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Effects of Nitrogen Fertilizer Rate, Timing, and Herbicide Use on Industrial Hemp (Cannabis Sativa)

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EFFECTS OF NITROGEN FERTILIZER RATE, TIMING, AND HERBICIDE USE ON INDUSTRIAL HEMP (*Cannabis sativa*)

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I dedicate this thesis to my parents, Ron and DeDe Anderson. Without all the love and support throughout my life I would not be where I am today if it wasn't for you two.

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EFFECTS OF NITROGEN FERTILIZER RATE, TIMING, AND HERBICIDE USE ON INDUSTRIAL HEMP (*Cannabis sativa*)

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Industrial hemp (*Cannabis sativa*) is an old crop being reintroduced into certain states; thus, very little information is known about growing the crop domestically. Two field experiments were established in Bowling Green, KY to evaluate various nitrogen fertilizer rates and timing applications to examine the effects on growth and yield of industrial hemp (*Cannabis sativa*). Each experiment was sprayed with 1.12 a.i. ha^{-1} pendimethalin on half of each plot as a pre – emergent herbicide. Hemp was planted at a rate of 43 kg/ ha into a conventionally tilled silt loam soil. Nitrogen rates studied were a control, 79, 157, and 236 kg N/ ha in the first experiment. In the second experiment, 157 kg N/ ha was applied at three timings along with a control. The three timings were: at establishment, side-dressed, and a split application of 78 kg at establishment and 78 kg side dressed. Plots were 6 m x 4.5 m and replicated 4 times for each experiment.

Data collected included stand counts 34 days after planting (DAP), plant heights at 34 and 55 DAP, and stand counts, plant heights, fresh biomass, dried biomass, seed yield, and fiber yield at harvest (89 DAP) for both experiments. Increased fertilizer rates increased fresh and dried biomass which correlated with increased fiber yields. Different fertilizer timings had no effect on seed, biomass, or fiber yields. Herbicide had an early effect on stand counts and plant height in both experiments (<20% reduction) but hemp overcame these effects and yields were not negatively impacted. Results indicated that higher nitrogen rates increase yields while nitrogen timing has no effect. Pendimethalin

utilized as a pre – emergent herbicide could be used with minimal industrial hemp injury if the crop becomes federally legal.

