Inhibitory Effects of Redroot Pigweed and Crabgrass on Germination and Growth—From Lab To Field

Xincun Hou (houxincun@grass-env.com)
Research and Development Centre for Grass and Environment, Beijing Academy of Agriculture and Forestry Sciences
https://orcid.org/0000-0003-3860-9797

Xu Hu
National Energy Conservation Center

Yuesen Yue
Beijing Academy of Agriculture and Forestry Sciences

Qiang Guo
Beijing Academy of Agriculture and Forestry Sciences

Chunqiao Zhao
Beijing Academy of Agriculture and Forestry Sciences

Xifeng Fan
Beijing Academy of Agriculture and Forestry Sciences

Juying Wu
Beijing Academy of Agriculture and Forestry Sciences

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Abstract

Interactions between weeds and crops often occur by resource competition or allelopathy. However, it is still unknown how local weed species influence artificially introduced switchgrass. In this study, four experiments were conducted to evaluate the inhibitory effects of redroot pigweed (*Amaranthus retroflexus*) and crabgrass (*Digitaria sanguinalis*) on germination and growth of switchgrass, the lowland tetraploid cultivar 'Alamo' (*Panicum virgatum* cv. Alamo). Switchgrass germination was inhibited significantly in Petri dishes, with 48.1% and 33.9% inhibition on germination rate by redroot pigweed and crabgrass root aqueous extracts at 0.1 g·mL\(^{-1}\) concentration, respectively. Significant inhibitory effects on switchgrass seedling biomass were observed at 5:5 proportion with redroot pigweed and crabgrass in glass jars, with 61.6% and 53.4% inhibition on plant biomass, respectively. Under the same root segregation, redroot pigweed had a stronger inhibitory effect on switchgrass seedling growth than crabgrass. Growth of transplanted switchgrass seedlings was inhibited significantly by local weeds in the field, with 46.2% and 11.7% inhibition on shoot biomass during the first and second growing seasons, respectively. However, no significant growth reduction in switchgrass was detected in the third growing season.

Introduction

Plants are able to perceive and respond to chemical cues emitted by their neighbors, and neighboring plants can affect plant survival and performance, either positively or negatively (Wang et al. 2020; Xia et al. 2016). Positive interactions, known as facilitation, range from protecting other plants from herbivore impacts, potential competitors, and climatic extremes to contributing to resource acquisition by canopy leaching, microbial enhancement, mycorrhizal networks, and hydraulic lift (Brooker et al. 2008). Alternatively, negative interactions occur, such as inhibiting seed germination through allelopathy or reducing growth of other plants through competition for resources (Brooker et al. 2008; Treutter 2005). These varied plant–plant interactions can regulate community and ecosystem composition by altering resource availability and environmental conditions (Brooker 2006), and may change from competition to facilitation when stress is high (García-Cervigón et al. 2013). However, because of their complexity, plant–plant interactions are not fully understood, and further elucidation of these interactions can help predict and potentially mitigate negative impacts.

Interaction between local weed species and crops often occurs by competing for resources or releasing allelochemicals. Weeds can greatly impact crop yield leading to economic losses. Substantial crop losses associated with weeds have been observed in various crops, such as rice (*Oryza sativa*) and wheat (*Triticum aestivum*) (Oerke and Dehne 2004). In agriculture, competition studies have often focused on minimizing the effect of weeds on crops by methods such as optimizing crop densities and developing predictive tools for weed management (Kropff and van Laar 1993). Allelopathy occurs when a living or dead plant releases allelochemicals into the environment, exerting an effect on the survival and persistence of other plants (Yang and Kong 2017). Allelochemicals can be released by both crops and weeds and may have either positive or negative effects (Cheng and Cheng 2015). Understanding the
complex weed–crop interactions may help to develop management strategies for weeds and reduce losses associated with reduced crop yield (Maxwell and O'Donovan 2007). Moreover, allelochemicals released by certain crop cultivars can be utilized to inhibit growth of weeds, providing a non-herbicidal alternative for weed management (Yang et al. 2017).

