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# Plant Suppression and Termination Methods to Maintain Intermediate Wheatgrass (*Thinopyrum intermedium*) Grain Yield

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**Abstract:** Intermediate wheatgrass (*Thinopyrum intermedium* (Host) Barkworth & D.R. Dewey; IWG) is a perennial sod-forming grass undergoing domesticated for use as a dual-use grain and forage crop with potential environmental benefits. IWG plant populations increase with stand age, which has been associated with reductions in grain yields after the second production year, thus management techniques are needed to maintain grain yields over time. We measured the effects of two between-row plant termination methods (cultivation and herbicide application) and two within-row suppression methods (burning and mowing), applied at different IWG physiological stages during the growing season. We measured IWG grain and straw yield, root biomass, and weed biomass. Treatments were initiated after the second year of grain harvest and applied for two consecutive years in southeast Minnesota. Grain yields were highest in production year 2 preceding any treatment application and declined in years 3 and 4 by 82% and 57% compared to year 2, respectively, across all management treatments. Termination methods reduced between-row IWG biomass and grain by up to 82% and 91% compared to the control but had no effect on within-row or total grain yield. Fall burning suppression treatments mitigated the negative effects of some termination treatments on grain yield and increased total straw yield. Spring mowing suppression treatments reduced grain and straw yield by 42% and 34%, respectively, compared to the control. Controls had minimal weed biomass while the termination treatments increased weed biomass, especially termination treatments that included herbicide application. No treatments sustained grain yields, but positive effects of some treatments were observed on total biomass and weeds and could be considered by growers.

**Keywords:** Kernza; perennial grains; agroecology; sustainable agriculture; plant competition

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## 1. Introduction

Intermediate wheatgrass (*Thinopyrum intermedium* (Host) Barkworth & D.R. Dewey; IWG) is a sod-forming perennial grass that is being domesticated to serve as a perennial grain crop [1,2]. One agronomic characteristic that limits the long-term viability of this perennial crop is that grain yields decline with stand age, and the rate of grain yield decline has been shown to vary across environments [3,4]. For example, Jungers et al. [3] found that grain yields declined during the third production year after fall seeding while spring seeded stands declined during the second production year. A potential physiologically mediated mechanism driving yield decline with stand age could be related to plant population density and competition among tillers [5]. IWG can reproduce vegetatively through tillering and rhizomatous growth, and increases in plant density have been attributed to decreasing grain yield via effects on light quantity and quality [3,6,7]. Increased population density from vegetative reproduction, along with potential seedling recruitment from shattered seeds, can result in dense root biomass, thus increasing competition for water and nutrients belowground [8].

A mechanical or chemical disturbance between planted rows can prevent increases in plant population density and subsequent competition for resources, which could sustain grain yields as stands age. Law et al. [5] found that fall strip tillage between IWG following the third harvest year increased grain yields in the following year by 61% compared to an untreated control by increasing the number of spikes per plant. Inter-row termination methods that do not require soil disturbance could better maintain soil structure, organic matter, and microbial diversity and activity [9]. Glyphosate has been used to manage plant populations for seed production of other perennial grass species such as Kentucky bluegrass (*Poa pratensis*) [10]. Both mechanical and chemical termination methods can promote weed growth by allowing more light to access the soil surface, a potential drawback to using these methods for maintaining perennial grain yields. Cultivation also moves weed seeds closer to the soil surface and can increase the temperature of the soil surface by breaking up crop residues, both further promoting weed growth.

Non-lethal disturbance of within-row plants has the potential to affect grain yields of perennial grasses varying in stand age. Mowing and burning initially suppress plant growth but can lead to enhanced longer-term productivity of perennial grasses [11,12]. Burning crop residue following seed harvest of perennial grasses was a common management practice to maintain seed yields, but concerns for air quality have resulted in regulations that restrict residue burning in North America [13,14]. Burning is effective because it consumes accumulated plant residue from previous growing seasons increasing light access to plant crowns and mineralizing plant tissues that may provide minerals such as K and Ca. Mowing has been utilized as an alternative to burning and can have similar effects on biomass productivity of perennial plants [15].

The goals of this study were to (1) determine the effects of between-row termination methods and application timings on IWG yields, root biomass, and weed biomass, and (2) determine the effects of within-row suppression methods and application timings on IWG yields, root biomass, and weed biomass.