INTRODUCTION AND LITERATURE REVIEW

I. Industrial Hemp Classification and Description

Industrial Hemp, *Cannabis sativa* L. (Cannabaceae), is a versatile crop and shares its botanical name with marijuana. Industrial hemp must have a delta-9 tetrahydrocannabinol (THC) concentration ≤ 0.3 percent on a dry weight basis (KYAGR, 2017) by law. Industrial hemp also differs from marijuana by having higher levels of CBD (cannabidiol) compared to THC. THC possesses psychoactive properties and is the only known psychoactive cannabinoid produced by *Cannabis sativa* (Hinterhuer, 2015). Hemp is an annual wind- pollinated plant and most cultivars are dioecious and indeterminate. In North America, hemp production is concentrated in Canada. Currently, most growers plant in the spring from mid-April (van der Werf et al., 1996) to mid-May (Cosentino et al., 2012). Planting outside this window can limit growth and yields, due to inadequate amounts of sun light or short day lengths (van der Werf et al., 1996). Most hemp cultivars are short-day plants. Earlier planting dates will boost vegetative growth. Four to five weeks after emergence the plants will have an accelerated vegetative growth period lasting five to six weeks; this allows plants to utilize longer photoperiods of the summer, doing this limits early flowering and maximizes stem growth (Bocsa and Karus, 1998). The stem is comprised of two components; the inner hurd and the outer bark (bast fiber) and can range from 2.5 – 5 cm in diameter. Plant population influences stem diameter and plant height (fiber length). Hemp leaves have six or more serrated lobes arranged in a palmate fashion. Seeding rate for seed varieties (ex. Delores, Joey, and Canda) is approximately 30 kg ha^{-1} and higher for fiber varieties (ex. Fedora 19, Felina 34, Futura 77) but not exceeding 80 kg ha^{-1} (Hall et al., 2014). A suitable soil for hemp growth should have a pH of 6.0 - 7.5 (Amaducci et al., 2014) with a soil texture of sandy loam for best growth. Clay loam could also be suitable, but heavy clays and sandy soils are not recommended (Li, 1982). Hemp root can reach depths of 2 - 2.5 meters in light well drained soils, and secondary root branches may grow 60 - 80 cm (Bocsa and Karus, 1998). Hemp cultivars reach a variety of different heights. Seed varieties first need a clean soil bed with little weed pressure. These varieties grow to 1-2 m and are typically planted on 15 – 18 cm row spacing (Purdue, 2017). The use of a burn down herbicide prior to planting is the only method of herbicide use that is approved (Alberta Agriculture and Forestry, 2015); otherwise tillage is utilized to control weeds prior to seeding. During the growing season, mechanical weed management is the only form of control. Techniques for harvesting seed include using a combine with a soybean head to extract the seed from the plants or manually stripping the plants of the flower material, this is very time consuming and reduces seed cleanliness. Fiber varieties range 2-4 m tall (Purdue, 2017). A burn down herbicide needs to be applied to give the crop time to grow with little interference from weeds. Tillage practices are also used for prevention of weeds. Fiber varieties are routinely planted at a recommended rate of 67 kg/ha (Alberta Agriculture and Forestry, 2015) which allows for dense populations resulting in slim, taller plants. Techniques for harvesting fiber include using a disk bar mower, metal weed eater blades, or silage cutter. After being cut the hemp is allowed to dry and ret. Retting is a process that allows for bacteria to breakdown the fibers that attach the bast fiber (outer bark) to the inner hurd. Two methods of retting are field retting and winter retting. Field retting is the process of allowing sun and moisture to fuel fungi to separate the fiber from the hurd. Winter retting is a process that keeps hemp in the field all winter and during spring a roller is used to break the stems, which are later baled (Alberta Agriculture and Forestry, 2015).

Afterward, fiber is taken to a processing plant where a decorticator separates the fiber from the hurd. Varieties grown for CBD are grown differently than for seed or fiber production. CBD production can utilize a burndown herbicide and tillage prior to field planting. However, CBD varieties are often started in green houses from propagated female clones, since CBD is concentrated in female flowers. Three to four weeks after establishment, they are transplanted. CBD plants have spacing's of approximately 1 m (can vary with cultivars) and follow a tobacco production scheme. Plants will reach a height intermediate between seed and fiber varieties, with more robust branching. Weeds are managed between rows by mowing, weed eating, or hoeing. Plants are cut by hand or using a metal disk blade attachment for a weed eater and impaled onto sticks and hung in barns to dry. After a few weeks of drying, the plants are transported to have flower materials stripped and processed for CBD concentrates. Hemp is considered a niche crop grown in small quantities around the world, in Europe only 15,000 ha are grown (Carus et al., 2013) and Canada grows 44,000 ha (Alberta Agriculture and Forestry, 2015). Hemp has not received the agronomic and breeding efforts that other more intensively grown crops have had over the past few decades, which has limited progress in developing new cultivars, machinery, and processing procedures (Campiglia et al., 2017).

