nutrient applications (except N in maize), and 2) ecological intensification (EI, high input), narrow rows, high seeding rate, and balanced nutrition (application of P, K, and S). Results indicated that APSIM simulated aboveground biomass and yield data for maize and soybean crops reasonably well in all site-years, with low relative root mean square error (RRMSE =18-21 and 18-31 for maize and soybean biomass and yield respectively), high model efficiency ($E = 0.92-0.81$ and $0.88-0.75$), and R^2 of $0.98-0.94$ and $0.88-0.75$. Model simulations for 37 years indicated an average maize

yield gap was 4.2 and 2.5 Mg ha⁻¹ for low- and high-input, respectively. Similarly, the soybean yield gap was 2.5 and 0.8 Mg ha⁻¹. We also found that modeled potential yield was more sensitive to weather changes for maize relative to soybeans. The 37-year simulation showed that the high-input system maintained more yield stability across all weather patterns. In addition, the size of the yield gap was reduced by approximately half under irrigated maize (4.2 vs. 2.5 Mg ha⁻¹) and soybeans (0.8 vs. 0.4 Mg ha⁻¹) with intensified management, from low- to high-input systems. Irrigation reduced yield variation in maize, reflected by lower CV relative to the rainfed scenario, with a lower impact on soybean yields. This study provides the first yield gap assessment of maize and soybean in Kansas to initiate dialogue (both experimental and modeling activities) on best practices to close the gaps in rainfed and irrigated systems.

Keywords: soybean, maize, yield gap, modeling, rotation, nutrient, biomass, APSIM.

INTRODUCTION

Yield potential is defined as the yield of a crop grown in an adapted environment, where nutrient and water are supplied with no restriction, and with pest, diseases, weeds and other stresses controlled. The difference between potential and actual yield is defined as the yield gap (Evans and Fischer, 1999). The potential yield is variable from year to year given specific temperature and radiation; the actual yield and the associated yield gap are likewise variable (Van Ittersum et al., 2013). Farmer management practices (planting date, row spacing, fertilization, disease control, weed control) will impact actual yields. A deep understanding of the main factors affecting yield is necessary to design sustainable and profitable cropping systems under the myriad array of environments. To analyze yield-limiting factors, the yield gap concept is very useful (Van Ittersum et al., 2013). Within this context, there are three production situations: i) potential yield (determined by CO₂, radiation, temperature, genotype) ii) yield limited by water and/or nutrients, and iii) actual yield influenced by biotic stressors (weeds, pest, diseases) in addition to water and nutrients.

Identifying best management practices such as row spacing, nutrient application rate and timing among others for closing yield gaps is challenging because of the dynamics associated with weather variability, soil conditions and genotype choices (Fischer et al., 2014). Furthermore, the wide range of options for crop and soil management, and genotypes as well the long time-scale underpinning the outcome of these choices, makes it difficult to develop sound strategies to close yield gaps purely based on field studies. Modeling has been reported in the literature as a tool to estimate yield gaps in different crops: soybean (Bhatia et al., 2013; Grassini et al., 2015a), chickpea (Bhatia et al., 2013), maize (Affholder et al., 2013; Grassini et al., 2011; Liu et al., 2012), rice (Affholder et al., 2013), millet (Affholder et al., 2013), wheat (Hochman et

al., 2016; Van Rees et al., 2014). Efforts have been made to estimate yield gaps at regional and global scales through modeling implementation (Guilpart et al., 2017; Rattalino Edreira et al., 2017; van Bussel et al., 2015; Van Ittersum et al., 2013; Van Wart et al., 2013).

Cropping system models can simulate crop growth, water balance and N cycling, and they can predict complex interactions between soil-crop processes. Models can facilitate the development of long-term strategies for sustainable farm management (Cao et al., 2015; Thorburn et al., 2005). The Agricultural Production Systems sIMulator model (APSIM, Holzworth et al., 2014; Keating et al., 2003) has been successfully used to simulate various aspects of cropping systems around the globe. Examples of APSIM application include: soybean planting date by maturity interactions (Archontoulis et al., 2014a), yield forecasting (Carberry et al., 2009; Togliatti et al., 2017), growth and development in legume species (Robertson et al., 2002), N fixation in legumes (Chen et al., 2016), irrigation support (Zhang and Feng, 2010), maize seeding rate (Lyon et al., 2003), and more recently long-term N fertilization responses in maize (Puntel et al., 2016) and water quality aspects (Dietzel et al., 2016; Martinez-Feria et al., 2016).

Kansas has a strong agricultural history, ranking number seven in maize and ten in US soybean production by state (Kansas Department of Agriculture, 2017). The climate in Kansas is warmer and drier compared to Midwestern states such as Iowa, Illinois, and Minnesota that are top producing maize states in the US (Kellner and Niyogi, 2015). Consequently, the range of management options that farmers have is larger than that of the other states, adding more complexity to the decision-making process. For example, the maize planting window is from April 5 to May 25 (USDA-NASS, 2010); the relative maturity (RM) range from 106 to 115 (DuPont Pioneer, 2017); and county-yield level ranges from 2 to 15 Mg ha⁻¹ (USDA, 2017). In

some Kansas regions, irrigation becomes an important production factor. The current challenge is to categorize different production levels for maize and soybean crops, quantify their respective yield gaps, and identify management systems to close those gaps.

To our knowledge, there is no prior yield gap analysis for Kansas targeting common rotation systems such as maize-soybean. Thus, the objectives of this study were to 1) calibrate the APSIM maize and soybean models using local field experimental data and 2) use the calibrated model to calculate a long-term (1980-2016) yield gap analyses as a function of management (high- vs low-input) and weather conditions (wet-warm, wet-cold, dry-warm and dry-cold years) in the western US Corn Belt.

MATERIALS AND METHODS

Experiments

A maize-soybean rotation was established in 2014 under rainfed and irrigated conditions at the Northcentral Kansas Experiment Field near Scandia, Kansas, US (39°49'54"N; 97°50'21"W). The data generated by these studies were used to calibrate the APSIM model. The soil is classified as a Crete silt loam (fine, montmorillonitic, mesic Panchic Argiustoll). Both phases of the rotation were present each year, except for rainfed maize in 2014. The climate at the site is classified as warm-humid continental (Peel et al., 2007), with a mean annual temperature of 11.8 °C and precipitation of 713 mm. About 75% of the rain occurs during the season (April-October).

A total of six datasets were developed to calibrate and validate the APSIM model. Each dataset had two years of field data:

- 1) irrigated, maize–soybean with low-input practices (common practices, CP);
- 2) irrigated, maize-soybean with high-input practices (ecological intensification, EI)
- 3) irrigated, soybean-maize with low-inputs (CP)
- 4) irrigated, soybean-maize with high-inputs (EI)
- 5) rainfed, soybean-maize with low-inputs (CP)
- 6) rainfed, soybean-maize with high-inputs (EI)

The irrigated treatments were used to calibrate the model, and the rainfed treatments were used as validation datasets. Table 1 presents the information for the low- and high- management inputs. Briefly, the low-input practices included seeding rates of 7.4 and 27.4 plants per m², respectively for maize and soybean; row spacing of 0.76 m, and no application of P, K, and S fertilizers. The high-input practices included seeding rates of 8.9 and 42.9 plants per m²,

respectively for maize and soybean; row spacing of 0.38 m, balanced nutrition (N, P, K, and S application), fungicides, and insecticide application as needed. For treatments receiving fertilizer application, nutrients were broadcasted at planting using MicroEssentials® SZ^(TM) (12-40-0-10S-1Zn) and Aspire® (0-0-58-0.5B) (Mosaic Co.) for both maize and soybean crops. Nitrogen source for maize was urea ammonium nitrate (UAN, 32-0-0). The criteria to decide P, K and S fertilizer rate was replacement by providing all nutrients according to the grain/seed removal driven by yield target. The criterion for N fertilization for maize was to follow a balanced approach, calculated as the difference of N provided by soil N (soil test) and crop N demand according to a yield target. Rates under irrigation and rainfed conditions were different since maximum attainable yield for the irrigated environment was greater. All fertilizer rates applied in maize and soybean are presented in Table 4.1. Plot size was 15 m length, and 6 m width replicated five times in a complete randomized block design. Maize plots in 2015 were placed in the same location as the soybean plots for the 2014 season with the goal of documenting residual effects from the management tested. In-season measurements were conducted in three replications out of five. Yield moisture was adjusted to 13 g kg⁻¹ for soybean and 15.5 g kg⁻¹ for maize. Aboveground biomass and nutrient concentrations were expressed on dry matter basis. Field experiments under irrigation during 2014 and 2015 growing season received 160 mm and 190 mm of irrigation respectively.

Measurements

Phenological stage, biomass and total N concentration in dry basis per organ (stem, leaves, grain) were measured at V₁₃, R₁, R₃, R₆ growth stages for maize (Ritchie et al., 1986) and at R₁, R₃, R₆, R₇ growth stages for soybean (Fehr, et al., 1971). Aboveground biomass was determined using a destructive method where 1 m² area was harvested each time. Plants were counted and

fractioned in stem, leaves and reproductive organs if present. All plant samples were dried at 65 °C to constant weight. Samples were ground (1 mm mesh) in preparation for N concentration (g 100 g⁻¹), which was determined via the combustion method (AOAC, 2000). Nitrogen content for each fraction was calculated as the product of N concentration multiplied by its dry mass.

APSIM Model Setup and calibration

The APSIM model (Holzworth et al., 2014; www.apsim.info) is an open source modular modeling framework utilized for the mechanistic analysis of agricultural systems. The APSIM version 7.7 was used in this study in a sequential node (starting Jan 1, 2014). APSIM was set up by connecting the following models: maize and soybean crop models (Keating et al., 2003; Robertson and Carberry, 1998); Soil N (soil N and C cycling model with default soil temperature model; Probert et al., 1998); SoilWat (a tipping bucket soil water model; Probert et al., 1998); SURFACEOM residue model, (Probert et al., 1998; Puntel et al., 2016; Thorburn et al., 2001, 2005) and the following management rules: planting, harvest, fertilizer, tillage, and rotations (Keating et al., 2003).