## 2. Materials and Methods

### *2.1. Study Area*

The experiment was conducted at Rosemount, MN (44°71′ N, 93°7′ W) from 2016 to 2019. Soil type is Tallula silt loam, 6% slope, eroded coarse-silty, mixed, superactive, mesic Typic Hapludolls. The mean annual air temperature is approximately 8.3 °C and mean annual precipitation is 813 mm. Seed was from an IWG population resulting from the fourth cycle of selection at The Land Institute (Salina, KS, USA). Seed was sown in September 2015 using a no-till drill (Truax Company, Inc., New Hope, MN, USA) following soybean (*Glycine max* L.) with openers adjusted to achieve a planting configuration of alternating 40.6 cm and 61 cm spacing between rows (Figure 1). At the time of treatment application in fall of 2017, seeded rows were distinguishable but IWG plants had spread into the between-row areas. The experiment was fertilized each spring with 80 kg N ha$^{-1}$ as urea (Table 1).

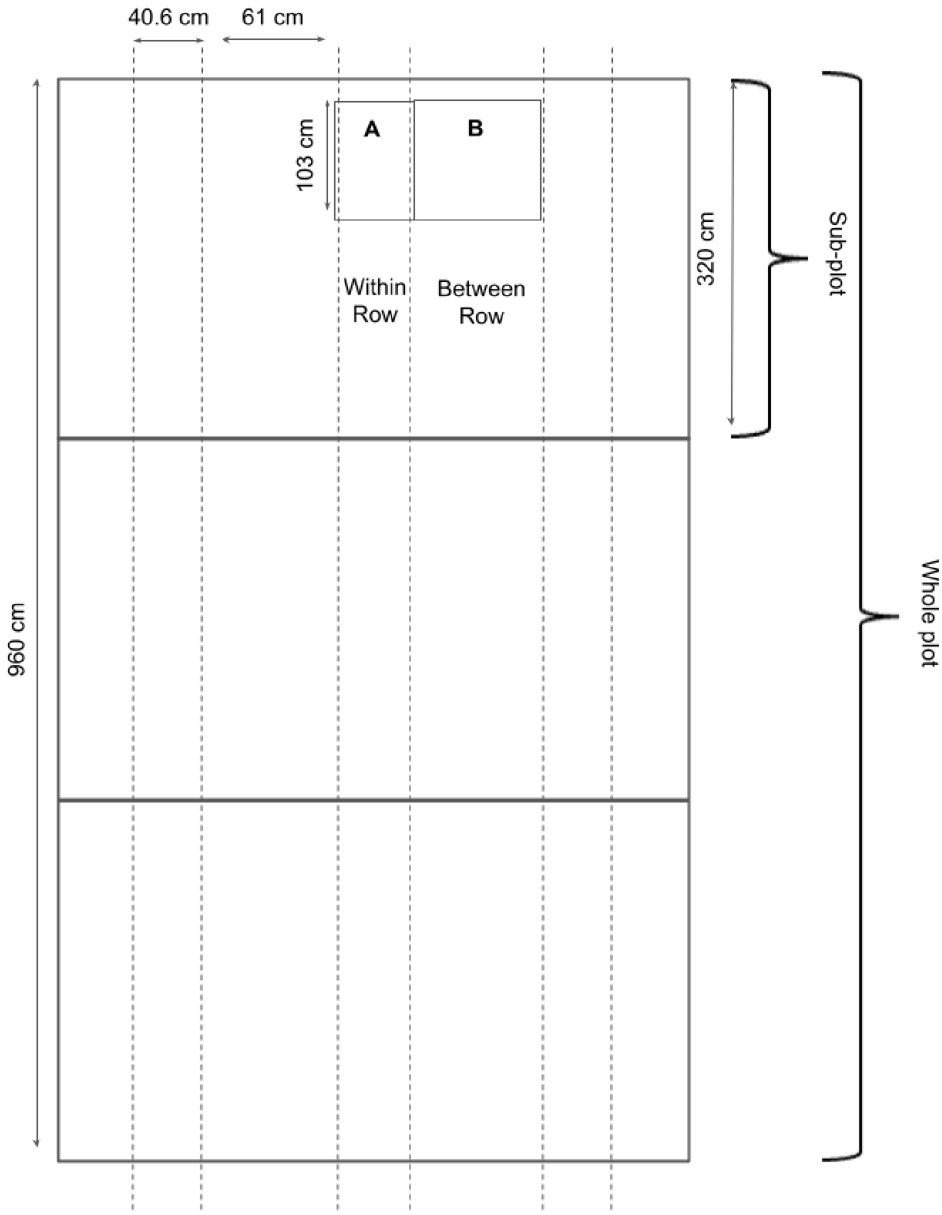


**Figure 1.** A diagram indicating three sub-plots within the main-plot boundary (thick solid line) positioned in relation to rows sown with IWG (dashed lines) and labeled with within and between row spaces. Boxes labeled A and B represent the sampled area within and between rows, respectively within sub-plots. Objects are not to scale.