II. History

Hemp originated in Central Asia over 8,500 years ago (Schultes and Hofmann, 1980). Hemp has been grown for fiber use for a few thousand years, but more recently, has been grown for seed, fiber, and CBD. Hemp is thought to be one of the oldest cultivated plant species, archeological evidences indicates its use during the Neolithic period (10,200 BC – 2000 BC) (Li, 1974) and hemp was among the first plants to be cultivated by humans (Swenson, 2017). Hemp fiber is valuable because it is a versatile textile whose products range from ropes and paper, to sails on ships. Hemp was an ideal fiber for these vessels because it was remarkably strong and able to withstand harmful weather. Hemp grown for fiber was introduced to western Asia and Egypt, and subsequently to Europe between 1000 and 2000 BCE (Small and Marcus, 2002). Hemp in colonial America was a common crop and had a large economic impact that helped shape the United States into what it is today. President Washington practiced hemp farming and at one point considered hemp to be a more lucrative cash crop than tobacco. He initially wanted to grow hemp to determine if it could be a viable crop in America. "Washington's diaries and farm reports indicate that hemp was cultivated at all of his 5 farms" (MountVernon.org, 2017). Most of the hemp President Washington grew was for textiles such as rope, thread, canvas, and fishing nets. "There were 11 state-sponsored fleets during the American Revolution, as well as the Continental Navy, and every single ship needed ropes and sails. A single vessel in the Virginia Navy called the "Brigantine Liberty," for example, required more than two miles of cordage." (MountVernon.org, 2017). In July 1798, the *USS Constitution* made its maiden voyage, and required more than 2000 tons of fiber, 55 tons of which went into making just the lines and rigging (Will, 2004). After the Civil War, Kentucky was the leader in hemp production (Will, 2004). Kentucky accounted for about 50% of national industrial hemp production during the 1800s (Hopkins, 1951). In the census of 1900, Kentucky produced 5800 hectares yielding 4700 metric tons (Moore, 1905). During World War II farmers across Kentucky were summoned to grow hemp for textiles. Farmers were sent to the University of Kentucky for lessons on growing hemp and after a few weeks of learning returned back to their farms with seed. During this time, hemp was grown for ropes and other textiles for the war effort.

III. Legalities

Industrial Hemp was a widely grown crop for many years, by many states, and for several products. There have been many laws in the United States over the last 80 years that have limited the hemp industry. The first law was the Marihuana Tax Act of 1937 (Kaiser and Cassady, 2014), which did not criminalize hemp production or use but instead, every farmer, processor, and importer was taxed and had to be registered by the federal government. Anyone noncompliant with this law could face 5 years in prison and a $2000 fine (U.S. Legal, 2017). It wasn't until the Controlled Substance Act of 1970 that industrial hemp and marijuana became illegal to grow, process, or distribute. This law classified any plant that contained trace amounts of THC to be classified as a Schedule I Narcotic. All *C. sativa*, no matter the intent of use, is governed by this act and heavily regulated. The Drug Enforcement Administration states that "Schedule I drugs, substances, or chemicals are defined as drugs with no currently accepted medical use and a high potential for abuse. Some examples of Schedule I drugs are: heroin, lysergic acid diethylamide (LSD), marijuana (cannabis), 3, 4-methylenedioxymethamphetamine (ecstasy), methaqualone, and peyote" (U.S.DEA, 2017). Many years passed before a spark of interest returned for hemp production. Kentucky passed Senate Bill 50 in 2013 that exempted hemp from the Controlled Substance Act of 1970 (Kaiser and Cassady, 2014). The U.S. also passed The Farm Bill 2014 that gave the states individual authority to grow hemp under regulations from the federal government and to have pilot programs to monitor the production and assist with research. In 2014 Kentucky began with 19 farmers, 7 universities, growing 12 hectares and as of 2017 there were 250 farmers, 6 universities, and 5,200 hectares granted through the pilot program (KDA, 2017). In July

2017 Rep. James Comer and many other authors, introduced Bill H.R.3530 – Industrial Hemp Farming Act of 2017 to the U.S. House of Representatives (Comer, 2017). This bill excludes hemp from being classified as marijuana and would make it legal to grow for industrial purposes (NORML, 2017). This would change the Controlled Substance Act and only classify marijuana as a Schedule 1 drug. Passage of this bill would likely increase interest in hemp as well as hemp market potential.