Switchgrass (*Panicum virgatum*), a warm-season C\(_4\) perennial grass species (Steffany et al. 2018; Yue et al. 2017), is considered a viable herbaceous second-generation bioenergy crop, suitable to marginal lands (Cherney et al. 2018; Mitchell et al. 2016). It was introduced to China's Loess Plateau in the 1990s and then cultivated on a large scale in Northern China (Ma et al. 2018). Switchgrass grows in the same period as some weed species during the growing season. Therefore, it is important to analyze weed–switchgrass interaction, in order to lay a foundation for weeds ecological control in switchgrass cultivation. Redroot pigweed (*Amaranthus retroflexus*), an annual dicotyledonous plant species (Ma et al. 2015), and crabgrass (*Digitaria sanguinalis*), an annual monocotyledonous plant species (Zhou et al. 2013a), are two common weed species around the world. In this study, redroot pigweed and crabgrass were selected to investigate weed–switchgrass interaction, based on the ecological relevance of these two weed species to switchgrass in Northern China.

Local weed species can be influenced by switchgrass through resource competition or allelopathy. In our previous research, allelopathic effects of switchgrass on redroot pigweed and crabgrass growth were investigated (Li et al. 2021), and flavonoid compounds from switchgrass extracts were identified as isorhamnetin-3-O-rutinoside, quercetin-3-O-galactoside, quercetin-3-O-glucoside, quercetin-3-O-rutinoside, quercetin-3-O-xyloside, quercetin-3-O-arabinoside, and quercetin-3-O-rhamnoside (Li et al. 2018). However, it is not known how local weed species influence switchgrass. In this study, the inhibitory effects of redroot pigweed and crabgrass on switchgrass germination and growth were evaluated in four experiments. First, the allelopathic effects of redroot pigweed and crabgrass root aqueous extracts on switchgrass germination were evaluated in a Petri dish experiment. Subsequently, in two glass jar experiments, the inhibitory effects of redroot pigweed and crabgrass on switchgrass seedling growth were further examined, at different proportions and under root segregation, respectively. Furthermore, the inhibitory effects of redroot pigweed, crabgrass, and other local weeds on transplanted switchgrass seedlings’ growth were investigated in a field experiment.

**Materials And Methods**

**Experimental site**

All experiments were conducted at National Experimental Station for Precision Agriculture in Xiaotangshan Town (40°10′ N, 116°26′ E, hereinafter referred to as Xiaotangshan Station), in Beijing suburb, Northern China. Xiaotangshan Station is characterized by a continental, semi-humid, and monsoonal climate. The mean annual rainfall is 640 mm, of which 80% occurs between June and August. The mean annual temperature is 11.5 °C, and the lowest and highest monthly mean temperatures are −4.1 °C in January and 25.8 °C in July, respectively. The annual sunshine hours are
2641.4 h. The active accumulated temperature (≥ 10 °C) is 4188.3 °C. The frost-free period is 198.0 days, approximately from mid-April to mid-October.

The topsoil, to a depth of 0–10 cm in the field, is clay loam, composed of 41% sand, 24% silt, and 35% clay, with bulk density of 1.37 g·cm⁻³, pH of 7.6, organic matter content of 1.4%, total nitrogen of 2.46 g·kg⁻¹, and total phosphorus of 0.63 g·kg⁻¹.

Switchgrass cultivar

The lowland tetraploid cultivar ‘Alamo’ of switchgrass (Panicum virgatum cv. Alamo, 2n = 4X = 36) (Young et al. 2012; Yue et al. 2017) was used for this research object as it is one of the most common cultivars cultivated in Northern China.

Bioassay experiment

During the growing season in 2017, a bioassay experiment was carried out to evaluate the allelopathic effects of redroot pigweed and crabgrass root aqueous extracts on switchgrass germination in Petri dishes. Plump seeds of switchgrass and roots of these two weed species were collected from the field at Xiaotangshan Station during growing seasons in 2016 and 2017, respectively.

After being washed with distilled water three times, redroot pigweed and crabgrass roots were lyophilized for at least 48 h at −80 °C in a freeze dryer, and then ground into powder to pass a 1-mm screen mesh. 10 g powder was homogenized in 100 mL distilled water for 30 min. The homogenate was filtered, and the filtrate was used as the root aqueous extract at 0.1 g·mL⁻¹ concentration, which was diluted to prepare a series of root aqueous extracts at concentrations of 0.005, 0.01, 0.02, and 0.05 g·mL⁻¹.

