

# The Reduction of Plant-Available Nitrogen by Cover Crop Mulches and Subsequent Effects on Soybean Performance and Weed Interference

M. S. Wells, S. C. Reberg-Horton,\* A. N. Smith, and J. M. Grossman

## ABSTRACT

A 3 site-year study was conducted to investigate the impact of roller-crimped rye (RC) (*Secale cereale* L.) mulches on soil N immobilization and subsequent effects on weed suppression and soybean [*Glycine max* (L.) Merr.] yield. Treatments consisted of: (i) RC, (ii) conventional tillage with neither rye cover crop nor weed control measures (WC), (iii) conventional tillage plus herbicide weed control (CT+HB), and, (iv) roller-crimped rye plus herbicide (RC+HB). The rye biomass varied between the sites with 4400, 8300, and 7084 kg ha<sup>-1</sup> dry matter (DM) for Goldsboro 2009, Kinston 2009, and Kinston 2010, respectively. During the season, the flow of soil inorganic N was monitored via ion-exchange probes and by direct extractions at two depths (0–10 and 10–25 cm) every 2 wk. Tissue data was collected every 2 wk on soybean and redroot pigweed (*Amaranthus retroflexus* L.) to determine the C/N ratio. For all sites, peak N immobilization occurred between 4 and 6 weeks after planting (WAP), indicated by a reduction in soil inorganic N. Results from the ion-exchange probes showed similar trends of the extractable soil inorganic N at all sites. Pigweed C/N ratios revealed a growing divergence between the two systems, with a severe N deficiency in the RC. Even with varying rye biomass production across environments the RC system created an extremely low N environment, suggesting that when a cereal cover crop is paired with a legume cash crop, reduced weed crop interference may result, with little reduction in soybean yield.

**A**POTENTIAL WEED MANAGEMENT practice, in the early stages of adoption by organic soybean producers, is the use of roll-killed cover crops. This system uses a roller-crimper implement immediately before planting of a cash crop to kill the mature cover crop, thereby producing a weed-suppressive mulch (Davis, 2010; Mirsky et al., 2012, 2013; Reberg-Horton et al., 2012). Cover crop mulches suppress weeds by physical and chemical mechanisms. Physical impedance and light deprivation are major means of controlling weed emergence (Teasdale and Mohler, 2000) with varying estimates of the importance of allelopathic chemicals from rye shoots (Kruidhof et al., 2009; Putnam and Defrank, 1983; Reberg-Horton et al., 2005). Both physical and chemical weed control mechanisms are dependent on cover crop biomass. Smith et al. (2011) reported excellent weed control in organic soybean when roll-killed rye biomass levels reached 9000 kg ha<sup>-1</sup> DM. The level of weed suppression is dependent on the quantity of rye mulch, with an exponential relationship between mulch mass and weed emergence (Teasdale and Mohler, 2000).

One factor that has not been well studied in the system is how the resulting changes in N availability affect the relative competitive ability of weeds and crop plants. The impacts of incorporated residues on N-mediated crop–weed interactions

have been well characterized, with high C/N residues causing rapid N immobilization that affects both crops and weeds (Burgess et al., 2002; Jin et al., 2008; Recous et al., 1995; Schomberg et al., 1994a). Less is known about the effect of surface residues on N immobilization in agricultural systems and their impacts on N cycles.

Surface residues have been better characterized in forestry systems. Hart and Firestone (1991) hypothesized that N transfers from soil to decomposing surface litter increase in ecosystems that have accumulations of high C/N ratio litter on the soil surface. Nitrogen transfer from soil to surface litter could account for the entire N immobilized in this layer during the first year of decomposition (Hart and Firestone, 1991). To further support the significance of surface residues on N-immobilization, Jaeger et al. (1999) showed unchanged, net microbial N immobilization throughout the summer in the forest surface soil, and then rapid increases in September after the forest leaf canopy senescence. An analogous N-immobilization effect was demonstrated when leafy material from *Theobroma grandiflorum* [(Wild. ex Spring) Schumann] and winter wheat (*Triticum aestivum* L.) straw, both of which have high C/N ratio litters (43:1 and 80:1, respectively), were applied to soil surfaces (Holland and Coleman, 1987; Schwendener et al., 2005). Similar N-immobilization effects should be expected in agricultural systems with high C/N surface residues.