IV. Usages

"Hemp, the new billion-dollar crop, could produce more than 25,000 products" was stated in Popular Mechanics Magazine in 1938. Industrial Hemp is grown for three primary commodities: seed, fiber, and CBD. Seed varieties are grown differently from the other styles of hemp. Hemp for seed is usually spaced farther apart than for fiber, this allows for more branching and production of seeds. Seeds from hemp produce a wide range of products. Hemp seed can be used in or on a wide range of foods such as salads, cereals, or even yogurts. Hemp seed may be crushed into oils for lotions, cosmetic products, massage, or cooking oils. The seed can be ground into a powder which is high in protein (36%) (Osburn, 1992). Hemp seeds also contain all amino acids including the 8 essential amino acids the human body cannot produce. The oil in the seed accounts for 35% of the weight in each seed and hemp seed contains 25% linolenic acid (Osburn, 1992). Fiber varieties of hemp are usually planted at high populations. Fiber crops need fewer branches or buds to encourage greater stem growth. Hemp fibers are produced for composite materials, rope, paper, construction materials, automotive components, carpeting and clothing (Kaiser and Cassady, 2014). Hemp fibers also contain antibacterial properties (Khan et al., 2014). Hemp plants also have an inner hurd that when separated by retting can be used for animal bedding. CBD (cannabidiol) varieties produce oils for creams, pharmaceuticals, waxes, and edible foods. Most forms come in oil and are administered orally. CBD has been growing in popularity because of its non-habit forming properties compared to pharmaceutical products. CBD differs from THC in marijuana because it does not confer any psychoactive properties. People utilize CBD to help with seizures, epilepsy, and other neurological disorders (Devinsky et al., 2014). CBD oils are also used as an anti-inflammatory to reduce pain.

V. Nutrient Management

Seventeen essential nutrients are needed for plants to grow and complete their life cycle. Little agronomic research on industrial hemp has been accomplished compared to crops such as corn, wheat or soybeans. Sound agronomic recommendations are needed for growers to maximize growth of the plant for seed, fiber, and CBD. With the growing interest in hemp around the world, agronomic practices are still scarce (Aubin et al., 2015). Nitrogen plays an important role in most soil fertility management plans; it is usually applied in higher concentrations compared to other nutrients. Nitrogen is a component of chlorophyll, amino acids and proteins and influences cell division and plant growth and will promote rapid vegetative growth. Plants with low levels of nitrogen will show deficiencies that include stunted growth along with leaf chlorosis (Tucker, 1999). Nitrogen can be found in either organic or inorganic forms in the soil. Organic forms of nitrogen are found when plant and animal material decomposes and releases proteins and amino acids back into the soil as part of the nitrogen cycle. When organic materials are broken down by bacteria they form inorganic nitrogen as ammonia, in a process called ammonification (Murphy, 2007). Inorganic nitrogen includes nitrates (NO_3^-), nitrite, (NO_2^-), ammonium (NH_4^+), and dinitrogen gas (N_2). European research had indicated hemp cultivars did not need as much nitrogen compared to wheat (Ranalli, 1999). Research in Canada demonstrated that hemp responded to nitrogen up to 120 kg N/ ha when soils were deficient, by increasing seed and biomass yield (Vera et al., 2009). Research in Saskatchewan showed a positive response to fertilizers, with nitrogen rates up to 150 kg N/ ha being optimal (Vera et al., 2009). In Quebec, Canada, additions up to 200 kg N/ ha showed a positive response with no plateau observed (Aubin et al., 2015).

Optimal nitrogen rates for industrial hemp vary according to usage and cultivar (Vera et al., 2009). Nitrogen has been seen to affect stem size but not have as much effect on inflorescence or seed production in areas near the Mediterranean (Campiglia et al., 2017). Fields that are not deficient in phosphorus or potassium may not respond to the addition of these (Aubin et al., 2015). Phosphorus applications impacted plant heights during certain growing seasons, but had no effect on seed and biomass yields between trials (Vera et al., 2004). There is very little research on potassium in industrial hemp. Some evidence has shown K to increase plant height under certain conditions, but seed yield and biomass were not increased (Vera et al., 2009).

VI. Weed Control

Mechanical weed control, which includes tillage and mowing, is one of the more common forms of weed management used worldwide. Not all weeds will be controlled by mechanical methods so hands-on methods such as pulling weeds or using a garden hoe may need to be implemented; however, these methods are very time consuming and labor intensive. Chemical control is most often utilized by farmers in the majority of crops grown and is often the most economical and convenient method. Canada states that no in-crop herbicide is labeled for the use on industrial hemp; however, Manitoba can use a burn-off herbicide such as glyphosate (Manitoba Agriculture, 2018). Manitoba Extension also suggests a tillage practice before seeding to help with weed management which allows time for the hemp to develop size and density before weeds can emerge.

