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Fertilizer and Population Affects Nitrogen Assimilation of Common Lambsquarters (*Chenopodium album*) and Redroot Pigweed (*Amaranthus retroflexus*)

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Weed growth and N assimilation usually increase with N application rate. With the increasing price of N fertilizer, a better understanding N assimilation by weeds is necessary to maximize economic return. Total plant yield is generally independent of population density, except when plants are very small or at very low population density. If plant yield is independent of population density, weed N assimilation may also be independent of population density. However, the effect of weed population density on N assimilation has not been thoroughly investigated. A 2011 controlled-environment study was established in East Lansing, MI, to evaluate the effect of weed population density and N application rate on growth and N assimilation by common lambsquarters and redroot pigweed. Study factors included four weed densities (1, 2, 4, and 8 plants pot^{-1}), three N application rates (0, 67, and 134 kg N ha^{-1}), and two weed species (redroot pigweed and common lambsquarters). Weeds were destructively harvested 3 wk after emergence, and shoot height, biomass, total N concentration, N use efficiency, and N assimilation were measured. Redroot pigweed was taller, had greater shoot biomass, and a greater shoot N assimilation than did common lambsquarters. With similar environmental conditions, redroot pigweed is expected to be more competitive than common lambsquarters. Shoot N assimilation increased with increasing weed population density, indicating that N assimilation was not independent of population density 3 wk after emergence because weeds were small or at low population density.

Nomenclature: Common lambsquarters, *Chenopodium album* L. CHEAL; redroot pigweed, *Amaranthus retroflexus* L. AMARE.

Key words: C_3 : C_4 plant metabolism, intraspecific weed competition, nitrogen concentration; root : shoot.

Weeds actively compete with crops for water, nutrients, and light, which can reduce crop yield (Dalley et al. 2006; Gower et al. 2003). Nitrogen (N) is often a limiting nutrient in nonleguminous crop production (Joern and Sawyer 2006). Weed growth and N assimilation usually increase with N application rate (Ampong-Nyarko and De Datta 1993; Andreasen et al. 2006; Berger et al. 2007; Blackshaw et al. 2003; Teyker et al. 1991). With the increasing price of N fertilizer, a better understanding weed growth and N assimilation is necessary to maximize economic return.

Individual, weed biomass accumulation is influenced by weed population density (Radosevich 1987). However, according to the law of constant final yield, total plant biomass accumulation is generally independent of population density, except when plants are very small or at a low population density (Fausey et al. 1997; Harper 1977; Radosevich 1987). Because plant yield is generally independent of population density, weed N assimilation may also be independent of population density. However, the effect of weed population density on N assimilation has not been thoroughly investigated for common lambsquarters and redroot pigweed. The effect of weed population density on common lambsquarters and redroot pigweed shoot N assimilation 3 wk after emergence was investigated in this study. Weed shoot N was measured 3 wk after emergence to reflect the timing of POST weed control in corn (*Zea mays* L.) grain production, which is recommended before weeds reach 10 cm in height (Gower et al. 2003).

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Weed growth tends to increase with N application rates (Blackshaw et al. 2003). Common lambsquarters and redroot pigweed are among the most-responsive weeds, both demonstrating high rates of shoot and root biomass accumulation with increasing N application rate (Blackshaw et al. 2003). An increase in weed-shoot biomass from N application has been noted in other studies (Barker et al. 2006; Iqbal and Wright 1997; Teyker et al. 1991). Generally, shoot biomass increases more than root biomass with increasing N levels because plants have less need to increase root growth when N supply is adequate (Blackshaw et al. 2003). Under N-limiting conditions, velvetleaf (*Abutilon theophrasti* Medik.) root biomass increased, whereas shoot biomass decreased, suggesting that the species may compete less for light as N becomes limiting (Bonifas et al. 2005).

Nitrogen concentration of weeds increases with N application rate with a maximum total N concentration reaching 4 to 5% (Blackshaw and Brandt 2008; Blackshaw et al. 2003; Iqbal and Wright 1997). However, the degree of response is species specific (Blackshaw et al. 2003), which may be partly attributed to the functional differences between C_3 and C_4 plant metabolism (Abouziena et al. 2007). Plants with C_3 metabolism often have higher shoot N concentrations, whereas plants with C_4 metabolism tend to produce more dry matter per unit shoot N (Brown 1978, 1985; Sage and Pearcy 1987). Photorespiration occurs in C_3 plants, requiring a greater total N concentration to maintain plant metabolism, whereas a C_4 plant uses carbon dioxide more efficiently (Jackson and Volk 1970). When N is limiting, plants with C_3 metabolism increase root growth proportionally more than C_4 plants do to maintain a sufficient level of tissue N and sustain photosynthetic rates and dry matter accumulation (Bonifas et al. 2005).