The APSIM model requires management information, weather and soil conditions as well as initial values (day one of simulation) for soil-root-residue parameters to provide outputs. Initial model conditions such as root mass, surface residue mass, soil water, soil nitrate, and soil organic carbon (SOC) pool partitioning were obtained by starting the model six years prior to the start of the experiment, similar to Dietzel et al. (2016). Model initialization matched soil organic matter and pH measurements (data not presented). Hydrological parameters needed to characterize the soil profile in APSIM such as field capacity and others were retrieved from the Web Soil Survey (Web Soil Survey, 2016) and converted to APSIM format using the

methodology outlined in Archontoulis et al. (2014a). The profile parameter values are provided in Table B.1.

A 110-relative maturity maize hybrid and a maturity group 4 soybean variety were initially selected from the APSIM cultivar database to represent the genetic characteristics of the cultivars used in the field experiment. These cultivars were developed using data from US Midwest and were used as a starting point in this exercise (Archontoulis et al., 2014a, b). During model calibration, some cultivar parameters were modified to better fit experimental data. The calibrated cultivar parameters are provided in Table B.2. In addition, we updated the radiation use efficiency (RUE) crop parameter for maize from 1.6-1.4 to a constant value of 1.85 (Lindquist et al., 2005). The default RUE consistently underestimated yields and biomass production.

Solar radiation (MJ m^{-2}), precipitation (mm), and maximum and minimum temperature ($^{\circ}\text{C}$) at daily intervals were obtained from the Scandia Weather Data Station (Mesonet K-State, 2017) located at 300 m from the experiment. Historical weather data (1980-2016) were retrieved from Daymet (Mesonet K-State, 2017; Thornton et al., 2017). The model output variables for both crops were: phenology, biomass production and partitioning to stem, leaves, and seeds, tissue nitrogen concentration and content per organ. Biomass and yield data reported here are on dry basis.

Long-term simulation

A 37-yr long-term maize-soybean rotation was simulated with APSIM to quantify differences between CP (low-input) and EI (high-input) in both rainfed and irrigated scenarios. A total of eight scenarios were simulated as results of the combination of crops, water scenarios and management systems. Four additional simulations were performed with no water and N

limitations to estimate potential yields per year and to calculate the associated yield gap (potential minus actual yield). All 12 simulations were ran (37 times) using weather records from 1980 to 2016 (Mesonet K-State, 2017; Thornton et al., 2017). Annual yield was the main output retrieved from the model.

Weather conditions

Weather conditions for the period 1980-2016 have been summarized in Fig. B.1 by plotting mean cumulative precipitation and mean temperature for each season (April 1st– October 1st) by year. Dotted vertical and horizontal lines indicate normal mean temperature and cumulative precipitation for the mentioned period. This analysis yields four weather patterns: cool and wet (upper left panel; i.e., 2008, 1982), warm and wet (upper right panel; i.e., 2001, 2016), cool and dry (lower left panel; i.e., 1997, 2009) and warm and dry (lower right panel; i.e., 2000, 2012). Values close to the vertical and horizontal dotted lines indicate weather close to normal (for the long-term historical 1980-2016 evaluation). Weather conditions for the period demonstrated a wide range of environments from the four patterns described above.

Statistical analysis

To evaluate the performance of APSIM simulated outputs were compared with observed data. Relative root mean square error (RRMSE, the lower the value, the better -Eq.1-), modeling efficiency (E, the higher the value the better-Eq.2-) and the coefficient of determination (R^2 , the higher the value the better-Eq.3-) (Archontoulis and Miguez, 2015) were calculated as follows:

$$RRMSE = \frac{RMSE}{\bar{o}} \times 100 \quad RMSE = \sqrt{\frac{\sum_{i=1}^n (S_i - O_i)^2}{n}} \quad (\text{Eq.1})$$

$$E = 1 - \frac{\sum (S_i - O_i)^2}{\sum (O_i - \bar{O})^2} \quad (\text{Eq.2})$$

$$R^2 = 1 - \frac{SS_{residual}}{SS_{total}} = 1 - \frac{\sum_{i=1}^n (S_i - O_i)^2}{\sum_{i=1}^n (S_i - O_i)^2 + \sum_{i=1}^n (S_i - \bar{O})^2} \quad (\text{Eq.3})$$

Observed and simulated values were plotted using GraphPad Prism 7 (Motulsky and Christopoulos, 2003). Observed yield means for maize and soybeans were compared with analysis of variance, after testing normality and variance homogeneity (R Core Team, 2016). Separation of means was evaluated by conducting a Fisher LSD test with a significance level of 0.05.

RESULTS

Field Results: Yield, biomass, and plant N content for maize and soybean

For maize under irrigation, there were no statistical differences between CP and EI in the 2014 season, with yield averaging 14.3 Mg ha⁻¹ (Fig. 4.1A). In the 2015 season, maize yield differential was 1 Mg ha⁻¹ under irrigation, with a maximum yield for EI of 14.1 Mg ha⁻¹. Under rainfed conditions, EI outyielded CP by 1.4 Mg ha⁻¹, with EI achieving a maximum yield of 13.5 Mg ha⁻¹ (Fig. 4.1B). Maize total biomass production at R₆ in 2014 for CP and EI under irrigation was 21.6 and 22.9 Mg ha⁻¹, respectively (Fig. 4.2A, B). In 2015, EI produced 24.5 Mg ha⁻¹ (+14% than CP) and 20.2 Mg ha⁻¹ (+10% than CP) for irrigated and rainfed conditions, respectively (Fig. 4.2 C-F).

In soybean, EI statistically outyielded CP when averaged across both seasons under irrigation (Fig. 4.1 C, D). Larger yield differences between treatments (EI > CP) were observed in 2014 relative to 2015. In 2014 under irrigation, EI outyielded CP by 3.0 Mg ha⁻¹, doubling the yield recorded in the CP treatment (2.9 Mg ha⁻¹). Likewise, in the rainfed scenario, EI outyielded CP by 2.1 Mg ha⁻¹ (Fig. 4.1 C). In 2015, intensification (EI) increased seed yield by 0.7 Mg ha⁻¹ for rainfed and 1.2 Mg ha⁻¹ for irrigated scenarios (Fig. 4.1 D). Total biomass in soybeans for 2014 at R₇ for the EI treatment was 10.5 Mg ha⁻¹ under irrigation (Fig. 4.2 D) and 7.0 Mg ha⁻¹ for rainfed conditions (Fig. 4.2 F). Total soybean biomass production in 2014 at R₇ for the CP treatment was 6.4 and 9.8 Mg ha⁻¹ under irrigation and rainfed conditions, respectively (Fig. 4.2 C, E). In 2015, soybean biomass differences between treatments followed the same trend as in 2014 (Fig. 4.2 A, B).

For maize, plant N content at R₆ ranged from 200 to 324 kg ha⁻¹ (Fig. 4.3). Major plant N content differences between intensification treatments occurred in 2015 season under irrigation, with 285 kg ha⁻¹ in EI and 224 kg ha⁻¹ (-21%) in CP. Under rainfed conditions, plant N content for the EI was 32% greater than for the CP treatment (Fig. 4.3). For soybean, plant N content ranged from 179 to 425 kg ha⁻¹. Averaging both seasons, under irrigation, plant N content for EI was 359 kg ha⁻¹ while for rainfed it was 193 kg ha⁻¹ (Fig. 4.3). For the CP treatment, across seasons, plant N content was 208 and 300 kg ha⁻¹ for rainfed and irrigated scenarios, respectively.

Modeling: Crop phenology and field data simulation

Phenology dates from all the simulations were compared with the recorded dates collected in the field for all site-years evaluated (Table 4.2). Overall, APSIM simulated with adequate accuracy phenology for both maize and soybean crops. Pooling together both seasons, average predictions for soybean flowering and physiological maturity were 6.5 days earlier than observed values. Predictions for maize were more accurate with an average deviation of 2 days for flowering (R₁) and 1 day for physiological maturity (R₆) stage.

The APSIM modeling component reasonably simulated yield, total biomass and plant N content (high E and R²; low RRMSE) for both maize and soybean crops in the rotation (Figs. 4.2-4.4). Observed and predicted values for plant traits under analysis were presented for the six simulations performed in APSIM (Fig. 4.2, yield and plant biomass; and Fig. 4.3, plant N content and grain N content) with the observed values following direction and magnitude of the simulated values. Relative RMSE of grain yield was 18%, and E was 0.76 for maize and with greater RMSE (31%) and lower E (0.53) for soybean seed yield. Plant biomass was predicted with an E of 0.92 for maize and 0.81 for soybean (Fig. 4.4, Table 4.3). The E on the prediction of

plant N content was 0.72 and 0.77 for maize and soybean, respectively. Grain N content for maize was predicted with an E of 0.67 (RRMSE= 19%), while for soybean seed N content the E was lower with a value of 0.43 (RRMSE= 29%) (Fig. 4.4, Table 4.3).

Maize-soybean rotation long-term simulation and yield gap

To address the impact of seasonal weather variability, yields were grouped into four weather scenarios defined above (Fig. B.1). Simulated yields were greater with EI compared to CP (Table 4.4). Maize yield for CP ranged from 7.0 to 8.5 Mg ha⁻¹ (warm and dry to warm and wet) and from 10.5 to 11.2 Mg ha⁻¹ (warm and dry to cool and wet) under both water scenarios. For EI, yields ranged from 6.7 to 9.1 Mg ha⁻¹ (warm and dry to cool and wet) and from 12.6 to 13.7 Mg ha⁻¹ (cool and dry to cool and wet). Soybean yield for CP ranged from 2.1 to 3.1 Mg ha⁻¹ (warm and dry to warm and wet) and from 3.6 to 4.1 Mg ha⁻¹ (warm and dry to cool and dry). The high input system (EI) for soybean seed yields ranged from 2.0 to 3.7 Mg ha⁻¹ (warm and dry to cool and wet) and from 3.9 to 4.5 Mg ha⁻¹ (warm and dry to cool and dry) under both water scenarios (Table 4.4).