In the United States, industrial hemp is still classified federally as a Schedule I Narcotic thus no pesticides have been labeled for use. Therefore, it is illegal to use herbicides on hemp unless it is for research and will be destroyed instead of processed for sale. Researchers are investigating different herbicides in the future that could potentially be utilized and labeled by companies for weed control in industrial hemp. Cultural control is another method that can be very beneficial for farmers. Cultural methods include crop rotation and maintaining good soil fertility. Most crop rotations in Kentucky include a grass such as winter wheat, followed by a double crop of soybeans, and followed the next year by corn. In Midwestern areas the rotation might be one year of corn, followed by soybeans. This allows for fields to continue to be maintained weed free by planting different crops so different modes of actions can be used in a management plan. Alberta

Agriculture and Forestry (2015) suggests hemp follow cereals because cereals are more easily managed for broadleaf weeds.

MATERIALS AND METHODS

Experiment 1

Field plots were established at the Agriculture Research Education Complex in Bowling Green, KY in 2017 to determine the optimal rate of nitrogen fertilizer application and the effects of herbicide application on crop growth and yield. Soil was conventionally tilled with a roto-tiller prior to planting into a Crider silt loam (Typic Paleudalf). On June 2, 2017, cv. Helena was seeded with a Flex II Seeder (Truax Company, Minneapolis, Minnesota, USA) at a rate of 43 kg/ ha. Herbicide and nitrogen were applied on June 3, 2017. Herbicide used was pendimethalin (group 3) at 1.12 kg a.i. ha $^{-1}$ which was surface applied to half of each 6 m x 4.5 m plot. Nitrogen as urea (46-0-0) was applied at 0, 79, 157, and 236 kg ha^{-1} at planting (Table 1) and each treatment was replicated four times. Stand counts were collected at 34 days after planting (DAP) by selecting random m^2 areas in each plot and counting the total population of plants. Plant heights were collected at 34 and 55 DAP by measuring 5 random plants. At 89 DAP stand counts, plant heights, and fresh biomass was taken. Biomass was obtained from a m^2 area within each plot. At 96 DAP seed yield was collected from a m^2 area where material (flowers and buds) were stripped, bagged, and dried in a forage dryer at 68° C for 48 hours prior to removal of plant material from seed via a 5-mesh soil screen. Dry biomass was measured 117 DAP by drying fresh biomass material in the field and subsequently weighing it. Fiber yield was recorded 117 DAP by macerating dry biomass bundles to separate bast fiber from inner hurd. The macerator was set at a 0.818mm gap with 40 PSI pressure on the steel roller and 60 PSI on the rubber roller. After collecting each sample, fiber was dried at 68° C in a forage dryer for at least 48 hours, before weighing. Data collected was analyzed using SAS 9.4 software (SAS/STAT, 2013). Normality was analyzed using Sapirio – Wilks test by PROC UNIVARIATE. Homogeneity of variances was analyzed using Brown – Forsythe test by PROC GLM. Data was analyzed in a multi-way ANOVA using PROC GLIMMIX. Least Square Means was separated using a PDIFF option with a Tukey adjustment. Significance was determined at $\alpha = 0.05$.

Table 1. Plot Diagram for Nitrogen Rate Experiment

104-P	104		204-P	204		304-P	304		404-P	404
103-P	103		203-P	203		303-P	303		403-P	403
102-P	102		202-P	202		302-P	302		402-P	402
101-P	101		201-P	201		301-P	301		401-P	401

Nitrogen Rate Treatments	Plots
Control	102,203,303,404
79 kg N/ ha	101,202,304,401
157 kg N/ ha	104,204,301,403
235 kg N/ ha	103,201,302,402