Total plant yield increases with population density, then gradually approaches a final value that is independent of population density (Radosevich 1987). We hypothesize that

Table 1. Significance of *F* values for fixed sources of variation from statistical analyses across trials.

Dependent variable	Species (*S*)	N rate (*N*)	Population density (*P*)	*S* × *N*	*S* × *P*	*N* × *P*	*S* × *N* × *P*
	P > *F*						
Height	0.002	0.180	0.008	0.762	0.172	0.680	0.586
Root biomass	0.958	0.722	0.731	0.465	0.397	0.528	0.768
Shoot biomass	0.001	0.349	< 0.001	0.118	0.487	0.692	0.586
Root : shoot	0.096	< 0.001	0.069	0.474	0.485	0.822	0.356
Percentage of total N	< 0.001	< 0.001	< 0.001	0.950	0.756	0.946	0.536
N use efficiency	0.001	< 0.001	< 0.001	0.535	0.574	0.751	0.953
N assimilation	0.029	< 0.001	< 0.001	0.003	0.631	0.943	0.844

N assimilation will also increase with weed population density, then gradually approach a final value independent of population density. However, relationship between N assimilation of weeds and population density has not been previously examined for common lambsquarters and redroot pigweed. The influence of weed population density on N assimilation will increase our understanding of N loss from weeds. The objectives of our study were to examine the effects of N application rate and weed population density on common lambsquarters and redroot pigweed height, biomass, N concentration, N use efficiency (NUE), and N assimilation at 3 wk after emergence.

Materials and Methods

The study was conducted under controlled environmental conditions in the Michigan State University research greenhouses (East Lansing, MI) during 2011. The study was a split-plot, randomized, complete-block design with five replications of treatments and two experimental trials. Main plot factor consisted of weed species. Subplot factors included N application rate and weed population density. The first trial began on February 25, 2011, and the second trial began March 8, 2011. Factors included two weed species (common lambsquarters and redroot pigweed), four weed densities (1, 2, 4, and 8 weeds pot^{-1}), and three N application rates (0, 67, and 134 kg N ha^{-1}).

Weed seeds were planted into 100-cm^2 pots containing 1,000 g of steam-sterilized Spinks loamy sand (sandy, mixed, mesic Lamellic Hapludalfs). Soil pH was 7.5, and cation exchange capacity was 7.5 cmol kg^{-1}. The soil was 69.3% sand, 20.2% silt, and 10.5% clay. Total N concentration was 2.15 g kg^{-1}. Ammonium-N and nitrate-N concentration were 15.8 and 10.4 mg kg^{-1}, respectively. Bray phosphorus was 94 mg kg^{-1}, exchangeable K was 83 mg kg^{-1}, and exchangeable magnesium was 127 mg kg^{-1}.

Nitrogen was supplied as a 0.06% (w/w) N solution made by dissolving urea in deionized water and applying it to the soil surface the same day as planting. To incorporate the N solution, 100 ml of water was added to the soil surface. Weeds were thinned to the desired density at the cotyledon stage. To ensure water was not a growth-limiting factor, weeds were watered at least once daily. Pots were rotated by replication every week. Natural light was supplemented with artificial light at 400 µmol m^{-2} s^{-1} photosynthetic photon flux in a 16-h day. Conditions in the greenhouse were maintained at a day/night temperature of 27/24 C.

Three weeks after emergence, the weed height was recorded, and weeds were harvested. At harvest, shoot portions were separated from root portions by cutting the shoots at the soil surface. A 1-mm sieve was used to remove roots from the soil. Roots were gently washed with deionized water to remove adhering soil. Shoots and roots were dried in a forced-air dryer at 65 C for at least 72 h, and dry weights recorded. Shoot tissue was ground to pass through a 0.178-mm sieve. Shoot total N content was determined by the micro-Kjeldahl digestion (Jung et al. 2003) and colorimetric analysis through a Lachat rapid flow injector autoanalyzer (Lachat Instruments, Loveland, CO). Shoot NUE was evaluated by dividing weed-shoot biomass (milligrams per pot) by unit-shoot N assimilated by the weeds (milligrams of N per 100 mg of biomass). At least 150 mg of plant tissue was needed to conduct the micro-Kjeldahl procedure for measuring total N concentration. Because of limited root biomass, the total N concentration and NUE for weed roots were not able to be determined.