**Table 1.** Timing of field study management activities and treatment application from 2017–2019.

| | 2017 | 2018 | 2019 |
|---|---|---|---|
| Spring cultivation | – | 17 April | 14 May |
| Spring herbicide | – | 18 April | 15 May |
| Urea applied (80 kg N ha$^{-1}$) | 24 April | 15 April | 26 April |
| Spring forage harvest | 30 May | 23 May | 28 May |
| Grain and straw harvest | 22 August | 10 August | 14 August |
| Fall herbicide | 20 October | 18 October | – |
| Fall cultivation | 26 October | 22 October | – |
| Fall mow-burn | 17 November | 15 November | – |

*2.2. Experimental Design*

Treatments were randomly assigned to plots and applied in the fall of 2017 to the spring of 2019 in a split-plot design with four replications. There were seven main-plot treatments. Main-plot treatments received one of two between-row termination treatments—cultivation or glyphosate (N-(phosphonomethyl)glycine) herbicide application—each applied at three different timings (spring, fall, or spring + fall). A control without any between-row termination was included. Main plots were divided into three sub-plots, each receiving one of the following suppression treatments—a spring mowing, a fall burning, or a no-suppression control. Experimental units (sub-plots) were 3 m wide with 6 planted rows and 3.2 m long (Figure 1). All within-row biomass and grain yield samples were collected from the two central rows.

Between-row cultivation was achieved using a power take-off tool bar with three rotary units (Multivator®, Ford Distributing, Columbus, OH, USA), in which each set of rotary units tilled a zone 40 cm wide and approximately 10 cm deep. Rotary units were spaced to leave 61 cm wide strips of IWG. Glyphosate herbicide was applied at a rate of 1.7 kg active ingredient $ha^{-1}$ with a $CO_2$ backpack sprayer. Similar to the cultivation treatments, 40 cm wide strips were sprayed with herbicide with 61 cm strips of living IWG between strips. Custom built hoods were attached to each spray nozzle to avoid herbicide drift to planted IWG rows. Main plots included two termination strips and three living IWG strips. Suppression treatments were applied to split plots (Figure 1). Burning was performed in November of each year using a butane fueled flame-weeder. Aluminum siding pieces were used as flame barriers to contain the fire within the treatment area. A Carter forage harvester was used to apply the spring mowing treatment and remove all aboveground biomass within plots to a stubble height of 8 cm. Additional treatment details are listed in Table 1.

*2.3. Grain and Biomass Sampling*

IWG grain and straw biomass were collected in early August at physiological maturity when seeds had approximately 40% moisture content [16]. Yields were measured prior to when renovation treatments were imposed in 2017, and then after renovation treatments were imposed in 2018 and 2019. Data were collected from within rows after treatment application (within-row data, WR) and between rows (between-row data, BR; see Figure 1 for details). Plant biomass was collected by hand to a height of ~10 cm. Within-row samples were collected by cutting two 103 cm lengths of parallel rows within each experimental unit—equivalent to an area of 0.42 $m^2$. Between-row samples were harvested from plants growing in the 61 cm space between planted IWG rows. The following procedures to determine yields were conducted on both WR and BR samples separately. Weeds were separated from IWG in each experimental unit in 2019, and both weeds and IWG were dried at 60 °C for five days before weighing again for dry matter yield determination. After drying, IWG seeds were threshed using a laboratory grain thresher (Wintersteiger LD-350; Salt Lake City, UT, USA) following methods described by Frahm et al. [17]. Grain was sieved and weighed for dry matter yield determination. Grain yield was subtracted from total IWG biomass yield to determine straw yield. Weed, IWG straw, and IWG grain were measured for WR and BR separately and then summed and reported as total yields.

Intermediate wheatgrass spring forage yield was determined by harvesting total biomass (IWG and weeds) from both WR and BR areas immediately prior to the mowing treatment each spring using plant collection, drying, and weighing procedures described above.

Root biomass was collected from a subset of treatments. Soil cores were taken from both WR and BR areas in the control sub-plots within the cultivated termination treatment and the untreated controls. A hydraulic Gidding's soil probe (inside diameter = 3.8 cm) was used to extract two WR and two BR root samples to a depth of 60 cm. Root samples were washed of soil using a hydropneumatic elutriation system [18], cleaned of sand and other non-root debris, and then dried at 60 °C for five days before being weighed.