The reduction of surface soil inorganic-N could be a valuable weed management strategy when a cereal cover crop is paired with a legume cash crop such as soybean. By inducing a low-N environment through the use of high C/N mulch, weeds may be less able to compete for necessary N reserves, thereby

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Published in Agron. J. 105:539–545 (2013)  
doi:10.2134/agronj2012.0396

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**Abbreviations:** CT, conventional tillage; DM, dry matter; HB, herbicide; PRS, plant root simulator; RC, roller-crimped rye; WAP, weeks after planting; WC, weed control.

altering weed-crop competition in favor of crop growth. There are several studies demonstrating the effect of N dose on weed response. The competitiveness of high N-responsive species such as pigweed progressively improves as N rate increases (Blackshaw, 2004). Increasing levels of nutrient availability strongly influence the reproductive performance of sicklepod (*Senna obtusifolia*), a non-N<sub>2</sub>-fixing legume, and lead to increased leaf area and plant biomass (Tungate et al., 2002).

The effect of N on pigweed is of particular interest in the roller-crimper system. Organic farmers have led the adoption of this system (Rodale Institute, 2012) and pigweed (*Amaranthus* sp.) is a primary weed on many organic farms in the south-eastern United States (S.C. Reberg-Horton, personal communication, 2012; Ulloa et al., 2010). The observed low weed competition in roller-crimped soybean (Smith et al., 2011) produced in high rye biomass environments, could be due to N immobilization driven by the rye mulch residues. Most authors have presumed that allelopathy (Reberg-Horton et al., 2005) or soil shading (Teasdale and Mohler, 2000) explain the low weed incidence. If, however, N immobilization is playing a significant role, the system could be fine tuned to maximize weed control through N deprivation. The objective of this study was to determine the magnitude of N immobilization in the roller-crimper system and how that impacts pigweed/soybean competitive outcomes.

## MATERIALS AND METHODS

In 2009 and 2010, a field experiment was performed at the Center for Environmental Farming Systems (CEFS) in Goldsboro, NC (35.38291° N, -78.035846° W) and Caswell Research Farm in Kinston, NC (35.273206° N, -77.623816° W). The Goldsboro location was certified organic, and the Kinston locations (2009 and 2010) were managed mostly organic with two exceptions. The weed-free checks received herbicides and the rye cover crop received urea ammonium nitrate (UAN) applications due to the lack of availability of manure compost. Locations were selected from the North Carolina Department of Agriculture Research Stations that were representative in climate, proximity, and soil type to eastern North Carolina soybean producers. The soil type in Goldsboro for 2009 was Wickham loamy sand (fine-loamy, mixed, semiactive, thermic Typic Hapludults) with 2 to 6% slope, and the soil types in Kinston for years 2009 and 2010 were Johns loamy sand (fine-loamy over sandy or sandy-skeletal, siliceous, semiactive, thermic Aquic Hapludults) with 0 to 2% slope and Kenansville loamy sand (loamy, siliceous, subactive, thermic Arenic Hapludults) with 0 to 3% slope.

The experiment was a randomized complete block design with six replicates. Treatments consisted of: (i) RC, (ii) conventional tillage with neither rye cover crop nor weed control measures, referred to as weedy check (WC), (iii) conventional tillage plus herbicide (both pre- and post-emergent) for weed control (CT+HB) (i.e., weed free), and, at 1 site-year, (iv) RC+HB (both pre- and post-emergent) (i.e., weed free).

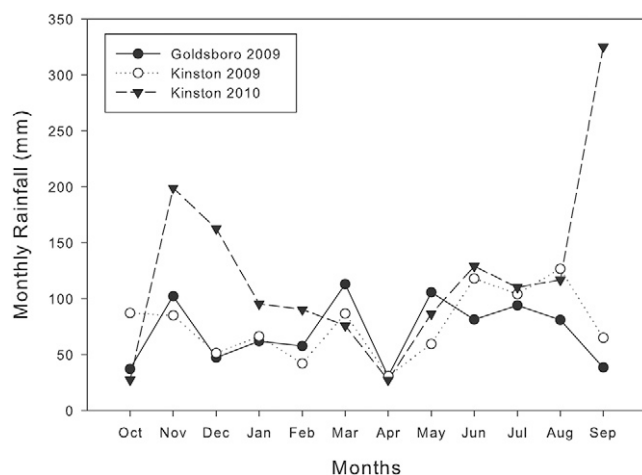
The entire field, regardless of treatments, at all locations was disked and field cultivated to remove any existing vegetation from the prior maize crop and lime, P, and K were applied according to soil tests before rye planting. Approximately 30 kg N ha<sup>-1</sup> was applied to the entire study (including

CT+HB and WC treatments) as UAN before fall seeding to ensure that adequate N was available for early growth of the rye cover crop. In addition, the entire study received UAN at 56 kg N ha<sup>-1</sup> at Kinston, and composted turkey (*Meleagris gallopavo*) manure with an available N equivalent of 56 kg N ha<sup>-1</sup> at Goldsboro were top-dressed before jointing in the spring.