Treatment effects of weed species, weed population density, and N application rate were examined using ANOVA in the PROC MIXED procedure of SAS (SAS Institute, Cary, NC; SAS Institute 2001) at $\alpha = 0.05$. Fixed variables included weed species, weed population density, N application rate, and the interactions of these factors. Laboratory trial, replication nested within laboratory trial, and the interaction of laboratory trial, replication, and weed species were considered random variables. Normality and equal variance assumptions were checked using Levene's procedure (Schultz 1985). Shoot-biomass data were log-transformed to meet the normality assumption. However, untransformed data are presented for readability, with statistical analysis based on transformed data. When the unequal variance assumption was not met, treatments were grouped by similar variances. If the effect of weed species, weed population density, N application rate, or their interactions were found significant, means were separated using a paired *t* test ($\alpha = 0.05$) using the LSMEANS option in SAS software.

Results and Discussion

Weed Height and Biomass. Weed height and shoot biomass were influenced by the main effects of weed species and population density (Table 1). Three weeks after emergence, the average height of common lambsquarters was 6.9 cm, whereas the average height of redroot pigweed was 9.0 cm (Table 2). Shoot biomass averaged 526 and 850 mg pot^{-1} for common lambsquarters and redroot pigweed, respectively. Differences in height and aboveground biomass accumulation of redroot pigweed and common lambsquarters may have been temperature dependent. Greenhouse day/night temperature was 27/24 C. Day/night temperatures of 29/24 C favor redroot pigweed shoot growth, rather than common lambsquarters (Chu et al. 1978). At a day/night temperature of 17/14 C, common lambsquarters has a greater relative growth rate than dies redroot pigweed (Pearcy et al. 1981).

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Table 2. Main effects of weed species, N rate, and weed population density on weed shoot height, root biomass, shoot biomass, and root : shoot.[a,b]

Factor	Shoot height	Root biomass	Shoot biomass	Root : shoot
	cm	—— mg pot^{-1} ——		
Species				
CHEAL	6.9 b	188 a	526 b	0.20 a
AMARE	9.0 a	108 a	850 a	0.23 a
N rate (kg N ha^{-1})				
0	8.3 a	174 a	641 a	0.27 a
67	7.9 a	144 a	742 a	0.21 b
134	7.8 a	125 a	681 a	0.17 b
Population (plants pot^{-1})				
1	7.7 b	74 a	501 c	0.19 a
2	7.4 b	107 a	510 c	0.20 a
4	8.3 a	182 a	732 b	0.24 a
8	8.6 a	227 a	1,010 a	0.22 a

[a] Abbreviations: CHEAL, common lambsquarters; AMARE, redroot pigweed.
[b] Means within a column and study factor followed by the same letter are not significantly different according to all pairwise comparisons among the treatments conducted using a *t* test at the 0.05 level of significance.

Table 3. Main effects of weed species, N rate, and weed population density on weed shoot N and nitrogen use efficiency (NUE).[a,b]

Factor	Nitrogen	NUE
	%	mg biomass mg^{-1} N
Species		
CHEAL	5.02 a	24 b
AMARE	3.46 b	35 a
N rate (kg N ha^{-1})		
0	3.47 c	37 a
67	4.36 b	28 b
134	4.89 a	24 b
Density (plants pot^{-1})		
1	5.02 a	23 c
2	4.65 a	26 bc
4	4.00 b	31 b
8	3.30 c	39 a

[a] Abbreviations: CHEAL, common lambsquarters; AMARE, redroot pigweed.
[b] Means within a column and study factor followed by the same letter are not significantly different according all pairwise comparisons among the treatments conducted using a *t* test at the 0.05 level of significance.