Each Petri dish, with 90 mm inner diameter, was lined with two layers of filter paper and moistened with addition of 5 mL root aqueous extract at different concentrations. Control Petri dishes only received 5 mL distilled water. Plump switchgrass seeds were surface sterilized with 75% alcohol for 5 min, and then with 3% sodium hypochlorite for 10 min, before being rinsed with distilled water three times. Fifty seeds were placed on the filter paper in each Petri dish. All Petri dishes were covered, sealed with parafilm and placed in a controlled environmental chamber with a 16:8 h light:dark photoperiod, 30:25 °C light:dark temperatures, and 70% relative humidity. A randomized complete block design with three replicates was used and the positions of all Petri dishes were randomized once a day.

After 12 days, germinated switchgrass seeds with radicle length (≥ 1 mm) were counted for germination rate calculation. Ten germinated switchgrass seeds were selected randomly from each Petri dish, for plumule and radicle lengths measurement.

Glass jar experiments

Mixture determination for weed-switchgrass system
During the growing season in 2016, a replacement experiment was conducted in glass jars, 100 mm inner diameter and 90 mm height, containing 500 g soil, in a greenhouse with 32:21 °C day:night temperatures and 65–90% relative humidity at Xiaotangshan Station. The objective of this experiment was to identify the optimal mixture proportion for the subsequent root segregation experiment.

Plump seeds of redroot pigweed, crabgrass, and switchgrass were collected from the field during the growing season in 2015, and seedlings were cultured in the greenhouse in mid-February 2016. A total of ten weed and switchgrass seedlings at three-leaf stage were planted in each glass jar. The proportions of weed to switchgrass were 1:9, 2:8, 3:7, 4:6, 5:5, 6:4, 7:3, 8:2, and 9:1, respectively. A monoculture of ten switchgrass seedlings served as the control (Fig. 1).

Root segregation of weed and switchgrass

During the growing season in 2016, a root segregation experiment of weed and switchgrass was conducted in glass jars, in the same greenhouse as described in the experiment on mixture determination. The objective of this experiment was to evaluate the role of different components involved in the soil root system on switchgrass seedling growth at 5:5 proportion with weeds.

The experiment was carried out in a series of glass jars, with the same dimensions as described above. A total of sixteen glass jars were divided into four groups: root segregation with plastic film (hereinafter referred to as complete segregation, as the control), root segregation with 0.45 µm Millipore filtration (preventing both mycorrhizal hyphae linkages and root penetration but allowing chemical and bacterial interaction, hereinafter referred to as partial contact 0.45), root segregation with 30 µm Millipore filtration (preventing root penetration but allowing microbial penetration and chemical interaction, hereinafter referred to as partial contact 30), and without root segregation (hereinafter referred to as full contact) (Fig. 2). Each glass jar contained a central cylinder, 70 mm inner diameter and 90 mm height, made of plastic mesh frame, where plastic film or Millipore filtration could be inserted.

Each glass jar was filled with 500 g soil, 250 g inside and 250 g outside the central cylinder. Seedlings of redroot pigweed, crabgrass, and switchgrass were cultured in the greenhouse in mid-May 2016. Five switchgrass seedlings and five weed seedlings, at three-leaf stage, were planted inside and outside the central cylinder, respectively, in each glass jar.

In the two glass jar experiments described above, the soil used was naturally air-dried topsoil sampled in the field at Xiaotangshan Station. Four replicates were used for each treatment or control in a randomized complete block design, and the positions of all glass jars were randomized once a day. Glass jars were irrigated daily with distilled water. Any seedlings, except those of tested species, were manually removed as soon as they were detected. After 30 days, switchgrass seedlings were harvested, washed three times, and their shoots and roots were dried for at least 48 h at 80 °C in an oven to measure their biomass.

Field experiment
During growing seasons from 2014 to 2016, a field experiment was conducted to evaluate the inhibitory effects of local weeds on transplanted switchgrass seedlings’ growth at Xiaotangshan Station over a three-year period.