The plot size at both locations was four rows wide (76-cm row spacing) by 15-m long. Before planting in the fall (mid-October), all fields were disked with a tandem offset disk and cultivated. Rye (cultivar Rymin) was subsequently sown to all treatments (including the CT+HB and WC) via grain drill with 13-cm spacing between rows as a cover crop at a rate of 134 kg ha<sup>-1</sup> (approximately 2 bushels per acre). Rye was terminated for both CT+HB and WC treatments early spring via light tillage (Goldsboro 2009) and glyphosate at Kinston 2009 and 2010. In mid-May, the rye cover crop was roll-killed (i.e., terminated) (Feeke's growth stage 11 or Zadoks growth stage 85) with a 3.1 m roller-crimper (I&J Manufacturing, Gap, PA). Immediately after the rye cover crops were roll-killed, soybean (cultivar Hutcheson), Maturity Group V, were planted with a no-till planter (Monosem, Edwardsville, KS) parallel to the roller-crimper direction at 370,500 live seed ha<sup>-1</sup>. Weed control methods varied between the two locations. At Goldsboro, CT+HB (i.e., weed-free) plots were maintained weed-free via clove oil applied 6 WAP as directed with an under-canopy in-row spray at 18.7 L a.i. ha<sup>-1</sup> (10% concentration). At Kinston, CT+HB plots were kept weed-free with s-metolachlor applied at planting for pre-emergent weed control at a rate of 1.9 kg a.i. ha<sup>-1</sup> and post-emergent weed control was achieved with imazethapyr applied 3 WAP post-emergent at a rate of 74.7 g a.i. ha<sup>-1</sup>. Hand weeding was done as needed to ensure weed-free conditions at all locations.

Crop parameters collected included rye biomass, soybean stand count, weed densities, soybean and pigweed tissue percent N and C, and soybean yield. Rye biomass was collected from all plots on a 0.5-m<sup>2</sup> quadrats, dried at 60°C for 72 h, and dry weights were recorded. Stand counts were taken twice during the season at 4 and 6 WAP on 1 m of soybean row. Weed density, was collected by species with the exception of *Amaranthus* which were not identified beyond genus during late summer on all plots by counting above-canopy weeds in the two middle soybean rows (20.9 m<sup>2</sup>). Soybean and pigweed tissues (aboveground plant biomass) of uniform growth stage and size were collected at 2-wk intervals after planting in each plot in a randomly selected 1-m row of soybean, or within a 0.5-m<sup>2</sup> area basis for pigweed. Since pigweed maturity and species composition could greatly effect plant tissue C/N ratios, visual inspection ensured both that tissues collected were from redroot pigweed and of similar growth stages. Plant samples were separated into component species, dried at 65°C, weighed, ground, and analyzed for total N and C by a PerkinElmer (Norwalk, CT) Model 2400 CHN elemental analyzer. Before the soybean harvest in mid-October, potential edge effect was minimized by end trimming the plots. Using a small plot combine, soybean yield was harvested from 12.2 m of the two center rows in each plot.

Soil parameters recorded were soil-extractable inorganic N, plant available N, soil moisture (used only for the calculation of extractable soil N kg ha<sup>-1</sup>) and monthly precipitation (Fig. 1).