### *2.4. Statistical Analysis*

All analyses were conducted with R version 3.4.4 (R Core Team 2018). Mixed-effects models were used to test for effects of main-plot treatments (termination), sub-plot treatments (suppression), year, and all potential two-way and three-way interactions on grain, straw, and spring forage biomass. This original model also included main-plot treatments nested within blocks as random effects. For all response variables, a two-way interaction between main-plot treatment and year or between sub-plot treatment and year were always observed. Therefore, data were analyzed for each year separately using linear mixed-effects models with restricted maximum likelihood to conduct two-way analysis of variance (ANOVA) tests with block as a random effect and main-plot treatments nested within blocks to account for pseudo-replication with a split-plot design. Termination treatment, suppression treatment, and their interaction were modeled as fixed effects. Degrees of freedom were lower for root biomass data because root data were only collected from a subset of main treatments across control split plot treatments. Means were compared by obtaining estimated marginal means (package, code: emmeans, emmeans; Lenth 2016) and adjusted for Tukey's honestly significant difference test. Results from ANOVA tests are listed in Table 2. Assumptions of normally distributed residuals, independence of error and constant variance were checked for each linear model using qq-plots, histograms of residuals, and plots of the residuals against fitted values.

**Table 2.** Analysis of variance (ANOVA) test results for the effects of termination (T) and suppression (S) treatments on grain, straw, and spring forage yield, weed biomass, and root biomass between- and within-rows of planted intermediate wheatgrass (IWG). Root sampling depth from 0–60 cm. Spring forage yield was only measured in mowed suppression sub plots while roots were only measured in the control suppression sub plots. Weeds were not measured in 2018.

| Year. | Source of Variation | Grain (Total) | Grain (WR) | Grain (BR) | Straw (Total) | Straw (WR) | Straw (BR) | Spring Forage (Total) | Weeds (Total) | Weeds (WR) | Weeds (BR) | Roots (WR) | Roots (BR) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 2018 | T | - | - | * | . | * | *** | - | NA | NA | NA | * | - |
| | S | - | - | - | *** | *** | - | NA | NA | NA | NA | NA | NA |
| | T × S | - | - | - | - | - | - | NA | NA | NA | NA | NA | NA |
| 2019 | T | * | * | ** | - | . | ** | * | *** | ** | *** | - | * |
| | S | *** | *** | - | *** | *** | - | NA | ** | ** | * | NA | NA |
| | T × S | * | * | - | *** | *** | - | NA | - | - | - | NA | NA |

. = 0.05 < $p$ < 0.10; * = 0.01 < $p$ < 0.05; ** = 0.001 < $p$ < 0.01; *** = $p$ < 0.001; - = $p$ > 0.05; NA = Not measured.

## 3. Results

### *3.1. Grain Yield*

Total grain yields averaged 391 kg ha$^{-1}$ in 2017, declined to 70 kg ha$^{-1}$ in 2018, and then increased to 168 kg ha$^{-1}$ in 2019. Termination and suppression treatments did not affect total grain yields in 2018 but did in 2019. Within row grain yields were also not affected by treatment in 2018 but BR grain yields that remained following termination treatments did vary among termination treatments (Table 2). In 2018, BR grain yields were significantly greater in the fall cultivated treatment (4.5 ± 4.0 kg ha$^{-1}$) compared to the spring herbicide (0.1 ± 0.3 kg ha$^{-1}$) and spring + fall herbicide (0.1 ± 0.1 kg ha$^{-1}$) applications. In 2019, there were significant main effects of termination treatment, suppression treatment, and their interaction on total grain yield and WR grain yield, while BR grain yields only varied by termination treatment (Table 2). For total and WR grain yield in 2019, no main termination treatment means differed from the untreated control, but the fall herbicide treatment did reduce yields compared to spring herbicide treatment (Table 3). The mowing suppression treatment reduced total and WR grain yields compared to the control and the burning treatment in 2019. Differences in WR and total grain yields were significant when comparing all combinations of termination and suppression treatments, but no combination resulted in higher grain yields compared to the control (Table 4). However, the spring herbicide termination treatment combined with the burn suppression treatment did increase grain yields compared to fall herbicide with and without mowing, and spring herbicide with mowing (Table 4). For 2019 BR grain yields, no interaction between management treatments occurred and grain yield from control termination plots was significantly greater than all other termination treatments except for fall cultivation (Table 2).

**Table 3.** Mean (±SE, $n$ = 4) of grain and straw yields by termination treatments (averaged over suppression treatments) and suppression treatments (averaged over termination treatments) in 2019. Means sharing a letter within a column are similar ($p < 0.05$).