After 37 years of simulation, under irrigation, maize yield for EI averaged 12.5 Mg ha⁻¹ (CV 15%) versus 10.9 Mg ha⁻¹ (CV 7.6%) for CP, with a yield gap of 15%. Under rainfed conditions, the yield gap was 7.5% (half the irrigated yield gap), with 7.8 Mg ha⁻¹ (CV 39%) and 7.3 Mg ha⁻¹ (CV 40%) for CP and EI management systems, respectively. Irrigation reduced variation in yield, reflected by the lower CV relative to the rainfed scenario. For soybean, under irrigation EI yielded 4.2 Mg ha⁻¹ (CV 10%) versus the CP yield of 3.8 Mg ha⁻¹ (CV 10%), representing an overall yield gap of 10.5% for the high- vs. low-input system (Table 4.4).

Potential yield for maize was 13.9 Mg ha⁻¹ (warm and dry), 14.7 Mg ha⁻¹ (warm and wet), 15.5 Mg ha⁻¹ (cool and wet) and 16.0 Mg ha⁻¹ (cool and dry). Potential yield for soybean was 4.5 Mg ha⁻¹ (cool and wet), 4.6 Mg ha⁻¹ (warm and wet), 4.7 Mg ha⁻¹ (warm and dry) and 4.8 Mg ha⁻¹ (cool and dry). Maize potential yield was more impacted by the weather than was soybeans (Fig. 4.5).

The average maize yield gap across weather patterns for low-input management (CP) was 7.3 and 4.2 Mg ha⁻¹ under rainfed and irrigated conditions, respectively. For high-input management, the yield gap was 6.3 Mg ha⁻¹ under rainfed and 2.5 Mg ha⁻¹ for irrigated conditions. Soybean average yield gap for CP (low-input) was 2.0 Mg ha⁻¹ for rainfed and 0.8 Mg ha⁻¹ for irrigated conditions; while under high-input (EI) yield gap was 1.8 Mg ha⁻¹ for rainfed and 0.4 Mg ha⁻¹ for irrigated conditions. In soybean, intensification under irrigation reduced the yield gap by 50%, and by 11% for the rainfed scenario (Table 4.4).

For the four long-term weather patterns, high-input (EI) under irrigation produced more yield and reduced the yield gap in both maize and soybean crops relative to the low-input (CP) scenario. In warm and dry conditions, the soybean yield gap was the same for low or high input systems (2.6 Mg ha⁻¹). For both maize and soybean crops in cool years (Fig. 4.5 A, C, E, G), intensification (high input) closed yield gaps in both rainfed and irrigated scenarios.

DISCUSSION

This study quantifies the yield gap for maize and soybean crops grown in rotation in Kansas, USA for the first time. Such information is important to initiation of discussions between stakeholders and modelers to identify and test practices that close the gap while improving profitability and environmental sustainability. Furthermore, this study provides original experimental data on crop yields under different management intensification levels and a calibrated version of the APSIM model for this environment for further use.

Yield gap analyses were previously executed for a number of row crops in different environments using a suite of simulation models (Bhatia et al., 2013; Grassini et al., 2011, 2015b; Liu et al., 2012; Specht et al., 1999; Van Oort et al., 2015; Van Roekel and Purcell, 2014). Previous yield gap assessments focussed on individual crops, but not the cropping system that accounts for the carry-over effects on soil water and nitrogen from one season to another (Iqbal et al., 2018). In this study we used the cropping systems approach and the APSIM model, that is rarely used in the literature (Ref from other environments). Our yield gap analysis was further expanded to clarify maize and soybean yield gaps per weather categories (hot-dry etc.) We found that in most weather patterns, intensification contributed to closing the yield gaps with a greater impact when crops were irrigated.

Overall, the APSIM model performed well in predicting crop yields and N dynamic (Figs. 2.1-2.2). The RRMSE of about 18% for maize yield is comparable to other simulation studies (Archontoulis et al., 2014b; Basche et al., 2016; Bourguignon et al., 2017; Chen et al., 2008; Gaydon et al., 2017; Mohanty et al., 2012; Puntel et al., 2016; Robertson et al., 2002). No attempts were made to further improve the RRSME to values below 18% because according to

He et al., (2017) targeting model calibration with lower RRMSE values is pointless as the simulated error cannot be lower than the inherent error in the measured data used for calibration.

For soybean, narrowing rows from 0.76 to 0.38 m resulted in a positive impact in grain yield; similar responses are portrayed in the literature for other environments (Chen and Wiatrak, 2011; Conley et al., 2008; Costa et al., 1980; Cox and Cherney, 2011; De Bruin and Pedersen, 2008; Ethredge et al., 1989; Hanna et al., 2008; Kratochvil et al., 2004; Lambert and Lowenberg-DeBoer, 2003). The narrow row spacing resulted in more light interception and greater biomass production in our study. For maize, an increase in seeding rate was reported to increase yields as the attainable yield environment improves (Andrade et al., 2002; Assefa et al., 2016; Barbieri et al., 2012; Ciampitti and Vyn, 2011, 2012; Jones et al., 1977). Several authors reported positive response to narrow row spacing in maize (Andrade et al., 2002; Fawcett et al., 2015; Johnson and Hoverstad, 2002; Jones et al., 1977; Lambert and Lowenberg-DeBoer, 2003; Shapiro and Wortmann, 2006), while some have reported none (Farnham, 2001) to negative impact on yield (Maddonni and Martínez-Bercovich, 2014). When selecting best management practices to close yield gaps special attention needs to be paid to maintaining high efficiency, farmer profitability, and to reduced environmental impact.

This research shows that intensification can reduce yield gaps and improve overall yield stability, especially under irrigated conditions for both maize and soybean crops, in concordance with previous studies that evaluated the effect of intensification on closing yield gaps (Cassman, 1999; Fischer et al., 2014; Mueller et al., 2012; Wood et al., 2000). For maize, wheat and rice, climate, fertilizer application, and irrigation can help to explain 60 to 80% of yield variability (Mueller et al., 2012). APSIM differentiated the high- and low-input systems (EI and CP), and

thus the model characterized these systems in a long-term fashion accounting for the year-to-year variability as evaluated by Puntel et al. (2016).

Our study shows that the yield gap in both crops was affected by yearly weather patterns. Analyzing what happened in previous years will help in the process of farmer's decision making. Weather and climate influence cropping area and intensity; different studies in the literature focused on the effect of weather variability on food production from a historical point of view. Weather variability impacted food production differently by region (Iizumi and Ramankutty, 2015; Lobell et al., 2008, 2011). A global scale assessment in yield gaps found that global yield variability is heavily controlled (45 to 70% in most crops) by fertilizer use, irrigation and climate. Thus, research on management practices that include nutrients and irrigation is fundamental to reducing production variability and closing yield gaps.

The challenges of real-world agriculture are to meet the constant increase in food demand while decreasing agriculture's global environmental footprint. Closing yield gaps and improving resource use efficiency will help to achieve these challenges. For example, addressing imbalances and inefficiencies in N and P application by 28% and 38% in maize, wheat and rice will not only impact yield but also greatly decrease agriculture's footprint (Mueller et al., 2012). The APSIM model can also assess the potential negative impacts of N fertilization (Akponikpè et al., 2010; Archontoulis et al., 2014b; Chen et al., 2016; Kisaka et al., 2016; Puntel et al., 2016; Thorburn et al., 2005). There is a need to investigate site-specific management practices for sustainably improving agricultural intensification (Pradhan et al., 2015). Future modeling efforts can explore the impact of different levels of intensification by management zones within a field.

Because there is substantial regional and weather variation in yield gaps, a simulation modeling approach is needed to estimate those gaps. Quantifying the exploitable yield gap in major cropping systems locally and around the globe can establish the basis for future research on crop intensification, irrigation and climate change impacts (Aramburu Merlos et al., 2015), and help estimate future food production capacity. The adoption of intensification should be reserved for appropriate environments and systems so as to minimize undesirable consequences (Ju et al., 2009; Vitousek et al., 2009).

The approach developed in this research represents an important tool to determine if intensification in a specific production system can positively impact the productivity of the farming system and assist in closing yield gaps. The latter task should be assigned priority as a research topic to provide input for local and global food security policy (Licker et al., 2010; Lobell et al., 2009; Van Ittersum et al., 2013; van Oort et al., 2017). The dataset collected, and analysis conducted in this research could be the starting point for another researcher to extract more information via implementation of modeling and simulation of different scenarios. Thus, data repository and data sharing should become essential to moving the science forward through collaborative initiatives among research groups.

CONCLUSIONS

Field data from a maize-soybean experiment with low and high input management systems was collected and analyzed using the APSIM model. Yield gaps were estimated in relation to weather patterns and management intensification in the Western US Corn Belt. Across 37-yr of simulations APSIM determined yield gaps for maize (2.5-7.3 Mg ha⁻¹) and soybean (0.4-2.0 Mg ha⁻¹) under contrasting input levels and water scenarios. For yield gap calculation, modeled potential yield was more sensitive to weather changes in maize than in to soybeans. The high-input system had more yield stability across all weather patterns. The size of the yield gap was reduced by approximately half under irrigation for maize (4.2 vs. 2.5 Mg ha⁻¹) and soybeans (0.8 vs. 0.4 Mg ha⁻¹) with intensified management, from low- to high-input systems.

Overall our results support the use of APSIM as a tool to develop future management strategies to close yield gaps in both rainfed and irrigated maize and soybean systems in the Western Corn belt, USA.