**Fig. 1. Monthly precipitation (mm) rainfall for Goldsboro and Kinston, NC (2009–2010). Weather data provided by the State Climate Office of North Carolina.**

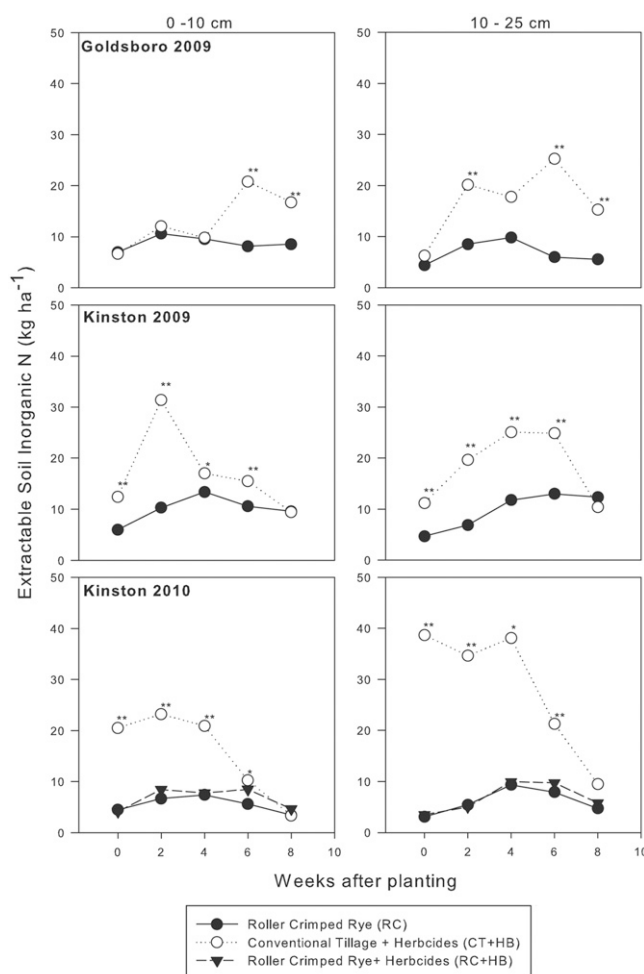
Twenty soil cores were collected, every 2-wk after planting, using a tee-handled soil push probe from two depths, “shallow” (0–10 cm) and “deep” (10–25 cm). Soil samples were air-dried, shaken with 1 M KCl (8 g soil in 50 mL) for 1 h, filtered, and analyzed for total inorganic N on Quick Chem 8000 Lachat (Keeney and Nelson, 1982). Plant available N was measured via plant root simulator (PRS) probes (WesternAG Innovations, Saskatoon, SK). The PRS probes were installed at soybean planting and exchanged every 2-wk starting 2 WAP for 8 wk following recommended protocol (Qian and Schoenau, 2002). To prevent plant root competition for available N, PRS probes were installed vertically into 10-cm diam., 15-cm deep root exclusion cylinders made of PVC pipe.

A combined analysis was attempted on the three trials, but significant treatment by site-year interactions prevented pooling over the site-year (Steel et al., 1996). Each site was analyzed separately using proc MIXED with time treated as a repeated measure. The correction to the variance structure was chosen based on consideration of several different variance models using the Akaike Information Criterion (Freund and Littell, 2000). Within each site-year, treatments were fixed and blocks were random effects and means were separated using pre-planned contrasts (SAS Institute, 2006; Steel et al., 1996). Weed densities were log transformed for analysis and means were back transformed for presentation.

## RESULTS AND DISCUSSION

### Soil Inorganic Nitrogen and Plant-Available Nitrogen as Affected by Rye Residue

Rye biomass dry matter production varied among the 3 site-years with 4450, 8367, and 7084 kg ha<sup>-1</sup> DM at Goldsboro and Kinston (2009), and Kinston (2010), respectively. At roll-killing and through 6 WAP, the extractable soil inorganic N and plant available N levels (probe N) were significantly lower among RC and RC+HB when compared to CT+HB plots at Kinston (2009 and 2010) (Fig. 2). There was no detectable difference in either soil inorganic N or plant available N between RC and RC+HB (Fig. 2 and 3). In Kinston, at the shallow sampling depth, initial soil inorganic N concentrations for CT+HB and RC were 12.4 and 6.0 kg ha<sup>-1</sup> N in 2009,