All plot numbers marked with -P were sprayed with pendimethalin

Experiment 2

Field plots were established at the Agriculture Research Education Complex in Bowling Green, KY in 2017 to determine the optimal rate of nitrogen fertilizer application and the effects of herbicide application on crop growth and yield. Soil was conventionally tilled with a roto-tiller prior to planting into a Crider silt loam (Typic Paleudalf). On June 2, 2017, cv. Helena was seeded with a Flex II Seeder (Truax Company, Minneapolis, Minnesota, USA) at a rate of 43 kilograms of seed a hectare. Herbicide and nitrogen were applied on June 3, 2017. Herbicide used was pendimethalin (group 3) at 1.12 kg a.i. ha^{-1} which was surface applied to half of each 6 m x 4.5 m plot. Nitrogen as urea (46-0-0) was applied at establishment (E), side dressed (SD) 3 weeks after establishment, half at establishment and half side dressed (S) and a control with each treatment replicated four times (Table 2). Stand counts were collected at 34 days after planting (DAP) by selecting random m^2 areas in each plot and counting the total population of plants. Plant heights were collected at 34 and 55 DAP by selecting 5 random plants. At 89 DAP stand counts, plant heights, and fresh biomass was taken. Biomass was obtained from a m^2 area within each plot. At 96 DAP seed yield was collected from a m^2 area where material (flowers and buds) were stripped, bagged, and dried in a forage dryer at 68° C for 48 hours prior to removal of plant material from seed via a 5-mesh soil screen. Dry biomass was measured 117 DAP by drying fresh biomass material in the field and subsequently weighing. Fiber yield was recorded 117 DAP by macerating dry biomass bundles to separate bast fiber from inner hurd. The macerator was set at a 0.818 mm gap with 40 PSI pressure on the steel roller and 60 PSI on the rubber roller. After collecting each sample, fiber was dried at 68° C in a forage dryer for at least 48 hours, before weighing. Data collected was analyzed using SAS 9.4 software (SAS/STAT, 2013). Normality was analyzed using Sapirio – Wilks test by PROC UNIVARIATE. Homogeneity of variances was analyzed using Brown – Forsythe test by PROC GLM. Data was analyzed in a multi-way ANOVA using PROC GLIMMIX. Least Square Means was separated using a PDIFF option with a Tukey adjustment. Significance was determined at α= 0.05.

Table 2. Plot Diagram for Nitrogen Timing Experiment

104-P	104		204-P	204		304-P	304		404-P	404
103-P	103		203-P	203		303-P	303		403-P	403
102-P	102		202-P	202		302-P	302		402-P	402
101-P	101		201-P	201		301-P	301		401-P	401

Treatments	Plots
Control	102,203,303,404
157 kg N/ ha @ establishment	101,202,304,401
157 kg N/ ha side dressed	104,204,301,403
79 kg N/ ha @establishment + 79 kg N/ha side dressed	103,201,302,402

All plot numbers marked with -P were sprayed with pendimethalin

Results

Experiment One

Stand Counts

Nitrogen rate did not affect stand count ($p \geq 0.21$) at 34 DAP or at harvest (Table 3). Pre-emergent herbicide reduced stand counts by 27% ($p < 0.01$) at 34 DAP as compared with the control (Table 4). At harvest, pre-emergent herbicide reduced stand counts by 42% ($p < 0.01$) as compared with the control (Table 4).

Table 3. Effect of Nitrogen Fertilizer Rate on Industrial Hemp Stand Count

Variables	Nitrogen rate (kg/ ha)				SEM[1]	*P* - value	
	0	79	157	236		N*H	N
	plants/ ha x 1000						
6 July 2017	1518.7	1469.2	1397.6	1549.9	211.4	0.93	0.82
Harvest	728.6	665.3	553.6	655.3	90.9	0.88	0.21

Means within a row with different superscripts differ (P < 0.05).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Table 4. Effect of Herbicide on Industrial Hemp Stand Count

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	plants/ ha x 1000				
6 July 2017	1709.2[a]	1258.5[b]	178.8	0.93	<0.01
Harvest	823.9[a]	477.6[b]	71.6	0.88	<0.01

Means within a row with different superscripts differ (P < 0.05).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Plant Height

At all three evaluation dates, nitrogen rate had no effect ($p > 0.12$) on plant height (Table 5). Plant heights were reduced 24% ($p < 0.01$) at 34 DAP from pre-emergent herbicide being applied (Table 6). As the growing season progressed, plants ($p \geq 0.29$) overcame the stunting (Table 6).