The root : shoot ratio was greatest for weeds grown with no N application, indicating that root growth was greater, relative to shoot growth, when N was limiting (Table 2). There was no difference in root : shoot ratio when weeds were grown at 67 or 134 kg N ha^{-1}. Most summer, annual weeds increase root growth relative to shoot growth at low N levels (Blackshaw et al. 2003; Bonifas et al. 2005). At low N levels, plants respond by partitioning biomass to roots to maximize the capture of N and thereby increase plant growth rate and carbon acquisition (Bloom et al. 1985; McConnaughay and Coleman 1999).

Average weed height was 8.3 and 8.6 cm when grown at 4 and 8 plants pot^{-1}, respectively (Table 2), but only 7.7 and 7.4 cm tall at 1 and 2 plants pot^{-1}, respectively. Weeds may have been taller at higher densities because of shade avoidance strategies that include accelerated stem extension, which has been observed in pigweed species and common lambsquarters (Brainard et al. 2005; Gramig and Stoltenberg 2009; Morgan and Smith 1976; Rajcan et al. 2002). Shoot biomass also increased with increasing population (Table 2).

At 1, 2, 4, and 8 plants pot^{-1}, whole-pot shoot biomass was 501, 510, 732, and 1,010 mg pot^{-1}, respectively. The law of constant final yield suggests that total plant yield is generally independent of population density (Harper 1977). Weeds grown at 1 to 8 plants pot^{-1}, did not have the same whole-pot shoot biomass, indicating that the weeds were too small or too widely spaced to be independent of density (Radosevich 1987). Before POST weed control, common lambsquarters and redroot pigweed shoot biomass are likely to increase with increasing population density. Similarly, the relationship between weed population density and yield loss is hyperbolic, indicating that crop yield losses initially increase linearly with increasing weed population density, but as weed density as weed density reaches a threshold value, crop yield losses become constant (Fausey et al. 1997).

Total N Concentration, NUE, and N Assimilation. Total shoot N concentration and NUE were influenced by the main effects of weed species, N application rate, and population density (Table 1). Nitrogen assimilation was influenced by the interaction of weed species and N application rate and by the main effect of population density.

Average shoot N concentration was 5.02 and 3.46% for common lambsquarters and redroot pigweed, respectively (Table 3). Common lambsquarters shoot N concentrations ranging from 1.20 to 4.12% have been recorded previously (Blackshaw et al. 2003; Qasem 1993; Vengris et al. 1953). Redroot pigweed shoot N concentrations have been found to range from 1.44 to 4.40% (Blackshaw et al. 2003; Qasem 1993; Teyker et al. 1991). Total shoot N concentration is variable because it is influenced by environmental conditions, weed size, and soil nitrogen levels (Blackshaw et al. 2003). In this study, weeds were at a similar growth stage to weeds controlled in the field at POST herbicide application and reflect shoot total N concentration of weeds growing in a field.

Redroot pigweed had a smaller total N concentration and greater shoot biomass than did common lambsquarters, which resulted in greater NUE of redroot pigweed compared with common lambsquarters (Tables 2 and 3). Plants with high NUE can produce more biomass per unit of N than can plants with low NUE. In situations of low soil-nitrogen levels, plants with high NUE are expected to be more competitive than are plants with low NUE. NUE is species dependent. Plants with C_4 metabolism tend to have a higher NUE than do plants with C_3 metabolism, whereas C_3 plants tend to have a greater total N concentration (Brown 1985). Photorespiration occurs in C_3 plants, requiring a greater total N concentration to maintain plant metabolism, whereas a C_4 plants use carbon dioxide more efficiently (Jackson and Volk 1970). Additionally, photorespiration slows the reduction of nitrate to ammonium (Oaks 1994), which is subsequently converted to amino acids.

Average shoot N concentration was 3.47, 4.36, and 4.89% when weeds were grown at 0, 67, and 134 kg N ha^{-1}, respectively (Table 3). An increase in total N concentration with increasing N application has been noted in several other studies (Andreasen et al. 2006; Blackshaw et al. 2003; Blackshaw and Brandt 2008). Conversely, NUE increased with decreasing N application rate (Table 3), also similar to previous studies (Ampong-Nyarko and De Datta 1993; Craswell and Vlek 1979). NUE is controlled by genetic and

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Table 4. Interaction of weed species and N application rate on N assimilation and main effect of weed population density on N assimilation.[a,b]

Factors	N rate	N assimilation
Species	kg N ha^{-1}	mg N pot^{-1}
CHEAL	0	25 d
	67	29 c
	134	30 BC
AMARE	0	22 d
	67	33 b
	134	39 a
Population plants pot^{-1}		
1		22 d
2		26 c
4		33 b
8		38 a