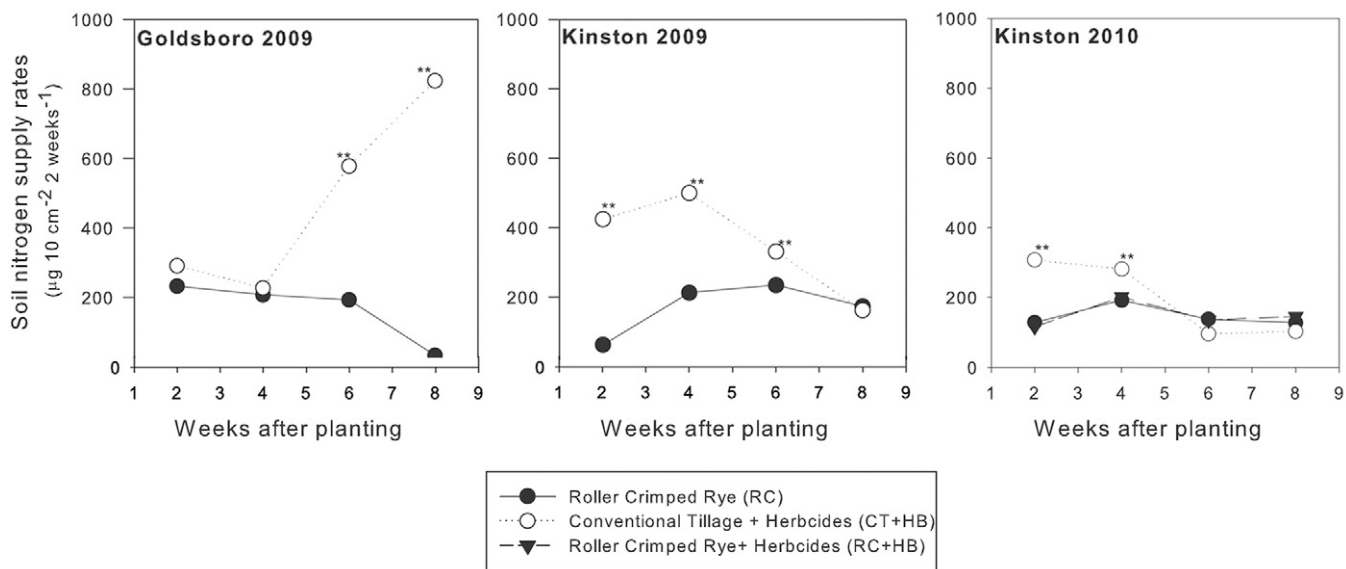


**Fig. 2. Extractable inorganic soil N from roll-crimped rye and conventional tilled plots. Asterisks indicate significant differences between treatments at a given time interval as determined by pre-planned contrast (\* $p \leq 0.05$ , \*\* $p \leq 0.01$ ). North Carolina.**

respectively, and 20.5 and 4.5 kg ha<sup>-1</sup> N in 2010, respectively (Fig. 2). Rye has been shown to be an excellent scavenger for N (Groffman et al., 1987; Jewett and Thelen, 2007; Meisinger et al., 1991) and its high C/N ratio biomass decomposes slowly, temporarily sequestering N in the accumulation of rye biomass. During growth, rye monocultures consistently result in lower levels of soil inorganic N when compared with legume monocultures (Ranells and Waggoner, 1997). A primary mechanism responsible for rye’s scavenging abilities is a fibrous and extensive root system. Generally, a rye cover crop has 20 to 30% of its total DM in roots, and rooting depth and density enable rye to scavenge and sequester soil inorganic N (Meisinger et al., 1991). The amount of rye residue grown in the southeastern United States is extremely high, with rates in excess of 9000 kg ha<sup>-1</sup> (Reberg-Horton et al., 2012). Such high mulch levels would mean that substantial levels of soil N are typically scavenged and accumulated into rye biomass in this system, resulting in a large pool of immobilized N that is temporarily unavailable for subsequent weeds.

Results show that at two of the three sites, soil inorganic N remained lower and more consistent in the RC and RC+HB plots until, but not including, the 8 WAP sampling interval (Fig. 2). Upward spikes in soil N availability were observed in CT+HB





**Fig. 3. Plant available N as measured by plant root simulator probes (PRS probes) from roll-cripped rye and conventional tilled plots. Asterisks indicate significant differences between treatments at a giving time interval as determined by pre-planned contrast (\* $p \leq 0.05$ , \*\* $p \leq 0.01$ ). North Carolina.**

plots in response to wetting and drying cycles (soil moisture data not shown). These spikes are typical in soil N data sets (Burgess et al., 2002; Coppens et al., 2007; Frey et al., 1999; Lundquist et al., 1999) as microbial biomass increases with moisture and increasing mineralization rates as microbes decompose soil organic matter. The continued low N in the RC plots suggests portions of C-rich rye residue were being decomposed and prevented the net N mineralization seen in the CT+HB plots.