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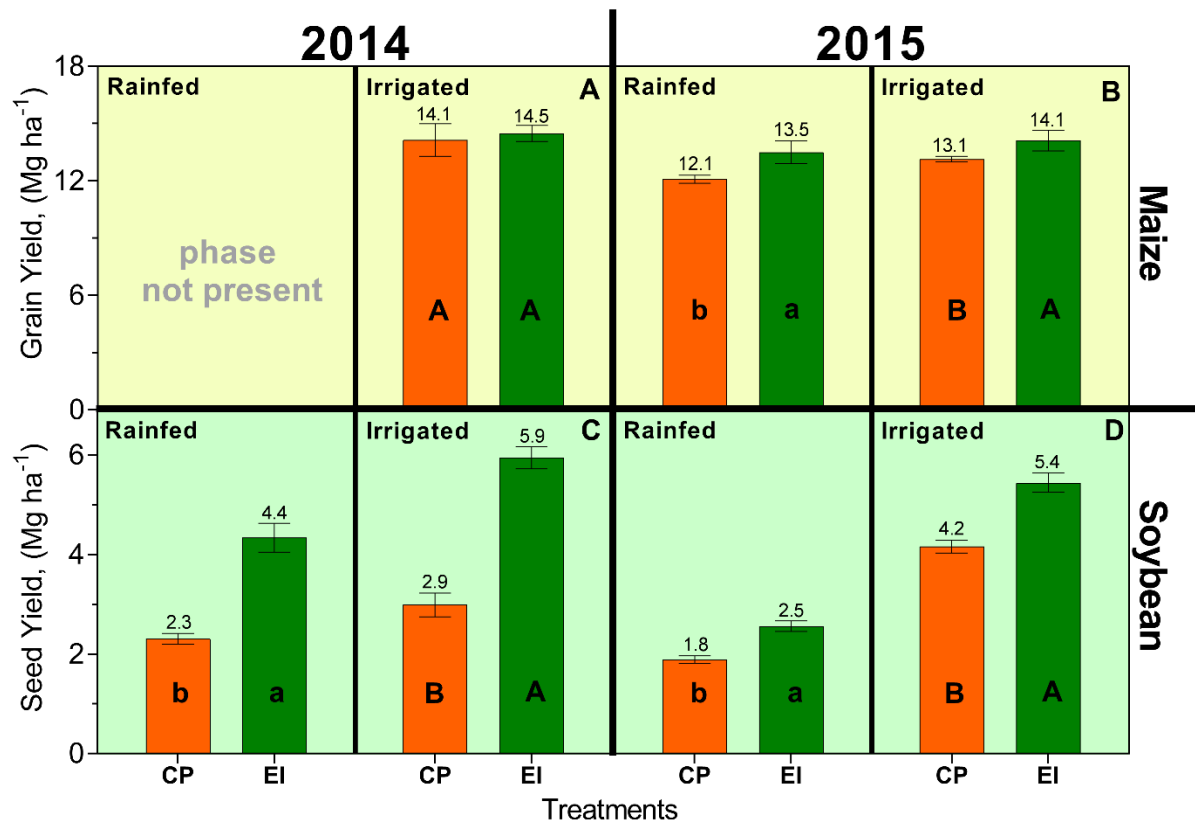


Figure 4.1 Maize grain yield 2014 (a), 2015 (b) and Soybean seed yield 2014 (c) 2015 (d) growing season under irrigated and rainfed scenario by treatment. CP, Common Practices, and EI, Ecological intensification, Scandia KS.

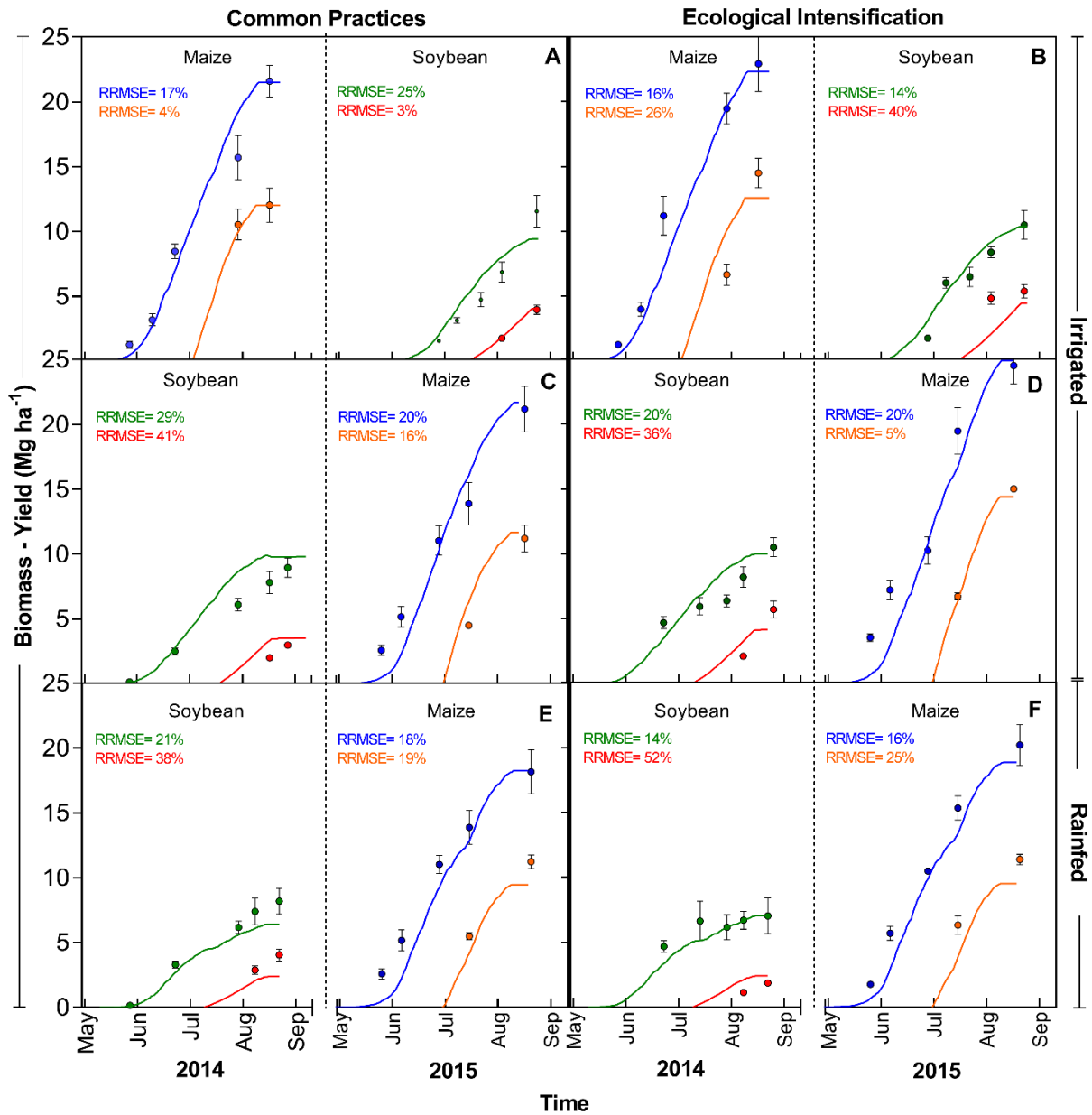


Figure 4.2 APSIM Simulated vs. observed yield (Mg ha^{-1}) and above ground biomass (Mg ha^{-1}) in maize and soybean for Common practices (a, c, e) and ecological intensification (b, d, f) for 2014 and 2015 growing seasons. Blue and orange lines indicate simulated maize biomass and yield; green and red lines indicate simulated soybean biomass and yield. Blue and orange circles indicate maize biomass and yield observed; green and red circles indicate soybean biomass and yield observed. Lines in circles indicate standard error; RRMSE, relative root mean square error. Scandia, KS.