During the growing season in 2013, plump switchgrass seeds were collected from the field and were sown in the same greenhouse as described in the above experiment on mixture determination in mid-February 2014. In mid-May 2014, six field plots of 4.0 × 4.0 m^2 were prepared and sixteen switchgrass seedlings at six-leaf stage were transplanted into 2.4 × 2.4 m^2 central area of each field plot with inter-plant and inter-row spacing of 0.8 m. In three field plots, all weeds were allowed to grow naturally with switchgrass, while, in the other three field plots, all weeds were manually removed as soon as they were detected, as the control. All field plots, with weeds and without weeds, were arranged in a randomized complete block design with three replicates.

The field plots were irrigated immediately after transplantation to ensure switchgrass establishment, but not thereafter during the experimental period. The equivalent of 150 kg·ha^{-1} N:P:K fertilizer (15:15:15) was applied as basal fertilizer, followed by 150 kg·ha^{-1} urea application in mid-June during the first growing season.

During growing seasons from 2014 to 2016, a sampling quadrat of 1.6 × 1.6 m^2 was set in the central area of each field plot. In each sampling quadrat, there were four switchgrass plants, of which three were labeled randomly. After anthesis, tiller number was counted, and twenty tillers were selected randomly from these three plants for plant height measurement in situ. At the end of November in each year, when switchgrass stems had fully senesced, shoots of these three plants were harvested at stubble height of 5 cm. Twenty tillers with relatively uniform size were selected from these three plants for phytomer number calculation in situ. These twenty tillers and other shoots were washed three times, and then dried for at least 96 h at 80 °C in an oven, for shoot and phytomer biomass measurement, respectively.

**Data analysis**

Plant biomass was calculated by shoot biomass + root biomass in glass jar experiments. Weed effects, as percentages, were calculated by (control – treatment)/control × 100%, based on germination characteristics in the Petri dish experiment, and growth characteristics in glass jar and field experiments, respectively.

Analysis of variance, multiple comparisons, and independent t-test were performed in Microsoft Excel 2007 for Windows. Data were presented as means ± standard error in each experiment.

**Results**

**Switchgrass seed germination with weeds root aqueous extracts**
Redroot pigweed and crabgrass root aqueous extracts affected germination rate of switchgrass seeds (Fig. 3A), and plumule and radicle lengths (Fig. 3B, C) in Petri dishes.

Redroot pigweed root aqueous extracts at concentrations of 0.02, 0.05, and 0.1 g·mL⁻¹ affected switchgrass germination rate significantly, with 16.9%, 31.4%, and 48.1% inhibition ($P=0.025$, $P=0.009$, $P<0.001$), respectively (Fig. 3A). Switchgrass plumule length increased significantly at 0.005 g·mL⁻¹ concentration, with 23.4% promotion ($P=0.009$), and was inhibited significantly at concentrations of 0.02, 0.05, and 0.1 g·mL⁻¹, with 14.5%, 28.2%, and 39.3% inhibition ($P=0.029$, $P=0.021$, $P=0.002$), respectively (Fig. 3B). Switchgrass radicle length decreased significantly at 0.05 and 0.1 g·mL⁻¹ concentrations, with 20.7% and 34.6% inhibition ($P=0.036$, $P=0.003$), respectively (Fig. 3C).

Crabgrass root aqueous extracts inhibited switchgrass germination rate significantly at concentrations of 0.05 and 0.1 g·mL⁻¹, with 25.9% and 33.9% inhibition ($P=0.003$, $P=0.004$), respectively (Fig. 3A). Switchgrass plumule length increased significantly at 0.005 g·mL⁻¹ concentration, with 20.3% promotion ($P=0.012$), and decreased significantly at 0.1 g·mL⁻¹ concentration, with 28.9% inhibition ($P=0.030$) (Fig. 3B). Switchgrass radicle length increased significantly at 0.01 g·mL⁻¹ concentration, with 23.0% promotion ($P=0.034$), and decreased significantly at 0.1 g·mL⁻¹ concentration, with 26.9% inhibition ($P=0.025$) (Fig. 3C).