The lack of initial extractable soil N treatment differences in Goldsboro (2009) may be attributed to high soil temperatures coupled with low soil moistures (M.S. Wells et al., unpublished data, 2010, 2011). Soil moisture and temperature regulate N mineralization rates (Frey et al., 1999; Jaeger et al., 1999; Ranells and Wagger, 1997; Six et al., 2002). At lower soil moisture contents, reduced supply rates of soil inorganic N are commonly observed as microbial activity declines (Qian and Schoenau, 2002). An increase in soil inorganic N was observed until 4 WAP in the deeper 10- to 25-cm fraction in the CT+HB treatments for all site-years (Fig. 2). This increase of soil inorganic N in the deep profile is likely the result of a series of rain events occurring 0 and 2 WAP (Fig. 1) that leached inorganic N into the lower soil profile (Fig. 2).

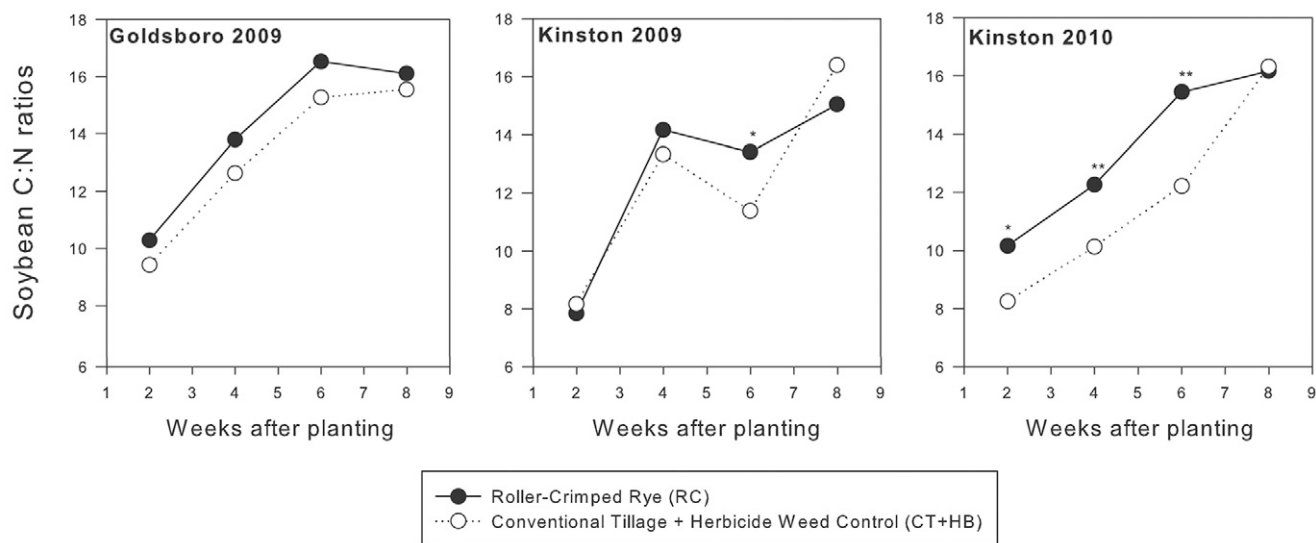
Several rye biomass factors are possibly contributing to the observed N immobilization. Even though rye roots account for 20 to 30% of the overall rye biomass, their C/N ratios (mature rye roots) range from 22:1 to 28:1, much lower than the C/N ratios of the rye surface residue (60–80:1); thus, rye roots are likely to contribute minimally to soil inorganic N immobilization (Malpassi et al., 2000). However, easily soluble C can contribute to bacteria-driven N immobilization by leaching from the rye shoots on the surface into the soil profile (Cochran et al., 1980). Another proposed mechanism is the N translocation to surface residues by fungal hyphae from the surrounding surface soils. This possibility has been raised in both agricultural and forestry systems. Reports demonstrating increased N content in surface litter in wheat (Christensen, 1986; Holland and Coleman, 1987; Schomberg

et al., 1994b) suggest transport of N from soil to surface residues went without confirmation until work on fungal transport of N revealed a similar pattern (Frey et al., 2000, 2003). Frey et al. (2000) demonstrated a 43% reduction in surface soil N immobilization when fungal populations were inhibited by fungicides along with a 52 to 86% reduction of total N transferred from mineral soil to wheat surface residues. Saprophytic decomposers in no-till systems can bridge the soil–litter interface while simultaneously transporting significant quantities of soil inorganic-N into the surface residues via fungal hyphae (Frey et al., 2000).