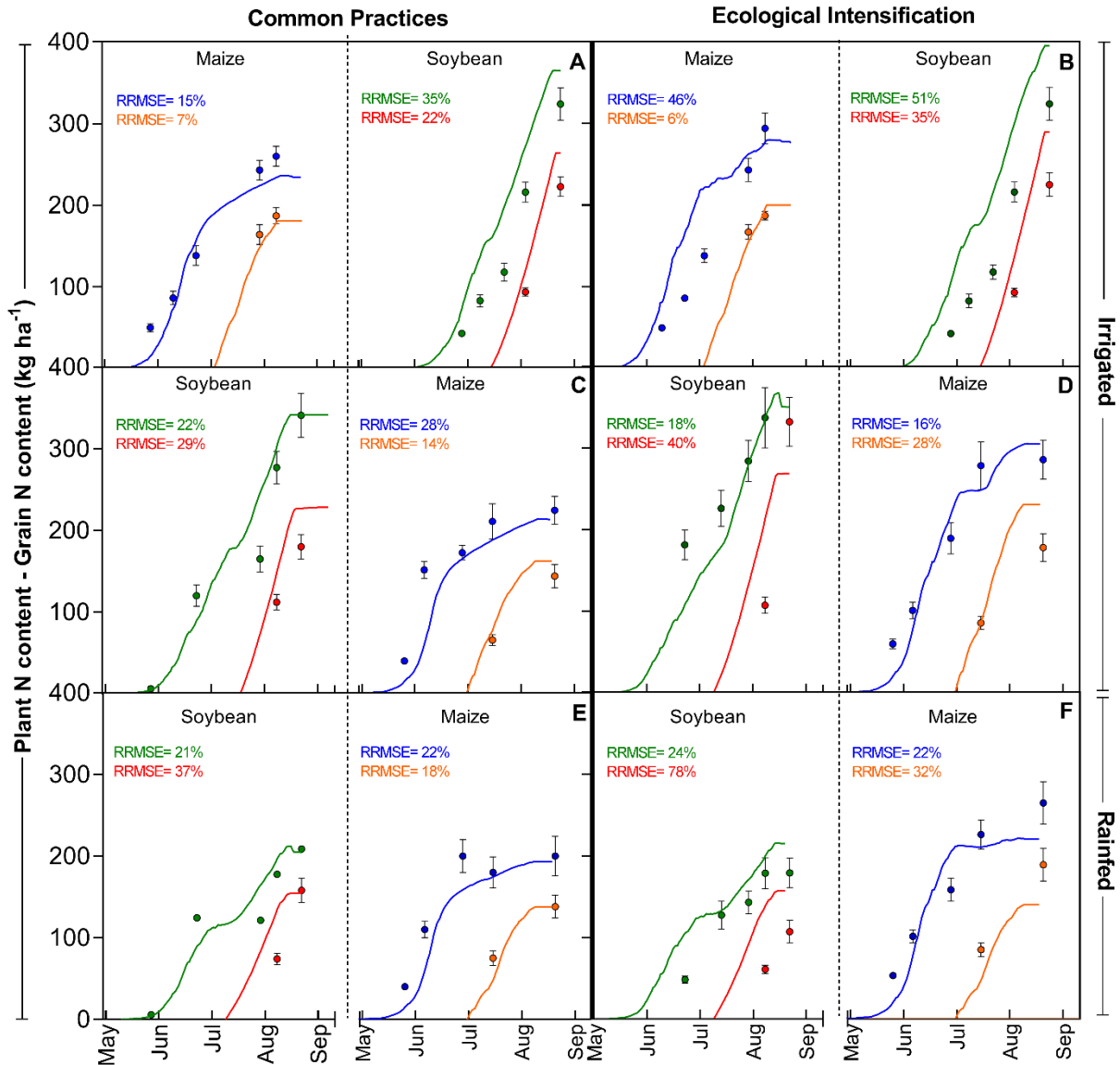


Figure 4.3 APSIM Simulated vs. observed plant N content (kg ha^{-1}) and seed/grain N content (kg ha^{-1}) in maize and soybean for Common practices (a, c, e) and ecological intensification (b, d, f;) for 2014 and 2015 growing seasons. Blue and orange lines indicate maize plant N content and grain N content simulated; green and red lines indicate soybean plant N content and seed N content observed. Blue and orange circles indicate maize plant N content and grain N content observed; green and red circles indicate soybean plant N content and seed N content observed. Lines in circles indicate standard error; RRMSE, relative root mean square error. Scandia, KS.

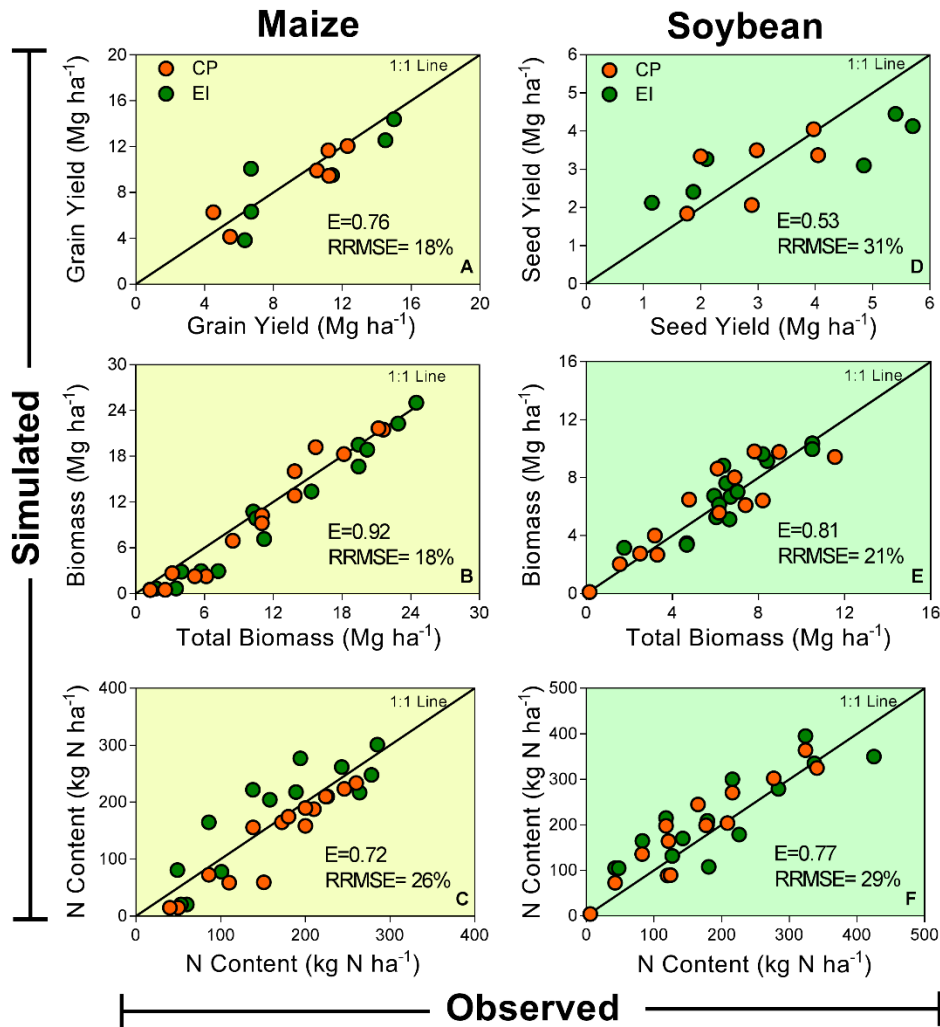


Figure 4.4 APSIM simulated versus observed yield (a, maize; d, soybean); plant biomass (b, maize; e, soybean) and N content (c, maize; f, soybean) combining all rotations. Orange circles, Common Practices (CP); green circles, Ecological Intensification (EI); E, Efficiency of the model; RRMSE, relative root mean square error.

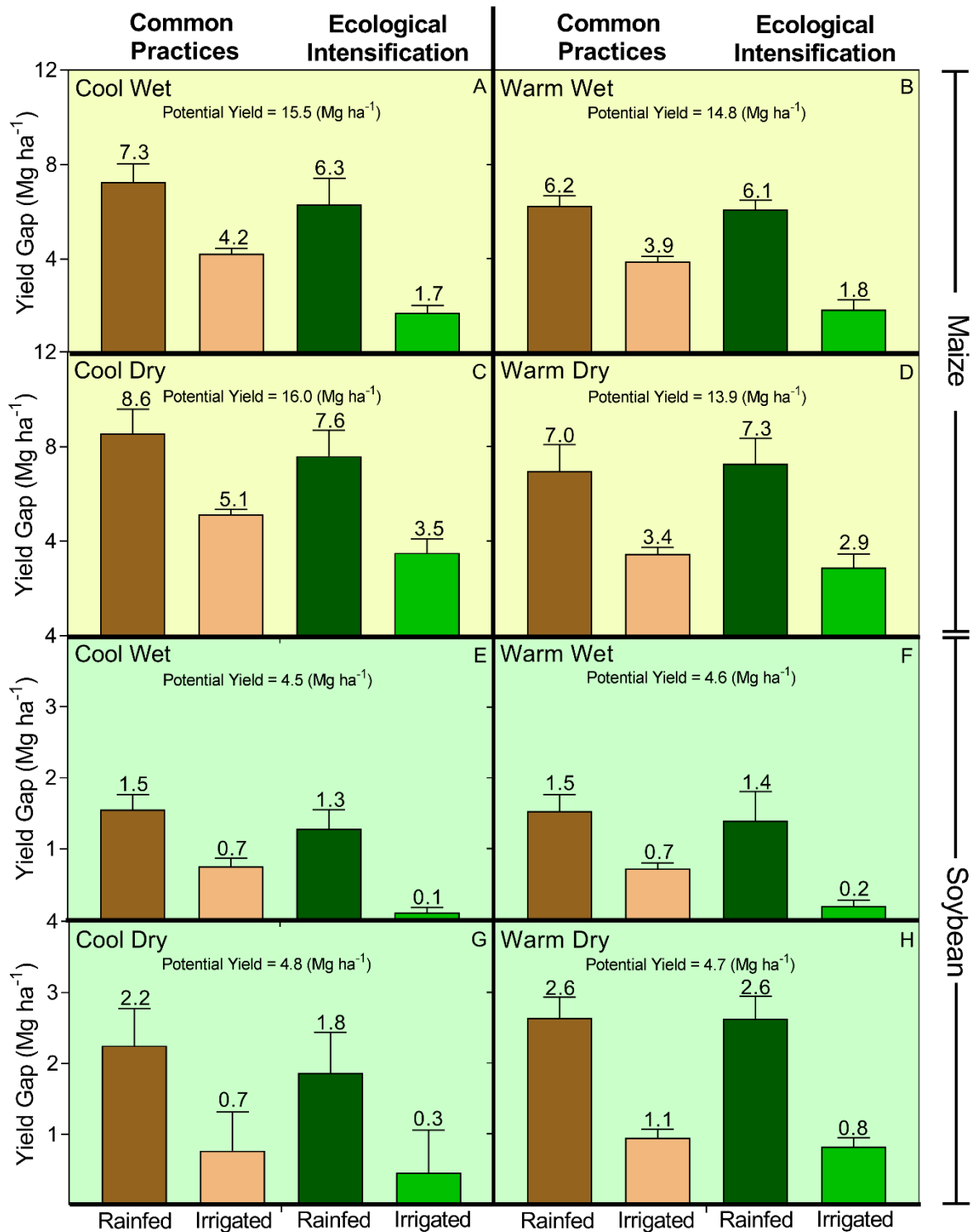


Figure 4.5 Potential yield and yield gap for long-term simulation (1980 – 2016) for Common Practices (lo input) and Ecological Intensification (hi input) under rainfed and irrigated conditions for maize (a, cool and wet; b, warm and wet; c, cool and dry; d, warm and dry) and soybean (e, cool and wet; f, warm and wet; g, cool and dry; h, warm and dry).

Table 4.1 Treatment description for maize and soybean treatments at Scandia KS for 2014 and 2015 growing season.