**Switchgrass seedling growth with weeds at different mixture proportions**

Redroot pigweed and crabgrass inhibited switchgrass seedling growth significantly at different mixture proportions in glass jars, except on root biomass at 2:8 proportion in redroot pigweed–switchgrass system (Fig. 4B), and on shoot (Fig. 4A), root (Fig. 4B), and plant (Fig. 4C) biomass at 1:9 proportion in crabgrass–switchgrass system.

Within redroot pigweed–switchgrass system, the inhibitory effects on switchgrass seedling growth increased as redroot pigweed seedling population increased from 1:9 to 5:5 proportion, on the whole. When redroot pigweed seedling population was close to switchgrass seedling population, the most significant inhibitory effects were observed, with biomass reductions of 60.1%, 64.9%, and 61.6% on shoot, root, and plant ($P<0.001$, $P=0.031$, $P=0.005$) at 5:5 proportion, respectively. From 6:4 to 9:1 proportion, the inhibition decreased to some extent. Differences between proportions of 6:4, 7:3 and 5:5 were significant for shoot or plant biomass, but non-significant for root biomass. Non-significant differences were observed for shoot, root, or plant biomass at proportions of 8:2 and 9:1 compared with 5:5 proportion.

Within crabgrass–switchgrass system, the inhibitory effects on switchgrass seedling growth increased as crabgrass seedling population increased from 1:9 to 5:5 proportion. The most significant inhibitory effects were observed when crabgrass seedling population was close to switchgrass seedling population, with biomass inhibitions of 50.5%, 60.3%, and 53.4% on shoot, root, and plant ($P<0.001$, $P=0.036$, $P=$...
0.008) at 5:5 proportion, respectively. From 6:4 to 9:1 proportion, the inhibition decreased to some extent. Differences were significant for shoot biomass, but non-significant for root biomass, at proportions of 6:4, 7:3, 8:2, and 9:1 compared with 5:5 proportion. Differences for plant biomass were significant at proportions of 6:4, 8:2, and 9:1 compared with 5:5 proportion, but non-significant between proportions of 7:3 and 5:5.

According to the above analysis, it was clear that the inhibitory effects on switchgrass seedling growth depended on the mixture proportion of weed and switchgrass. There was a strong competitive relationship between these two weed species and switchgrass when weed seedling population was close to switchgrass seedling population. The highest biomass inhibition was observed, on shoot at 4:6 proportion, and on root and plant at 5:5 proportion, respectively. No significant differences were observed between shoot, root, or plant biomass at proportions of 4:6 and 5:5. Hence, the subsequent experiment on root segregation of weeds and switchgrass was conducted at 5:5 proportion in glass jars.

**Switchgrass seedling growth under root segregation with weeds**

Redroot pigweed and crabgrass inhibited shoot (Fig. 5A), root (Fig. 5B), and plant (Fig. 5C) biomass of switchgrass seedlings significantly, at 5:5 proportion under root contact in glass jars.

Partial and full contacts lead to significant inhibition of redroot pigweed on switchgrass seedling growth in glass jars, with shoot biomass inhibition of 26.2% for partial contact 0.45, 37.6% for partial contact 30, and 49.2% for full contact (all at $P < 0.001$). Root biomass was reduced 22.30% ($P = 0.010$), 41.8% ($P < 0.001$), and 54.7% ($P < 0.001$), while plant biomass was reduced 25.0%, 38.9%, and 50.9% (all at $P < 0.001$), for partial contact 0.45, partial contact 30 and full contact, respectively. Differences in shoot, root, or plant biomass were significant under partial contacts 0.45 and 30, and full contact.

Partial and full contacts led to significant inhibition of crabgrass on switchgrass seedling growth in glass jars, with biomass inhibition of 20.7%, 32.3%, and 35.5% on shoot ($P = 0.013$, $P = 0.002$, $P < 0.001$), 19.8%, 41.2%, and 46.2% on root ($P = 0.016$, $P = 0.003$, $P < 0.001$), and 20.4%, 34.9, and 38.6% on plant ($P = 0.008$, $P = 0.001$, $P < 0.001$) for partial contact 0.45, partial contact 30 and full contact, respectively. Differences between partial contacts 0.45 and 30 were non-significant for shoot biomass, but significant for root or plant biomass. Shoot, root, or plant biomass differences were significant between partial contact 0.45 and full contact, but non-significant between partial contact 30 and full contact.