### Soybean and Pigweed Carbon/Nitrogen Ratios

At Kinston (2010) soybean tissue C/N ratios were higher in the RC plots for the first 6 wk but decreased to similar ratios detected in the CT+HB plots by 8 WAP (Fig. 4). Even though soybean tissue from RC plots at both Goldsboro and Kinston (2009) followed similar trends as those observed in Kinston (2010), only during the 6 WAP interval at Kinston (2009) were the differences significant ( $p < 0.05$ ) (Fig. 4). This time frame (6 WAP) corresponds to growth stages V2 to V3, when biological N<sub>2</sub> fixation is beginning in soybean nodules, suggesting that beyond Week 6 the soybean plants were sufficiently meeting their N requirements through N<sub>2</sub>–fixation. Nitrogen deprivation in soybean can reduce biomass production before nodulation (Heckman and Kluchinski, 1995) but the lack of difference in yields between RC+HB and the CT plots (Table 1) suggests that the low soybean N state in that system for the first 6 wk had limited impact on crop development.

Redroot pigweeds suffered a more severe response to N deprivation than the soybean. At both Goldsboro and Kinston during 2009, pigweed C/N ratios continued to diverge between the two treatments (CT and RC) over the 8-wk sampling period (Fig. 5). Comparable N deprivation in other weed species can reduce shoot biomass, seed number, and total seed mass and as a result, the offspring can be less competitive in low-N environments (Tungate et al., 2006).



**Fig. 4. Soybean C/N ratios from roll-crimped rye and conventional tilled plots. Asterisks indicate significant differences between treatments at a giving time interval as determined by pre-planned contrast (\* $p \leq 0.05$ , \*\* $p \leq 0.01$ ). North Carolina.**

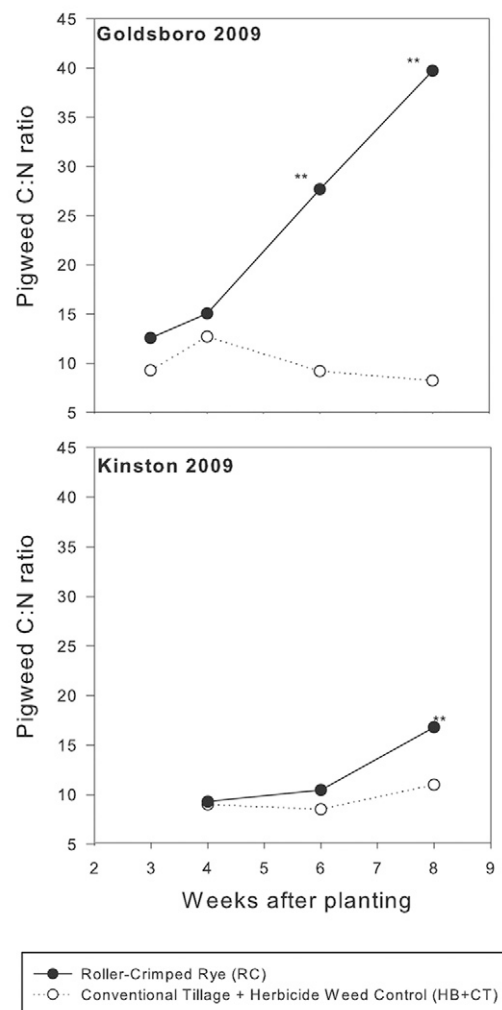
**Table 1. Weed management treatment effect on soybean yield.**

Treatment	Kinston	
	2009	2010
— yield kg ha <sup>-1</sup> —		
Weed management		
Roller-crimped rye	3525a†	3439a
Roller-crimped rye + herbicide	na‡	4020b
Conventional tillage + herbicide	6279b	4118b

† Means within columns followed by the same letter are not significantly different based on Fisher's Protected LSD ( $\alpha = 0.05$ ).

‡ na, not applicable.