| | Total Grain | WR Grain | BR Grain | Total Straw | WR Straw | BR Straw |
|---|---|---|---|---|---|---|
| | kg ha$^{-1}$ | | | | | |
| Termination treatment | | | | | | |
| Fall cultivation | 213 (26) $^{ab}$ | 185 (24) $^{ab}$ | 28 (10) $^{bc}$ | 2970 (356) | 2623 (294) | 347 (104) $^{ab}$ |
| Spring herbicide | 220 (38) $^{b}$ | 217 (38) $^{b}$ | 3 (3) $^{a}$ | 3574 (506) | 3470 (517) | 103 (26) $^{a}$ |
| Spring cultivation | 144 (16) $^{ab}$ | 140 (17) $^{ab}$ | 5 (2) $^{a}$ | 2827 (197) | 2720 (211) | 107 (31) $^{a}$ |
| Fall + spring herbicide | 147 (26) $^{ab}$ | 143 (26) $^{ab}$ | 4 (2) $^{a}$ | 2294 (417) | 2137 (364) | 156 (100) $^{a}$ |
| Fall + spring cultivation | 182 (23) $^{ab}$ | 178 (23) $^{ab}$ | 4 (2) $^{a}$ | 2975 (191) | 2878 (208) | 97 (31) $^{a}$ |
| Control | 175 (18) $^{ab}$ | 140 (15) $^{ab}$ | 35 (5) $^{c}$ | 2758 (210) | 2215 (183) | 544 (75) $^{b}$ |
| Fall herbicide | 93 (18) $^{a}$ | 87 (16) $^{a}$ | 6 (3) $^{ab}$ | 2068 (310) | 1974 (300) | 94 (28) $^{a}$ |
| Suppression treatment | | | | | | |
| Fall burn | 213 (17) $^{b}$ | 199 (16) $^{b}$ | 14 (5) | 3399 (209) $^{c}$ | 3133 (195) $^{b}$ | 266 (66) |
| Control | 184 (17) $^{b}$ | 174 (17) $^{b}$ | 10 (3) | 2985 (230) $^{b}$ | 2812 (227) $^{b}$ | 173 (42) |
| Spring mow | 106 (10) $^{a}$ | 94 (9) $^{a}$ | 12 (3) | 1958 (143) $^{a}$ | 1777 (143) $^{a}$ | 181 (42) |

**Table 4.** Mean within-row and total grain and straw yield (±SE, $n$ = 4) for suppression and termination treatment combinations in 2019. Different lower-case letters denote statistical significances between each suppression and termination treatment combination at $p < 0.05$.

| | Termination | Within-Row Grain | Total Grain | Within-Row Straw | Total Straw |
|---|---|---|---|---|---|
| | | kg ha$^{-1}$ | | | |
| Burn | Control | 155 (30) $^{abc}$ | 185 (35) $^{abcd}$ | 2509 (328) $^{abcde}$ | 3005 (349) $^{abcde}$ |
| | Fall cultivation | 228 (24) $^{abc}$ | 272 (44) $^{bcd}$ | 3335 (369) $^{b\,de}$ | 3850 (651) $^{b\,de}$ |
| | Fall herbicide | 140 (25) $^{abc}$ | 150 (30) $^{abcd}$ | 2382 (618) $^{abcde}$ | 2483 (654) $^{abcde}$ |
| | Fall + Spring cultivation | 173 (51) $^{abc}$ | 177 (51) $^{abcd}$ | 2950 (452) $^{abcde}$ | 3088 (393) $^{abcde}$ |
| | Fall + Spring herbicide | 210 (31) $^{abc}$ | 218 (34) $^{abcd}$ | 3368 (376) $^{cde}$ | 3749 (545) $^{cde}$ |
| | Spring cultivation | 174 (37) $^{abc}$ | 177 (37) $^{abcd}$ | 2902 (416) $^{abcde}$ | 3040 (352) $^{abcde}$ |
| | Spring herbicide | 310 (48) $^{c}$ | 312 (48) $^{d}$ | 4482 (513) $^{e}$ | 4574 (484) $^{e}$ |
| Control | Control | 167 (11) $^{abc}$ | 208 (16) $^{abcd}$ | 2484 (256) $^{abcde}$ | 3024 (356) $^{abcde}$ |
| | Fall cultivation | 221 (49) $^{abc}$ | 231 (47) $^{abcd}$ | 3107 (235) $^{b\,de}$ | 3360 (280) $^{b\,de}$ |
| | Fall herbicide | 75 (20) $^{ab}$ | 80 (23) $^{abc}$ | 1892 (343) $^{abcd}$ | 2001 (395) $^{abcde}$ |
| | Fall + Spring cultivation | 230 (33) $^{abc}$ | 235 (33) $^{abcd}$ | 3314 (282) $^{abcde}$ | 3384 (237) $^{abcde}$ |
| | Fall + Spring herbicide | 127 (41) $^{abc}$ | 128 (42) $^{abcd}$ | 1625 (572) $^{ab}$ | 1673 (582) $^{ab}$ |
| | Spring cultivation | 133 (26) $^{abc}$ | 141 (22) $^{abcd}$ | 2788 (422) $^{abcde}$ | 2908 (420) $^{abcde}$ |
| | Spring herbicide | 264 (58) $^{bc}$ | 265 (58) $^{cd}$ | 4475 (768) $^{e}$ | 4547 (770) $^{e}$ |
| Mow | Control | 99 (24) $^{ab}$ | 134 (33) $^{abcd}$ | 1651 (189) $^{abcd}$ | 2246 (328) $^{abcde}$ |
| | Fall cultivation | 106 (16) $^{ab}$ | 136 (13) $^{abcd}$ | 1427 (184) $^{a\,c}$ | 1699 (207) $^{a\,c}$ |
| | Fall herbicide | 45 (18) $^{a}$ | 50 (14) $^{a}$ | 1648 (632) $^{abcd}$ | 1718 (610) $^{abcd}$ |
| | Fall + Spring cultivation | 130 (15) $^{abc}$ | 133 (14) $^{abcd}$ | 2370 (200) $^{abcde}$ | 2454 (204) $^{abcde}$ |
| | Fall + Spring herbicide | 92 (46) $^{ab}$ | 94 (46) $^{abc}$ | 1419 (476) $^{ab}$ | 1460 (462) $^{ab}$ |
| | Spring cultivation | 111 (17) $^{ab}$ | 115 (15) $^{abcd}$ | 2469 (323) $^{abcde}$ | 2532 (284) $^{abcde}$ |
| | Spring herbicide | 78 (12) $^{a}$ | 82 (12) $^{ab}$ | 1454 (227) $^{abcd}$ | 1600 (193) $^{abcd}$ |