**Transplanted switchgrass growth with local weeds**

Local weeds inhibited tiller number, phytomer biomass and shoot biomass of transplanted switchgrass (Fig. 6) during the first and second growing season in the field at Xiaotangshan Station.

Plant height and shoot biomass increased markedly during the three growing seasons between 2014 and 2016. Plant height of control plants at the end of the 2014 growing season was 153.0 cm and reached
232.6 cm by the end of 2016. Shoot biomass of control plants increased from 597.2 g plant\(^{-1}\) in 2014 to 1676.8 g plant\(^{-1}\) in 2016 (Fig. 6A & B). Local weeds had no significant effect on plant height (Fig. 6A), but significantly reduced shoot biomass during the first and second growing seasons, with 46.2% and 21.0% inhibition, respectively (Fig. 6B).

Phytomer number remained relatively stable and was not affected significantly by local weeds during the first, second, and third growing seasons (Fig. 6D). On the other hand, tiller number and phytomer biomass increased markedly during the experimental period, and were inhibited by local weeds significantly during the first and second growing seasons, with 29.5% and 11.7% inhibition on tiller number (Fig. 6C), and 32.4% and 14.4% inhibition on phytomer biomass, respectively (Fig. 6E).

**Discussion**

**Allelopathic effects of weeds root aqueous extracts on switchgrass germination**

Weed allelopathy can alter crop physiological functions, such as their enzymatic activity, protein synthesis, respiration, and cell division and enlargement, and can thereby substantially reduce crop germination (Zohaib et al. 2016). For example, aqueous extracts of purple nutsedge (*Cyperus rotundus*) increased germination time and decreased the germination index of soybean (*Glycine max*) seedlings (Darmanti et al. 2015).

In this study, root aqueous extracts were used to investigate if redroot pigweed and crabgrass could affect switchgrass germination characteristics by allelopathy. Root aqueous extracts of both weeds affected switchgrass germination. It was found that root aqueous extracts of both weeds at low concentrations tested promoted switchgrass seed germination in Petri dishes. Root extracts of these weeds at 0.005 g·mL\(^{-1}\) concentration significantly increased switchgrass plumule length, and root extract of crabgrass at 0.01 g·mL\(^{-1}\) promoted switchgrass radicle length (Fig. 3B & C).

On the contrary, root aqueous extracts of both weeds at high concentrations had inhibitory effects on switchgrass germination in Petri dishes. Germination rate of switchgrass was significantly reduced by redroot pigweed extracts at concentrations of 0.02, 0.05, and 0.1 g·mL\(^{-1}\), and by crabgrass extracts at concentrations of 0.05 and 0.1 g·mL\(^{-1}\). Switchgrass plumule length was significantly reduced by redroot pigweed extracts at concentrations of 0.02, 0.05, and 0.1 g·mL\(^{-1}\), and by crabgrass extracts at 0.1 g·mL\(^{-1}\) concentration, while switchgrass radicle length was significantly reduced by redroot pigweed extracts at concentrations of 0.05 and 0.1 g·mL\(^{-1}\), and by crabgrass extracts at 0.1 g·mL\(^{-1}\) concentration, respectively (Fig. 3A, B, C).

These findings strongly indicate the role of plant allelopathy in interactions among these plants. The inhibitory effects observed might be the result of the release of allelopathic compounds, or the increase of root extract concentrations in the soil. More research is required to further identify the mechanism of
allelopathic promotion / inhibition of weeds on switchgrass germination. In future research, allelopathic effects of root aqueous extracts of other local weeds on switchgrass germination will be evaluated to investigate physiological and ecological processes involved.

Inhibitory effects of weeds on switchgrass seedling growth

Weeds can inhibit crop seedlings growth by various mechanisms, such as by releasing allelochemicals into soil or decreasing available nutrients in the soil (Batish et al. 2005). Previous studies found that various weed species significantly inhibited wheat seedling growth (Dongre and Singh 2007). Additionally, purple nutsedge extracts decreased the weight and length of soybean seedlings (Darmanti et al. 2015), and ragweed parthenium (Parthenium hysterophorus) residue significantly inhibited radish (Raphanus sativus) and mustard (Brassica campestris) seedling length and dry weight (Batish et al. 2005). Similarly, aqueous extracts and leachates of nutgrass (Cyperus rotundus) inhibited rice seedlings growth (Quayyum et al. 2000).