These effects could play an important role in determining weed–crop competitive outcomes. The role of N in weed–crop competition has largely been studied in non-legume, N<sub>2</sub>-fixing cropping systems (Blackshaw, 2004; Henson and Jordan, 1982; Suspasilapa et al., 1992). In the current study, the RC plots had sustained lower soil inorganic-N levels ranging from 3.0 to 12.0 kg N ha<sup>-1</sup> throughout the season when compared to the CT plots. In the N-constrained RC plots, it may be possible to exploit the N responsiveness of some weed species during the critical period of weed competition. Species having the highest growth rates in fertile environments can be the most sensitive to suboptimal nutrient concentrations (Shipley and Keddy, 1988). Even though there can be a transfer of N from soybean to weeds, transfer of N is very low in weed species such as pigweed that are known to be non-hosts for arbuscular mycorrhizae (Moyer-Henry et al., 2006). The low soil inorganic-N in the RC plots could restrict the growth of weed species such as pigweed during the critical weed-free period (Eyherabide and Cendoya, 2002) between 2 and 7 wk after emergence, thus reducing weed–crop competition. During this time frame, soil inorganic-N in the RC plots was constantly lower than 12.0 kg N ha<sup>-1</sup> for all three sites. Four WAP the soil inorganic-N in RC and RC+HB plots at Kinston (2009 and 2010) continued to decline (Fig. 2), while pigweed C/N ratios showed increasing signs of N deprivation (Fig. 5).



**Fig. 5. Pigweed C/N ratios from roll-crimped rye and conventional tilled plots. Asterisks indicate significant differences between treatments at a giving time interval as determined by pre-planned contrast (\* $p \leq 0.05$ , \*\* $p \leq 0.01$ ). North Carolina.**

**Table 2. Weed management treatment effect on weed density.**

Treatment	2009		2010
	Goldsboro	Kinston	Kinston
	— weed counts m <sup>-2</sup> —		
Weed management			
Roller-crimped rye	7.5a†	1.2a	0.40a‡
Roller-crimper rye + herbicide	—	—	0.05b
Conventional tillage + herbicide	7.3a	0.99a	0.01b
Weedy check (WC)	29b	11b	1.37c

† Means within columns followed by the same letter are not significantly different based on Fisher's Protected LSD ( $\alpha = 0.05$ )

‡ Means were log-transformed for analysis due to lack of homogenous of variance and back transformed for presentation.

### Weed Control and Soybean Yield

The predominant weed at both locations was pigweed with the WC plots ranging from 1 to 29 plants m<sup>-2</sup> (Table 2). Pigweed densities were greatest in Goldsboro (2009) in the WC plots followed by Kinston (2009 and 2010) (Table 2). Roller-crimped rye had fewer weeds than the WC plots for all site-years but weed counts were significantly higher relative to the CT+HB and RC+HB plots in 1 of the 3 site-years (Table 2). The lower soybean yields in the RC plot (Table 1) are likely a result of weed competition. The similarity in yields between the RC+HB and CT systems suggests the two systems share similar yield potential in the absence of weeds. None of the sites achieved the approximately 9000 kg DM ha<sup>-1</sup> of rye biomass (with 4450, 8367, and 7084 kg ha<sup>-1</sup> DM at Goldsboro and Kinston [2009] and Kinston [2010], respectively) recommended to prevent yield loss from weeds in the RC system (Mohler and Teasdale, 1993; Smith et al., 2011). The already low N status of the soil and plants documented in this study would likely be reduced even further when rye biomass reaches this recommended level.

### CONCLUSIONS

Although the 3 site-years had varying rye biomass, the RC system created an extremely low N environment that affected N content in both pigweed and soybean before nodulation. Soybean plants appear able to recover from their N deprivation when nodulation becomes active, whereas pigweed continues to be affected. This suggests that when a cereal cover crop is paired with a legume cash crop, N deprivation can play a substantial role in weed suppression. The management implications of this finding may contradict previous recommendations. The need for extremely high levels of mulch to sufficiently suppress weeds (Teasdale and Mohler, 2000) has led to recommendations of fertilizing the cover crop to ensure maximal growth (Balkcom et al., 2008). If N applications to the cover crop result in higher soil N levels during the soybean phase, suppressing weeds with N deprivation may be unachievable. Finding a balance between maximizing cover crop growth and maintaining extremely low soil N during the soybean phase will require research on the rate and timing of N to the cover crop over a range of environments.

### ACKNOWLEDGMENTS

The authors gratefully acknowledge Carrie Brinton for her able technical support and the staffs of the Center for Environmental Farming Systems and Caswell Research Farm. This project was supported by SARE project number LS08-210.

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