	Maize		Soybean	
	CP	EI	CP	EI
Seeding rate (pl ha ⁻¹)	74,000	89,000	274,000	429,000
Row spacing (m)	0.76	0.38	0.76	0.38
Fertilization (kg ha ⁻¹)	56N	P-K-S* 56N+112N	No	P-K-S* 56N
Micronutrients	No	1x	No	1x
Fungicide	No	1x	No	1x
Insecticide	No	1x	No	1x

CP. Common Practices, EI. Ecological Intensification *Following university recommendations. Pl: plants. N expressed in kg ha⁻¹. Fertilizer rates added for maize and soybean in kg ha⁻¹: (63-10-35-9) and (63-15-48-12) for rainfed and irrigated scenario. Fungicide Priaxor (BASF) and Headline. Rates for maize 0.28 l ha⁻¹ and 0.42 l ha⁻¹ respectively; for soybean, only Priaxor was applied at the same rate (0.28 l ha⁻¹). Insecticide: for both crops was Fastac CS (BASF), 0.225 l ha⁻¹. Fertilization: For maize: Microessentials SZ (12-40-0-10S-1Zn) Rates for maize: 200 and 264 kg ha⁻¹ for rainfed and irrigated. Rates for soybean: 144 and 200 kg ha⁻¹ for rainfed and irrigated conditions. Aspire (0-0-58-5B). Rates for maize: 90 and 120 kg ha⁻¹. Rates for soybean 195 and 270 kg ha⁻¹. Micronutrients: Librel Zn (0.15Zn) (BASF).

Table 4.2 Observed and simulated dates for flowering and physiological maturity for maize and soybean during 2014 and 2015 growing season. Scandia KS.

Crop	Trt.	Season	Water condition	Flowering			Physiological Maturity		
				Obs.	Sim.	Obs-Sim	Obs.	Sim.	Obs-Sim
Soybean	CP	2014	Rainfed	22/07/2014	15/07/2014	7	9/10/2014	1/10/2014	8
	EI		Irrigated						
	CP	2015	Rainfed	28/07/2015	22/07/2015	6	28/09/2015	23/09/2015	5
	EI		Irrigated						
Maize	CP	2014	Irrigated	11/07/2014	14/07/2014	-3	2/09/2014	4/09/2014	-2
	EI		Irrigated						
	CP	2015	Rainfed	11/07/2015	12/07/2015	-1	8/09/2015	4/09/2015	4
	EI		Irrigated						

Trt., treatment; CP, Common Practices; EI, Ecological Intensification; Obs, Observed date. Sim, Simulated date.

Table 4.3 Statistical analysis for yield, plant biomass, plant N content and seed N content for observed vs. APSIM simulated data for maize and soybean, KS (2014 – 2015).

Trait	Crop	RRMSE	E	R ²
Yield	Maize	18	0.76	0.88
	Soybean	31	0.53	0.75
Plant Biomass	Maize	18	0.92	0.98
	Soybean	21	0.81	0.94
Plant N Content	Maize	26	0.72	0.89
	Soybean	29	0.77	0.99
Seed N Content	Maize	19	0.67	0.89
	Soybean	29	0.43	0.85

RRMSE. Relative Root Mean Square Error. E., Model efficiency. R² coefficient of determination.

Table 4.4 Mean, minimum, and maximum simulated yield and coefficient of variation for maize and soybean for common practices and ecological intensification treatments grouped by weather condition, and the average across all weather for the period 1980 – 2016. Rainfed and irrigated scenarios presented.

Weather	Treatment	Environment	Maize				Soybean			
			Mean	Min	Max	CV	Mean	Min	Max	CV
Cool and Dry	CP	Rainfed	7.5	3.2	10.7	36.1	2.9	0.8	4.0	43.0
		Irrigated	11.0	9.8	11.8	5.2	4.1	3.7	4.5	8.1
	EI	Rainfed	8.4	5.0	11.8	34.6	2.6	0.8	4.0	42.0
		Irrigated	12.6	10.2	14.6	12.5	4.5	3.8	4.9	7.8
Cool and Wet	CP	Rainfed	8.2	4.1	11.2	27.4	2.9	1.7	3.9	22.5
		Irrigated	11.2	9.6	12.1	6.6	3.7	2.9	4.4	10.2
	EI	Rainfed	9.1	4.1	13.6	38.2	3.21	1.7	4.5	26.7
		Irrigated	13.7	12.0	14.9	7.1	4.4	4.1	4.8	4.8
Warm and Dry	CP	Rainfed	7.0	3.5	10.0	35.6	2.1	1.3	2.9	27.1
		Irrigated	10.5	8.8	11.7	8.9	3.6	2.9	4.2	10.9
	EI	Rainfed	6.7	3.4	10.0	36.0	2.0	1.2	3.1	30.9
		Irrigated	11.0	7.3	13.7	19.0	3.9	3.2	4.5	12.0
Warm and Wet	CP	Rainfed	8.5	6.8	10.8	14.0	3.1	2.1	4.5	20.4
		Irrigated	11.0	9.7	12.0	6.5	3.8	3.3	4.3	7.3
	EI	Rainfed	8.0	5.3	10.6	21.0	3.0	2.1	4.0	16.5
		Irrigated	12.9	10.6	15.1	9.3	4.4	3.9	4.8	6.7
Average	CP	Rainfed	7.3	0.57	11.2	40.0	2.5	0.57	4.0	37.0
		Irrigated	10.9	8.8	12.1	8.0	3.8	2.9	4.5	10.0
	EI	Rainfed	7.8	1.8	13.6	41.0	2.6	0.51	4.5	40.0
		Irrigated	12.5	7.3	15.0	15.0	4.2	3.2	4.9	10.0

CP, Common Practices; EI, Ecological intensification; Min, minimum; max, maximum; CV, coefficient of variation.

Chapter 5 - General discussion

Conclusions and implications for agriculture

The historical analysis of soybean yields, biomass and nutrient related traits (Chapter 1) characterized changes that occurred from 1922 to 2015, and summarized available literature that included N, P and K data. The review showed that seed yield improved from 1.3 Mg ha⁻¹ in 1930 to 3.2 Mg ha⁻¹ in 2010, and that that increase was mostly driven by an increase in total biomass production with relatively low variation in HI. The amount of N and P that plants partition to seed increased, while seed nutrient concentration remained stable for N but declined for both P (18%) and K (13%). Nitrogen stover nutrient concentration remained stable, while P declined, and K increased. Nutrient IE increased for N (33%) and P (44%) but decreased for K (11%); and variations in nutrient IEs were primarily explained by changes in nutrient HIs for N and K, and by both PHI and Pseed.

From the four years of field experiments in a maize-soybean rotation, described in Chapter 2, intensified management systems based on seeding rate increase, narrow row spacing, and a balanced nutrition program resulted in soybean yield increase compared to common practices. For soybean, response to intensified practices with a balanced nutrition program was observed in yield environments above 4.5 Mg ha⁻¹. Soybean yields were more variable throughout all growing seasons compared to maize yields. Each unit of fertilizer applied in soybean produced more seed yield under intensification compared with a balanced nutrition program without intensification. For maize, yields show a productive environment with control plots yielding above 10 Mg ha⁻¹ under rainfed conditions and with potential to increase up to 14 Mg ha⁻¹ under irrigation and with intensive management systems. Maize demonstrated again that is a sensitive

crop regarding to nutrition, more specifically to N, since treatments without a balance nutrition program yielded considerably less than those that received balanced nutrition.

Chapter 3 summarized the research conducted to estimate biological N fixation (BNF) in soybeans. Estimations of BNF are critical to account for a N balance of the crop and its impact on the cropping system nutrient balance. The ^{15}N natural abundance is a relatively easy and inexpensive method for quantifying fixation. The method requires the quantification of the ^{15}N abundance of soybean growing in a N free substrate, to determine the B value. In this research, the B value was determined in a greenhouse experiment at different growth stages, plant parts, and for four soybean varieties. Overall, for the entire plant, the B value at the peak of biomass was of -1.97. The mode reported in the literature was of -1.70, with a minimum of -3.86 and a maximum of 0.20. When the B value was utilized to estimate BNF in field-grown soybeans, intensification (high input) significantly increased BNF rates.

The last chapter discussed the modeling approach implemented to simulate two of the five management systems evaluated at the field scale. Field data from the maize-soybean experiment with low and high input management systems was collected and analyzed using the APSIM model. Yield gaps were estimated in relation to weather patterns and management intensification in the Western US Corn Belt. After calibration, a long-term simulation was performed to analyze the yield gap for the low and high input management systems. Across 37-yr of simulations with APSIM determined yield gaps for maize (2.5-7.3 Mg ha⁻¹) and soybean (0.4-2.0 Mg ha⁻¹) under contrasting input levels and water scenarios. The modeled potential yield was more sensitive to weather changes in maize than in soybeans. The high-input system had more yield stability across all weather patterns. The size of the yield gap was reduced by approximately half under irrigation for maize and soybeans with intensified management, from low- to high-input systems.

Overall our results support the use of APSIM as a tool to develop future management strategies to close yield gaps in both rainfed and irrigated maize and soybean systems across different weather patterns in Western Corn belt, USA.

Contribution to science

The present research contributes to the knowledge of the complex interactions between genotype (G), environment (E), and management (M) practices in maize-soybean rotations. The soybean review study presented the first historical overview of yield and nutrient related traits, that advances the understanding of actual soybean characteristics related to yield and biomass production, and N, P, and K related traits. Field experiments did not follow the traditional kitchen sink approach where each management practice is evaluated by addition and subtraction of other management practices. The integrated approach of evaluation of five management systems defined by different combinations of management practices for a maize-soybean rotation and exploring rainfed and irrigated conditions are not reported in the literature. In this research, treatments placement was maintained in the same “plot” for four growing seasons capturing the residual effects of the management systems evaluated.

The research conducted on the B value to estimate BNF produced references on B values for modern varieties by growth stage and plant part. To the best of our knowledge, the data presented in this study is not available for modern soybean varieties. The values presented in this research study will be helpful for other research groups that are aiming to provide an estimate of BNF with the natural abundance technique and using B values from past experiments.

The modeling approach proposed in this research allowed for the extraction of more information from the data collected in the field studies. By combining field datasets with long term weather records, and simulations through computational modeling, we generated a variety of

possible scenarios as result of interaction of G x E x M that allowed us to respond to our objective with more certainty. Models are a powerful tool if we supplied them with quality data and if we calibrate them to represent the real crop rotations with accuracy. The modeling approach is novel too since the focus is not only on one crop, but rather on a maize-soybean rotation with two contrasting management systems, and through 37 years of weather data. This kind of approach can be followed by other researchers with datasets from field studies to add value to that information.