In this study, redroot pigweed and crabgrass decreased switchgrass biomass in glass jars. The inhibition depended on the proportion of weed and switchgrass seedlings in the replacement experiment. A strong adverse effect from these two weed species on switchgrass occurred at 5:5 proportion. Allelochemical response could be strongly related to neighboring plants. These effects were associated with the switchgrass density and in the subsequent root segregation experiment, 5:5 proportion was used according to these results.

Another factor that can influence the impact of allelopathy is the presence of mycorrhizal fungi. The relationship between allelopathic effects and mycorrhizal fungi is complex as mycorrhizal fungi have been found to serve as both targets and mediators of allelopathic effects between plants, with strong positive and negative effects previously detected (Cipollini et al. 2012). Arbuscular mycorrhizal fungi were found to lessen the allelopathic effects of an invasive plant, garlic mustard (Alliaria petiolata), on a herbaceous native North American plant, pale jewelweed (Impatiens pallida) (Barto et al. 2010), but the presence of ectomycorrhizal fungi substantially reduced nitrogen uptake in Scots pine (Pinus sylvestris) (Nilsson et al. 1993). Moreover, there is growing evidence that allelopathic plants can directly affect mycorrhizal fungi. For example, garlic mustard extracts were found to affect spore generation, growth, infection rates, and community structure in arbuscular mycorrhizal fungi (Cipollini et al. 2012).

In this study, shoot and root biomasses of switchgrass grown with these two weed species under partial contacts 30 was further reduced significantly, relative to that under partial contacts 0.45, except shoot biomass with crabgrass (Fig. 5), which indicated that allelopathic effects of redroot pigweed and crabgrass on switchgrass were mediated by both chemical signals and soil mycorrhizal hyphae. Switchgrass biomass under full contact with redroot pigweed decreased significantly, relative to that under partial contacts 30. While, with crabgrass, there were non-significant differences between switchgrass biomass under contacts 30 and full contact. These results indicated that resource competition of redroot pigweed was stronger than that of crabgrass in weed–switchgrass interaction.
In future research, effects of redroot pigweed, crabgrass, and other local weeds on physiological and ecological processes of switchgrass seedlings will be assessed. A comprehensive analysis of weed–switchgrass interaction need to be further studied, including possible aerial allelopathic effects, as well as interactions of resource competition and allelopathy between seedlings of weed species and switchgrass.

**Inhibitory effects of local weeds on transplanted switchgrass growth**

The presence of weeds can be problematic for crop production, especially because some weeds inhibit crop growth. Previous studies found that annual weed residues inhibited growth and nutrient uptake of maize (*Zea mays*) and soybeans (Bhowmik and Doll 1984), and aqueous extracts of various weeds suppressed plumule and radicle length of legumes and cereal crops (Bhatt et al. 2001).

In this study, the inhibitory effects of redroot pigweed, crabgrass, and other local weeds on several growth characteristics of transplanted switchgrass seedlings were significant, and these inhibitory effects decreased continuously by each growing season. During the first and second growing seasons, tiller number, phytomer biomass and shoot biomass were inhibited significantly by local weeds. During the third growing season, these inhibitory effects were non-significant.

In future research, long-term effects of redroot pigweed, crabgrass, and other local weeds on physiological and ecological processes of switchgrass seedlings will be evaluated in the field. Moreover, the degree of impact of resource competition and allelopathy between redroot pigweed, crabgrass, and other local weeds and switchgrass should be further evaluated. Such studies would lay a foundation for ecological control of weeds in long-term switchgrass cultivation.