[a] Abbreviations: CHEAL, common lambsquarters; AMARE, redroot pigweed.
[b] Means within a column and study factor followed by the same letter are not significantly different according all pairwise comparisons among the treatments conducted using a *t* test at the 0.05 level of significance.

environmental factors (Baligar et al. 2001; Oaks 1994). Weeds, including common lambsquarters and redroot pigweed, can assimilate and store excess N (luxury N consumption) in vacuoles, resulting in low NUE (Ampong-Nyarko and De Datta 1993). Additionally, at low N application rates, plant fiber and carbohydrate concentrations increase causing an increase in NUE (Chapin 1980).

Total N concentration was 5.02, 4.65, 4.00, and 3.30% when weeds were grown at 1, 2, 4, and 8 plants pot^{-1} (Table 3). As mentioned previously, total shoot biomass pot^{-1} increased with increasing population density, resulting in a dilution effect on the total N concentration in the shoot tissue (Blackshaw et al. 2003). Nitrogen use efficiency was greatest at 8 plants pot^{-1}. Nitrogen use efficiency decreased in situations when N was nonlimiting, such as at high N application rate and at low population density.

Shoot nitrogen assimilation was influenced by the interaction of weed species and N application rate (Table 1). Redroot pigweed grown at 134 kg N ha^{-1} had the greatest N assimilation, followed by pigweed grown at 67 kg N ha^{-1} and common lambsquarters grown at 134 kg N ha^{-1} (Table 4). Shoot N assimilation was dependent on shoot biomass and N concentration. Redroot pigweed had greater shoot biomass than did common lambsquarters (Table 2), and weeds grown at 134 kg N ha^{-1} had a greater N concentration than did the other N application rates (Table 3). Nitrogen assimilation increased with weed population density (Table 4). Shoot biomass was greatest at 8 plants pot^{-1} (Table 2). A higher shoot biomass resulted in higher total N assimilation compared with weeds grown at a low population density with less shoot biomass. If weeds were larger or at a higher population density, N assimilation may be independent of population density similar to the law of constant final yield (Harper 1977; Radosevich 1987). However, in the field at the time of POST herbicide application, N assimilation will increase with increasing weed population density.

Management Implications. Three weeks after emergence, redroot pigweed was taller, produced more shoot biomass, and assimilated more N than did common lambsquarters (Tables 2 and 4). Redroot pigweed also had a greater NUE than common lambsquarters did (Table 3). In a study comparing weed competitiveness, Roush and Radosevich (1985), concluded that redroot pigweed was more competitive than common lambsquarters was. Because their (Roush and Radosevich 1985) environmental conditions were similar to this study, redroot pigweed may be more competitive than common lambsquarters is. However, environmental conditions that favor growth of plants with C_3 metabolism (day/night temperatures of 17/14 C) may result in common lambsquarters being more competitive than redroot pigweed (Chu et al. 1978; Pearcy et al. 1981). The results of this study and previous studies indicate that the competitiveness of common lambsquarters and redroot pigweed is dependent on environmental conditions.

Nitrogen assimilation by common lambsquarters and redroot pigweed increased with increasing N application rate (Table 4). Nitrogen management strategies that limit N assimilation by weeds and favor crop growth may be beneficial to reduce crop–weed competition (Di Tomaso 1995). Weed interference with crops has been reduced by placing fertilizer in a band instead of broadcasting it (Blackshaw et al. 2004; Mesbah and Miller 1999). Timing fertilizer applications when crop assimilation is rapid may also reduce weed interference (Davidson 1984).

High weed densities resulted in greater N assimilation than that assimilated at low weed densities (Table 4). Weed management strategies that reduce the weed seed bank, and subsequently weed density, should reduce N loss to weeds early in the growing season. At a weed population of 8 plants pot^{-1}, N assimilation by weeds was equivalent to 85 kg N ha^{-1}, whereas at 1, 2, and 4 plants pot^{-1}, N assimilation by weeds was equivalent to 49, 58, and 74 kg N ha^{-1}, respectively. Two-pass herbicide programs can reduce early season weed competition and late-emerging weeds (Bradley et al. 2000; Gower et al. 2002), reducing the weed seed bank. N management and weed-control strategies that limit N assimilation by weeds and reduce weed density should be employed.

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