### *3.2. Straw Yield*

Total straw yields were affected by suppression treatment but not termination treatment in 2018 (Table 2) where mowing reduced straw yields compared to burning and the control. There was a significant effect of termination treatment on WR straw yields in 2018

($p$ = 0.034) although a mean comparison test did not detect any pairwise differences in yields. There was a significant effect of suppression treatment on WR straw yields in which mowing reduced yields compared to the control and burn treatments. Between-row straw yields were similar among the control, fall herbicide and fall cultivation treatments in 2018, while all other termination treatments reduced BR straw yields compared to the control.

A significant interaction between termination and suppression treatments was observed for total and WR straw yields in 2019 (Table 2). For 2019 total and WR straw production, the mowing suppression treatment resulted in significantly lower yields than the control and burn (Table 3). The BR straw yield varied by termination treatment in 2019 where yield in the control was significantly greater than all other treatments except for the fall cultivation (Table 3). The control plots for BR 2019 straw yielded significantly more than all others except for fall cultivated plots (Table 3). No combination of termination and suppression treatments resulted in greater straw yields than the control, but similar to grain yields, a spring herbicide with burn did result in greater WR and total straw yields compared to some other treatment combinations (Table 4).

### *3.3. Spring Forage Yield*

Since forage biomass was harvested only from the mowing suppression treatments, data were analyzed to determine the effects of termination treatments. A termination treatment effect was only observed in 2019 (Table 2), where mowed spring forage was greater in the spring herbicide and fall cultivation treatments compared to the fall herbicide; however, spring forage from all termination treatments was similar to the control.

### *3.4. Weed Biomass*

There was a main effect of both termination and suppression treatments on total, WR, and BR weeds in 2019 (Table 2). Compared to the control, total weed biomass was greater in all the herbicide termination treatments and in the mowed suppression treatments (Table 5). The fall + spring herbicide treatment had significantly more WR weeds than all other treatments (including the control) except for the fall herbicide treatment (Table 5). Mowing increased WR weeds compared to the burned treatments (Table 5). Similar to trends observed with total weeds, BR weeds were greater in the herbicide treatments compared to the control, with the exception of the spring herbicide treatment that was similar to the control. Mowing increased BR weeds compared to the burning treatment, and both were similar to the control (Table 5).

**Table 5.** Mean (±SE) of weed biomass within (WR) and between (BR) planted IWG rows by termination treatments (averaged over suppression treatments) and suppression treatments (averaged over termination treatments) in 2019. Different lower-case letters denote statistical significances between each suppression and termination treatment combination at $p < 0.05$.

| | Total Weeds | Weeds (WR) | Weeds (BR) |
|---|---|---|---|
| Termination treatment | | g plot$^{-1}$ | |
| Fall cultivation | 44 (15) $^{ab}$ | 18 (8) $^{a}$ | 25 (8) $^{a}$ |
| Spring herbicide | 145 (33) $^{bc}$ | 33 (9) $^{a}$ | 112 (27) $^{ab}$ |
| Spring cultivation | 70 (21) $^{ab}$ | 17 (9) $^{a}$ | 53 (15) $^{a}$ |
| Fall + spring herbicide | 328 (38) $^{d}$ | 108 (27) $^{b}$ | 221 (23) $^{c}$ |
| Fall + spring cultivation | 70 (18) $^{ab}$ | 18 (9) $^{a}$ | 52 (10) $^{a}$ |
| Control | 22 (10) $^{a}$ | 14 (7) $^{a}$ | 9 (4) $^{a}$ |
| Fall herbicide | 223 (44) $^{cd}$ | 51 (16) $^{ab}$ | 172 (37) $^{bc}$ |
| Suppression treatment | | | |
| Fall burn | 94 (21) $^{a}$ | 16 (5) $^{a}$ | 78 (18) $^{a}$ |
| Control | 118 (26) $^{a}$ | 38 (11) $^{ab}$ | 80 (18) $^{ab}$ |
| Spring mow | 174 (31) $^{b}$ | 56 (13) $^{b}$ | 118 (20) $^{b}$ |