Future research

After conducting this research, new questions arose when addressing the objectives of each chapter. In the case of the historical review, it will be interesting to focus in the future on plant nutrient ratios and their relations to crop growth rate. This will likely result in improved tools for nutrient management. The characterization for crops based on single nutrients has value in establishing basic nutrient physiological responses and dynamics, but research efforts should start focusing on nutrient ratios and how those ratios affect parameters such as crop growth rate.

Further research should be focused on evaluating the impact of management systems to find better strategies to increase productivity in a sustainable manner. In this research cover crops were not included in the rotation, but it would be good to include them to have more crops per year and to evaluate their performance in combination with the five management systems.

The work done to establish the B value and to determine the BNF for the soybean crop in the rotation can be complemented with a global N balance for the soybean rotation for both the high and low input management systems evaluated. This will bring light to the question of whether or not soybeans produce a positive or a negative N balance in cropping system. For the B value estimation, it will be interesting to conduct new greenhouse studies evaluating different maturity

groups and more than one inoculation strain. This will generate more data, especially for use in BNF estimations across all soybean production regions. Future research should also focus on more accurately quantifying N fixation for high-yielding soybean systems ($> 7 \text{ Mg ha}^{-1}$) for improving N budget estimations, since those systems require considerable amounts of N, and in most cases, it is assumed that the balance is neutral or positive, but there are not enough studies that accurately measure this.

Finally, a complete economic analysis quantifying the impact of each management system and the profit of each rotation combined with irrigation or rainfed scenario will provide support to stake holders, agronomist and farmers when evaluating how to increase productivity to meet the growing global food demand.

Appendix A - Figure and tables Chapter 1

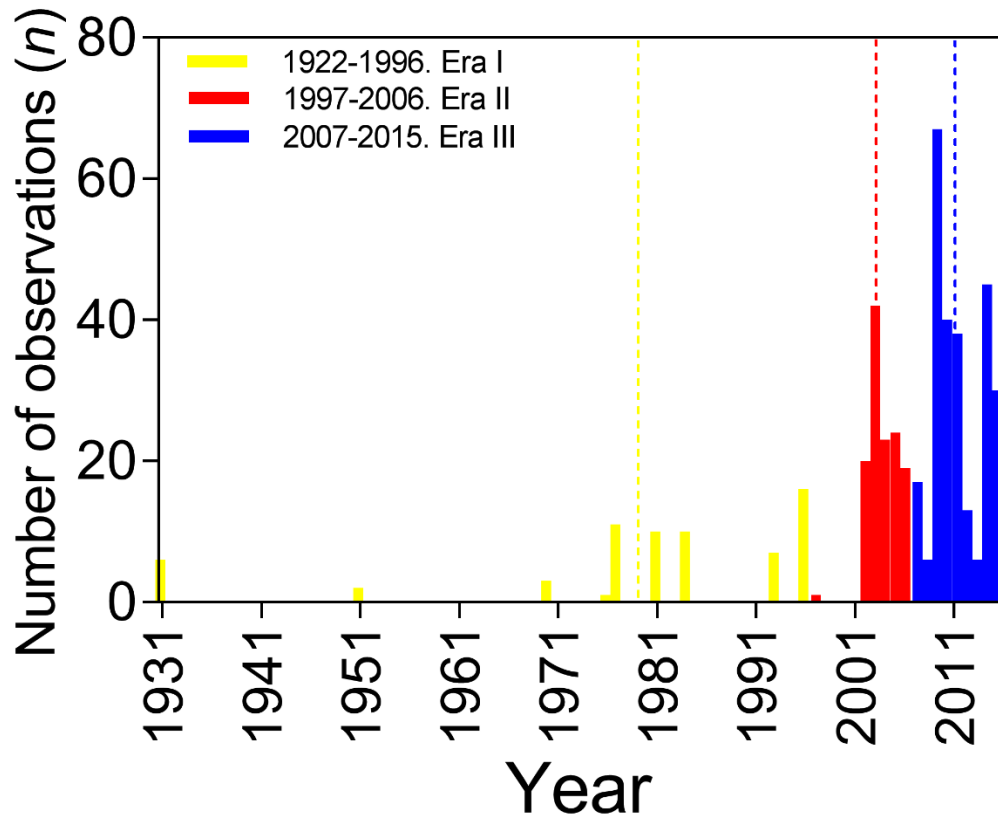


Figure A.1 (A) Soybean seed yield; (B) biomass; (C) harvest index; (D, I, and N) seed N, P, and K concentration; (E, J, and O) stover N, P, and K concentration; (F, K, and P) plant N, P, and K nutrient uptake; (G, L, and K) N, P, and K harvest index; and (H, M, R) nutrient internal efficiency across years for the pool data ($n = 322$). Solid lines represent linear regression. Green represents positive slope ($***p < 0.01$), red represents negative slope ($***p < 0.001$), and blue a slope not different from zero. NHI, N harvest index; NIE, N internal efficiency; KHI, K harvest index; KIE, K internal efficiency; PHI, P harvest index; PIE, P internal efficiency.

Table A.1: Linear function adjusted, R^2 , Slope, F value and P value for slope analysis of the relations presented in Figure 1.

Relation	Function	R^2	Slope analysis		
			Slope	F value	P value
Seed yield vs. Time	$Y=0.025*X-48$	0.09	0.025	30.85	0.0001
Total biomass vs Time	$Y=0.058*X-109$	0.06	0.058	20.96	0.0001
HI vs. Time	$Y=0.00077*X-1.14$	0.016	0.00077	5.07	0.0249
N_{seed} vs. Time	$Y=-0.0046*X+9.06$	0.0008	-0.0016	0.26	0.604
P_{seed} vs. Time	$Y=-0.0027*X+5.99$	0.11	-0.0027	39.9	0.0001
K_{seed} vs. Time	$Y=0.00044*X+0.9$	0.0002	0.0004	0.06	0.81
N_{stover} vs. Time	$Y=-0.0058*X+13.16$	0.0088	-0.0058	2.83	0.09
P_{stover} vs Time	$Y=-0.0012*X+2.86$	0.025	-0.0013	8.25	0.0043
K_{stover} vs. Time	$Y=0.0095*X-17.67$	0.055	0.0095	18.65	0.0001
Plant N Uptake vs. Time	$Y=1.44*X-2656$	0.038	1.44	12.48	0.0005
Plant P Uptake vs. Time	$Y=0.035*X-48.57$	0.003	0.035	1.06	0.30
Plant K Uptake vs. Time	$Y=1.229*X-2347$	0.081	1.23	28.21	0.0001
NHI vs. Time	$Y=0.0014*X-2.09$	0.0256	0.0014	8.29	0.0043
PHI vs. Time	$Y=0.0015*X-2.25$	0.019	0.0015	6.18	0.013
KHI vs. Time	$Y=-0.0017*X+4.03$	0.036	-0.002	11.92	0.0006
NIE vs. Time	$Y=0.027*X-41.3$	0.021	0.027	6.94	0.0088
PIE vs. Time	$Y=0.76*X-1408$	0.09	0.77	31.38	0.0001
KIE vs. Time	$Y=-0.085*X+197.9$	0.016	-0.08	5.23	0.023

nitrogen, N; phosphorus, P; potassium, K; harvest index, HI; N harvest index, NHI; P harvest index, PHI; K harvest index KHI; seed N concentration, N_{seed} ; stover N concentration, N_{stover} ; seed P concentration, P_{seed} ; stover P concentration, P_{stover} ; seed K concentration, K_{seed} ; stover K concentration, K_{stover} ; N internal efficiency, NIE; P internal efficiency, PIE; K internal efficiency, KIE; N to P ratio, N:P; N to K ratio, N:K; K to P ratio, K:P.

Table A.2: Descriptive statistics (Minimum, 25% Quartile, 75% Quartile, Mean, Maximum and Coefficient of Variation for seed yield, total biomass, harvest index; N, P and K biomass uptake, seed uptake, harvest index, seed content, stover content and internal efficiency; and N:P, N:K and P:K ratio.