**Allelochemicals of redroot pigweed and crabgrass**

Plant allelopathy is mostly mediated by plant secondary metabolites, namely allelochemicals (Kong et al. 2018). To fully reveal redroot pigweed and crabgrass allelopathy, allelochemicals need to be identified. Some redroot pigweed and crabgrass allelochemicals have been identified in previous studies. Docosane, triacontane, silane, and ethoxytrimethyl were identified as redroot pigweed allelochemicals, which reduced cucumber and wheat growth in a hydroponic greenhouse culture (Bakhshayeshan-Agdam et al. 2019). Alternatively, three crabgrass allelochemicals, veratric acid, maltol, and (−)-loliolide, were found to produce changes in the soil microbial community that resulted in inhibited growth of wheat, maize, and soybeans, and reduced microbial carbon (Zhou et al. 2013a). Moreover, 52 compounds were identified as crabgrass essential oil constituents and inhibited growth and allelochemical production of wheat (Zhou et al. 2013b).

In future research, allelopathic effects of these compounds and their modes of action on switchgrass germination and growth should be evaluated to enhance our understanding of allelopathy and its mechanism during weed–crop interaction.

**Conclusions**
Switchgrass germination was inhibited significantly by root aqueous extracts of weeds at high concentrations. Redroot pigweed extracts at concentrations of 0.05 and 0.1 g·mL\(^{-1}\) and crabgrass extracts at 0.1 g·mL\(^{-1}\) concentration significantly reduced switchgrass germination. At the same concentration, root aqueous extracts of redroot pigweed led to more significant allelopathic inhibition on switchgrass germination than those of crabgrass.

Significant inhibition on shoot, root and plant biomass of switchgrass seedlings occurred at 5:5 proportion of these two weed species and switchgrass in glass jars. This experiment indicated that allelopathy played a crucial role in the weed-crop interaction, which was further enhanced by soil mycorrhizal hyphae. The inhibition of redroot pigweed on switchgrass seedling growth was more significant than that of crabgrass in glass jars. Redroot pigweed grown in full contact with switchgrass reduced its growth more than the two partial contact treatments indicating strong resource competition of this weed.

Tiller number, phytomer biomass and shoot biomass of switchgrass were inhibited significantly by local weeds in the field and, this effect decreased continuously with increasing growing season. During the third growing season, local weeds did not inhibit switchgrass growth significantly.

On the whole, this study produced more evidence on the allelopathic effects of redroot pigweed and crabgrass on switchgrass germination in Petri dishes and their inhibitory effects on seedling growth. Field results showed adverse effects of local weeds on switchgrass growth during the first and second growing seasons. As switchgrass area under cultivation is increasing, these results can help in decision making in regard to timing of weed management in the field. The study enhances our understanding of weed-crop interactions at germination and seedling stage.

**Declareds**

**Conflicts of interest/Competing interests**

There are no conflicts/competing of interest.

**Availability of data and material (data transparency)**

We declare that materials described in the manuscript, including all relevant raw data, will be freely available to any scientist wishing to use them for non-commercial purposes, without breaching participant confidentiality.

**Code availability (software application or custom code)**

No previously unreported software application or custom code was described in our manuscript.

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**Figures**
Figure 1

Schematic diagram of experimental design on mixture determination for weed–switchgrass system in glass jars

Figure 2

Schematic diagram of experimental design on root segregation of weed and switchgrass at 5:5 proportion in glass jars
Figure 3

Allelopathic effects of weed root aqueous extracts on switchgrass germination in Petri dishes. ***, **, and * indicate significant differences between each treatment and control for the same germination characteristic within each weed species, at P < 0.001, P < 0.01, and P < 0.05, respectively, according to independent t-test.
Figure 4

Switchgrass seedling growth at different proportions with weeds in glass jars. Control: a monoculture of ten switchgrass seedlings. Bars with different letters indicate significant differences (P < 0.05) for the same seedling characteristic within each weed species, according to analysis of variance and multiple comparisons.
Figure 5

Switchgrass seedling growth at 5:5 proportion with weeds under root segregation in glass jars. Bars with different letters indicate significant differences (P < 0.05) for the same seedling growth characteristic within each weed species, according to analysis of variance and multiple comparisons.
Figure 6

Inhibitory effects of local weeds on transplanted switchgrass growth in the field. * indicates significant differences between treatment and control for the same growth characteristic during the same growing season, at $P < 0.05$, according to independent t-test.