### *3.5. Root Biomass*

Root biomass to a depth of 60 cm was collected only from the cultivated termination treatments and the control. There was a significant effect of termination treatments on WR roots in 2018 (Table 2), where WR root biomass averaged 8.1 Mg ha$^{-1}$ for the control, spring + fall cultivation and fall cultivation treatments, whereas the spring cultivation treatment averaged 4.2 Mg ha$^{-1}$. There was an effect of termination treatment on BR roots in 2019 ($p$ = 0.04, Table 2) where BR root biomass averaged 1.5 Mg ha$^{-1}$ for the control, fall cultivation, and spring cultivation treatments, whereas the spring + fall cultivation treatment averaged 0.5 Mg ha$^{-1}$.

## 4. Discussion

Perennial grain crops like intermediate wheatgrass are being developed to provide environmental benefits such as soil conservation, nutrient retention, and soil carbon sequestration because their perennial nature limits soil disturbance and promotes below-ground biomass and nutrient capture [19,20]. However, declining grain yields with stand age reduce the potential for profitability after the second or third year, which will motivate growers to terminate stands thus reducing environmental benefits. Identifying agronomic methods that prevent grain yield declines with stand age can prolong the duration of IWG in rotations and therefore reduce tillage and subsequent environmental issues such as soil erosion.

As IWG stands age, plant population increases as rhizomes and shattered seeds produce seedlings between planted rows [21]. Researchers have hypothesized that an increase in IWG plant population alters light penetration, which reduces seed head induction of established plants in fall [7]. Law et al. [5] and Pinto et al. [7] both showed that mechanically thinning IWG fields in fall increased the proportion of reproductive tillers (i.e., tillers that produced seed heads) compared to controls. However, only Law et al. [5] found evidence that more reproductive tillers manifested in higher grain yields. We did not measure the effects of termination and suppression treatments on yield components such as reproductive tillers, and although the mean total grain yield varied by treatment, no treatment was different from the control. This result is similar to what was found by Pinto et al. [7], who investigated the effects of thinning and residue management on IWG grain yields in the second year of production. In our study, termination treatments were very effective at terminating plants in between rows (evident by the low BR grain and straw biomass yields). It is important to note that the BR IWG plants that were terminated were contributing grain to total grain yields. In fact, 25% of the total grain yields in control plots were from plants in between rows. Increases in grain yield by plants left after thinning would have to exceed 25% of untreated yields to compensate for the removal of terminated plants, which did not occur in this study. Future studies should investigate the space within and between rows by varying the width of the terminated strips and the width of strips left standing for grain production.

Similar to our findings related to termination treatments, no suppression treatment consistently increased grain yields in this study. However, averaged over termination treatments, the mowing suppression treatment reduced grain yields compared to no suppression treatments. This is important in the context of dual-use management since a spring forage harvest can provide valuable feed for livestock on the farm or be sold for an additional revenue source. Puka-Beals et al. [22] showed that a single spring IWG forage harvest could provide up to USD 350 ha$^{-1}$ yr$^{-1}$ in net returns. Hunter et al. [23] found that net return from spring harvested IWG forage varied from USD 188 to 474 ha$^{-1}$ yr$^{-1}$ when harvested prior to stem elongation to ensure grain harvest, similar to this study. Hunter et al. [23] also found that a spring forage harvest increased grain yield in the first production year, but decreased grain yield in years 3 and 4. This aligns with our findings that spring mowing reduced grain yield. Whether or not returns from the spring forage harvest can compensate for reduced returns from a reduction in grain yield depend on the price of Kernza® grain.