Variable	Unit	1922-1996						1997-2006						2007-2015					
		Min	25%Q	Mean	75%Q	Max	CV	Min	25%Q	Mean	75%Q	Max	CV	Min	25%Q	Mean	75%Q	Max	CV
Seed Yield	Mg ha ⁻¹	0.75	1.35	2.25	3.10	5.99	0.49	0.71	1.55	1.99	2.48	6.77	0.41	0.81	2.07	3.03	3.91	7.88	0.44
Total Biomass	Mg ha ⁻¹	1.90	3.39	5.79	7.81	13.88	0.43	1.59	3.31	5.88	7.56	18.59	0.47	1.67	5.27	7.67	9.99	20.73	0.47
Harvest Index	dimensionless	0.16	0.35	0.39	0.46	0.62	0.26	0.2	0.29	0.38	0.48	0.62	0.26	0.19	0.37	0.41	0.47	0.88	0.22
Plant N Uptake	kg ha ⁻¹	67	126	235	366	465	0.54	38	103	156	196	614	0.44	53	162	238	310	549	0.44
Plant P Uptake	kg ha ⁻¹	5	12	22	33	50	0.54	6	12	20	22	58	0.61	5	16	22	28	47	0.39
Plant K Uptake	kg ha ⁻¹	20	43	90	118	272	0.61	14	42	75	104	359	0.62	18	79	122	149	336	0.54
N Seed Uptake	kg ha ⁻¹	32	100	152	197	412	0.44	27	72	111	145	226	0.43	36	133	187	229	501	0.42
P Seed Uptake	kg ha ⁻¹	7	11	16	22	34	0.43	3	8	11	14	25	0.37	3	11	16	20	44	0.39
K Seed Uptake	kg ha ⁻¹	16	30	52	69	146	0.53	9	28	38	47	81	0.37	8	33	55	68	165	0.55
NHI	dimensionless	0.3	0.47	0.61	0.8	0.87	0.31	0.62	0.71	0.78	0.86	0.9	0.10	0.44	0.67	0.73	0.81	0.91	0.15
PHI	dimensionless	0.25	0.56	0.63	0.72	0.86	0.26	0.27	0.57	0.68	0.87	0.89	0.28	0.38	0.63	0.72	0.82	0.9	0.18
KHI	dimensionless	0.17	0.35	0.49	0.61	0.76	0.36	0.19	0.45	0.48	0.54	0.77	0.26	0.21	0.34	0.44	0.51	0.8	0.27
N:P	dimensionless	3.0	9.3	11	12.6	18.0	0.28	3.9	5.4	9.4	11.3	22.7	0.47	3.8	8.4	10.9	12.6	22.5	0.32
N:K	dimensionless	0.87	2.1	2.9	3.4	5.4	0.31	1.0	1.6	2.5	3.7	4.7	0.48	1.1	1.7	2.1	2.3	6.4	0.31
P:K	dimensionless	0.13	0.2	0.3	0.3	0.55	0.32	0.1	0.2	0.3	0.3	0.8	0.51	0.1	0.1	0.21	0.3	0.6	0.41
Nseed	g 100 g ⁻¹	4.28	5.63	5.94	6.19	7.60	0.12	3.01	4.57	5.36	5.97	6.75	0.17	3.83	5.44	5.84	6.29	7.47	0.10
Pseed	g 100 g ⁻¹	0.35	0.55	0.65	0.74	0.92	0.21	0.30	0.49	0.57	0.65	0.73	0.18	0.32	0.45	0.53	0.59	0.85	0.21
Kseed	g 100 g ⁻¹	1.53	1.80	1.98	2.15	2.59	0.13	0.43	1.70	1.89	2.03	2.55	0.20	0.64	1.30	1.72	2.14	2.65	0.29
Nstover	g 100 g ⁻¹	0.3	0.78	1.48	1.68	3.63	0.69	0.11	0.79	1.30	1.84	2.70	0.45	0.53	0.95	1.38	1.67	4.74	0.46
Pstover	g 100 g ⁻¹	0.02	0.12	0.22	0.27	0.72	0.74	0.04	0.09	0.28	0.41	0.64	0.58	0.03	0.08	0.14	0.21	0.31	0.54
Kstover	g 100 g ⁻¹	0.11	0.67	1.16	1.27	4.07	0.78	0.12	1.38	1.71	2.04	4.77	0.38	0.27	1.18	1.48	1.67	4.36	0.31
NIE	kg kg ⁻¹	5	8	10	12	18	0.31	10	13	15	16	24	0.18	7	12	13	14	25	0.17
PIE	kg kg ⁻¹	31	83	100	128	192	0.39	44	100	115	136	175	0.29	76	114	139	164	207	0.23
KIE	kg kg ⁻¹	8	25	28	33	46	0.37	11	20	25	30	51	0.30	10	21	27	32	77	0.37

nitrogen, N; phosphorus, P; potassium, K; harvest index, HI, N harvest index, NHI; P harvest index, PHI; K harvest index KHI; seed N concentration, N_{seed}; stover N concentration, N_{stover}; seed P concentration, P_{seed}; stover P concentration, P_{stover}; seed K concentration, K_{seed}; stover K concentration, K_{stover}; N internal efficiency, NIE; P internal efficiency, PIE; K internal efficiency, KIE; N to P ratio, N:P; N to K ratio, N:K; K to P ratio, K:P.

Appendix B - Figure and tables Chapter 4

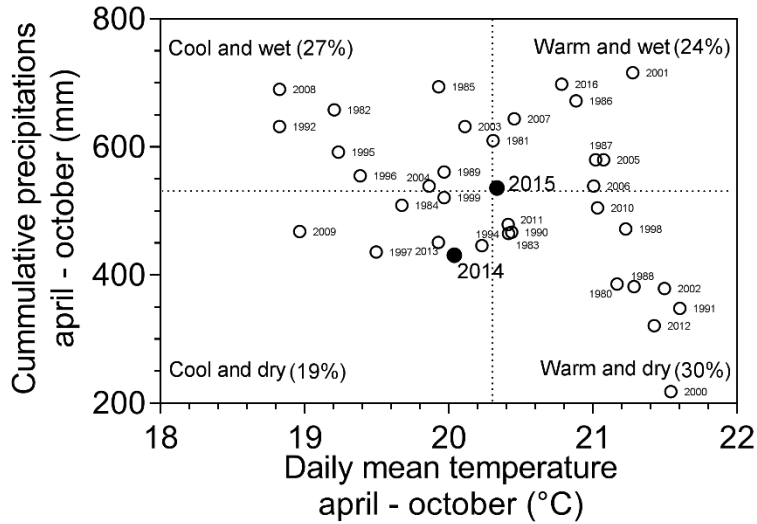


Figure B.1 Yearly (1980 – 2016) mean temperature and mean precipitation for the period April – October for the years. Black circles indicate seasons where experimental data was collected. Empty circles indicate years where experimental data was simulated. Dotted vertical and horizontal line indicates mean temperature (°C) and mean cumulative precipitation (mm) for the period. In parenthesis percentage of years in each category. Point for the year 1993 not shown (Temperature 18.9 °C – precipitation 967 mm).

Table B.1. Soil values from the initialization period (2005-2015). Values refers to the start of the simulation on 01/01/2005. OC, soil organic carbon; BD, bulk density; Fbiom, microbial SOC (fast decomposition); Finert, inert of soil organic carbon (not decomposing); Hum, humic SOC (medium decomposing); LL, lower limit; DUL, drained upper limit; SAT, saturated volumetric water content; SW, soil water; Maize and Soybean KL, parameters defining capacity to extract water per day; NO3, soil nitrates.

Soil layer cm	OC g 100g ⁻¹	BD Mg m ⁻³	Fbiom	Finert kg C ha ⁻¹	Hum	LL -----mm mm ⁻¹ -----	DUL	SAT	SW	Maize KL d ⁻¹	Soybean KL d ⁻¹	NO3 mg kg ⁻¹
0-5	1.74	1.37	590	4768	6560	0.11	0.265	0.459	0.338	0.08	0.08	7.5
5-15	1.74	1.37	957	10107	12772	0.11	0.285	0.459	0.301	0.08	0.08	12
15-25	1.45	1.38	475	10885	8648	0.158	0.295	0.455	0.251	0.07	0.07	1.5
25-38	1.45	1.38	244	19666	6103	0.158	0.3	0.455	0.251	0.06	0.06	2.5
38-63	0.87	1.33	113	23142	5672	0.182	0.32	0.473	0.204	0.05	0.05	2.1
63-83	0.47	1.38	31	10824	2116	0.192	0.29	0.455	0.195	0.04	0.04	2
83-101	0.29	1.34	6	6575	413	0.16	0.29	0.47	0.202	0.03	0.03	0.829
101-135	0.17	1.33	0.76	7611	76	0.15	0.29	0.473	0.205	0.02	0.02	0.221
135-175	0.17	1.33	0.89	8954	89	0.15	0.29	0.473	0.205	0.02	0.02	0
175-200	0.17	1.33	0.56	5596	55	0.15	0.29	0.473	0.2	0.00	0.00	0

Table B.2. APSIM maize and soybean cultivar and crop model specific parameter values used in this study. When more than one value is given (see soybean), this means that there is an array of values for the specific parameter.

Acronym	Value	Unit
Maize		
tt_emerg_to_endjuv (thermal time from emergence to end juvenile)	250	°C-days
tt_flower_to_maturity (thermal time from flowering to phys maturity)	812	°C-days
head_grain_no (potential kernel number per ear)	800	#
grain_gth_rate (grain growth rate)	9.17	mg/rain/day
tt_flower_to_start_grain (thermal time from flowering to start grain fill)	170	°C-days
tt_maturity_to_ripe (thermal time from maturity to harvest)	150	°C-days
Soybean		
x_pp_hi_incr (photoperiod)	1, 24	Hours
y_hi_incr (daily rate of harvest index)	0.01, 0.01	1/days
x_hi_max_pot_stress (stress index)	0.0, 1.0	(-)
y_hi_max_pot (maximum value for harvest index)	0.5, 0.5	(-)
tt_emergence (thermal time to emergence)	100, 100	°C-days
x_pp (photoperiod levels)	13.59, 14.6, 15.6, 16.6	Hour
y_tt_end_of_juvenile ¹ (thermal time to juvenile)	100, 133, 200, 400	°C-days
y_tt_floral_initiation (thermal time from end of juv to floral initiation)	128, 171, 256, 512	°C-days
y_tt_flowering (thermal time from flowering to start grain fill)	246, 328, 492, 1312	°C-days
y_tt_start_grain_fill (thermal time from start to end of grain fill)	499, 666, 999, 2664	°C-days
tt_end_grain_fill (thermal time from end grain fill to maturity)	20	°C-days
tt_maturity (thermal time from maturity to harvest)	70	°C-days
node_sen_rate (node senescence rate)	95	°C-days node ⁻¹
Twilight (twilight)	0	(-)
x_stage for N fixation (crop stage number)	3, 4, 5, 6, 7	stage #
N_fix_rate (Nn fixation rate)	0.0006, 0.0016, 0.0016, 0.0009	gN/gDM
x_stage for N concentration (crop stage number)	3, 6, 9	stage #
y_n_conc_min_leaf (minimum N concentration in leaves)	0.02, 0.01, 0.0085	gN/gDM
y_n_conc_crit_leaf (critical N concentration in leaves)	0.06, 0.05, 0.02	gN/gDM
y_n_conc_max_leaf (maximum N concentration in leaves)	0.06, 0.05, 0.025	gN/gDM
y_n_conc_crit_stem (critical N concentration in stems)	0.03, 0.02, 0.008	gN/gDM
y_n_conc_max_stem (minimum N concentration in stems)	0.03, 0.02, 0.008	gN/gDM
y_n_conc_crit_pod (critical N concentration in pods)	0.06, 0.06, 0.005	gN/gDM
y_n_conc_max_pod (maximum N concentration in pod)	0.06, 0.06, 0.008	gN/gDM

Appendix C - License Agreement Chapter 1

Balboa, G. R., Sadras, V. O., and Ciampitti, I. A. (2018). Shifts in soybean yield, nutrient uptake, and nutrient stoichiometry: A historical synthesis-analysis. *Crop Sci.* 58.

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