The effects of burning were positive for both grain and straw yields. For total grain yields, burning limited the negative effects of the fall herbicide termination treatment (Table 4). Without a fall prescribed burn, a fall herbicide resulted in reduced grain yields compared to other termination treatments (but not the control), but the burning suppression treatment increased grain yields to limit the negative effects of this termination treatment. Burning also increased total straw yields compared the control when averaged over all the termination treatments (Table 3). Prescribed burning is a common management method to promote plant species diversity and productivity in perennial grasslands in the Upper Midwest USA. Burning can increase overall productivity by enhancing nutrient cycling and availability, which could explain the increases in total straw yields observed in this study. Burning has also been found to increase flowering and seed production of some North American grass species. Researchers observed increases in the flowering of wet prairie grass species in response to prescribed burning and speculated that burning mimicked natural patterns of lightning-ignited fires, of which these species have evolved flower responses to [24]. Another hypothesis is that light quality and amount of exposure to plant crowns affects floral induction and seed yield in subsequent years [7]. Researchers found that shading crowns of Kentucky Bluegrass (*Poa pratensis* L.) reduced seed yields in subsequent years [25]. For decades, Kentucky Bluegrass seed producers in North America have burned fields between years of production. Concerns around air quality and public safety have resulted in burning restrictions in some US states [26], thus this management technique may not be available to all Kernza® growers in North America.

The timing of termination treatments appears to have been important for IWG grain yield. Total grain yields varied significantly by the timing of herbicide application in 2019 (Table 3); whereas the fall herbicide treatment resulted in the lowest total grain yield while the spring treatment resulted in the highest total grain yield. It is possible that this difference in herbicide response was because glyphosate is a systemic herbicide that may have been translocated from leaves and stems downward to roots in the fall as many perennial types of grass exhibit prior to winter dormancy [27]. Translocation of herbicide to roots may have resulted in greater levels of plant mortality, whereas in spring, phloem transport of carbohydrates in perennial grasses moves primarily from roots to shoots, thus limiting damage to plant crowns [27,28]. Therefore, a foliar glyphosate application in the spring may not have been translocated throughout the IWG plants, that is from the between-row space towards the plant crowns within-rows, to the extent that it would be in the fall.

An important concern regarding between-row termination is weed dynamics. Disturbed between-row space can be colonized by weeds, which can compete with crops and reduce yields for near and long-term crop production. Here, we report two important findings regarding weed dynamics. First, weed pressure in the untreated control plots was low to negligible in this study (Table 5). Other researchers have documented relatively high levels of weed abundance in the first one to two years of IWG grain production [29] and that weed pressure decreases with increasing IWG stand age [30]. Here, we show that by year four of production, weeds presented minimal concerns for grain production. We also found that the effects of herbicide and cultivation termination treatments varied in terms of weed biomass (Table 5). Plots that received any herbicide application, but particularly when applied in the fall, had significantly more weed pressure compared to cultivated treatments in 2019. This result was surprising in that we expected the cultivation treatments to stimulate BW weed abundance as the added soil disturbance would increase resource availability and conditions for weed seed germination. Weed abundance in-creased in both WR and BR areas in herbicide treated plots. We suspect that lateral movement and uptake of herbicides by WR IWG plants may have reduced productivity and limited their ability to compete with weeds. This hypothesis would be supported if there was evidence of reduced WR straw biomass in the herbicide treatments, and although the means were lower than some treatments, straw yields were not statistically different across treatments (Table 3).

We hypothesized that BR termination via cultivation would help stimulate WR IWG productivity by reducing competition for below-ground resources [31,32], but this was only supported with marginal significance in 2018 when fall cultivation resulted in greater straw yield that year. Root biomass can also be used to infer differences in belowground resources and competition based on optimal partitioning theory. We observed a decrease in WR root biomass in spring cultivated plots compared to other cultivation and control plots in 2018. Optimal partitioning theory would suggest that if terminating plants between rows would limit competition for belowground resources, WR root biomass would decrease. The fact that this was not observed consistently across cultivation treatments or years highlights other factors at play. By 2019, BR root biomass differed among treatments in that the most intensive cultivation treatment (spring + fall) had the lowest root biomass. This is expected as cultivation is effective at stimulating decomposition of root tissues in the soil. Research is needed to determine if BR cultivation leads to significant carbon emissions in a perennial grain cropping system.

## 5. Conclusions

Herbicide and cultivation effectively terminated volunteer IWG plants that had colonized the space between planted IWG rows, but this did not consistently improve plant productivity of IWG within planted undisturbed rows. Herbicide applied in the spring was among the highest yielding treatments for grain and straw, but when applied in the fall, the herbicide treatment caused significant yield reductions and annual weed growth. A spring herbicide termination treatment followed by a fall burning suppression treatment resulted in the greatest grain yields compared to most treatment combinations but not the control, thus this grain yield persistence approach warrants further testing. Spring mowing decreased total grain yields by 42%, but the spring forage that was produced during the mowing has the potential to generate an economic return equal to or greater than grain losses resulting from grain yield reductions. Along with additional research to test agronomic methods that prevent yield decline, studies are needed to determine the genetic control of yield longevity and the potential to select for this important trait in plant breeding programs.

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