Table 5. Effect of Nitrogen Fertilizer Rate on Industrial Hemp Plant Height

Variables	Nitrogen rate (kg/ ha)				SEM[1]	*P* - value	
	0	79	157	236		N*H	N
	meters						
6 July 2017	0.402	0.410	0.380	0.403	0.028	0.45	0.71
27 July 2017	2.164	2.238	2.315	2.327	0.096	0.30	0.16
Harvest	2.373	2.468	2.500	2.599	0.125	0.67	0.12

Means within a row with different superscripts differ (P < 0.05).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Table 6. Effect of Herbicide on Industrial Hemp Plant Height

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	meters				
6 July 2017	0.456[a]	0.345[b]	0.023	0.45	<0.01
27 July 2017	2.240	2.820	0.076	0.30	0.45
Harvest	2.453	2.518	0.106	0.67	0.29

Means within a row with different superscripts differ (P < 0.05).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Biomass

Nitrogen rates impacted ($p < 0.01$) fresh and dried biomass yields (Tables 7, 9). Biomass weights for 236 kg N/ ha or 157 kg N/ ha did not differ nor did 157 kg N/ ha or 79 kg N/ ha. Nitrogen applied at 236 kg/ ha produced greater bio0mass than the control plots. Pre-emergent herbicide did not impact ($p = 0.27$) fresh biomass or dried biomass (Tables 8, 10).

Table 7. Effect of Nitrogen Fertilizer Rate on Industrial Hemp Fresh Biomass

Variables	Nitrogen rate (kg/ ha)				SEM[1]	*P* - value	
	0	79	157	236		N*H	N
	kg/ ha						
Harvest[2]	22360[c]	27740[bc]	30140[ab]	36560[a]	3050	0.29	<0.01

Means within a row with different superscripts differ ($P < 0.05$).

[1] Standard Error of the Mean

[2]Data were analyzed using a square root transformation due to failure of normality.

N*H = interaction between nitrogen and herbicide

Table 8. Effect of Herbicide on Industrial Hemp Fresh Biomass

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	kg/ ha				
Harvest[2]	30020	27950	2490	0.29	0.27

Means within a row with different superscripts differ ($P < 0.05$).

[1] Standard Error of the Mean

[2]Data analyzed using square root transformation due to failure of normality.

N*H = interaction between nitrogen and herbicide

Table 9. Effect of Nitrogen Fertilizer Rate on Industrial Hemp Dried Biomass

Variables	Nitrogen rate (kg/ ha)				SEM[1]	*P* - value	
	0	79	157	236		N*H	N
	kg/ ha						
Harvest	6840[c]	8410[bc]	8590[ab]	10130[a]	770	0.21	<0.01

Means within a row with different superscripts differ ($P < 0.05$).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Table 10. Effect of Herbicide on Industrial Hemp Dried Biomass

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	kg/ ha				
Harvest	8740	8250	620	0.21	0.27

Means within a row with different superscripts differ ($P < 0.05$).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Seed Yield

Nitrogen rates impacted ($p < 0.01$) seed yields (Table 11). Seed yield for 236 kg N/ ha, 157 kg N/ ha, and the control did not differ. Seed yields for 236 kg N/ ha and 79 kg N/ ha did statistically ($p < 0.01$) differ (Table 11). Pre-emergent herbicide did not impact (p = 0.06) seed yield (Table 12).

Table 11. Effect of Nitrogen Fertilizer Rate on Industrial Hemp Seed Yield

Variables	Nitrogen rate (kg/ ha)				SEM[1]	*P* - value	
	0	79	157	236		N*H	N
	kg/ ha						
Harvest	870^{ab}	748^{b}	1090^{a}	1180^{a}	130	0.26	<.01

Means within a row with different superscripts differ (P < 0.05).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Table 12. Effect of Herbicide on Industrial Hemp Seed Yield

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	kg/ ha				
Harvest	890	1050	100	0.26	0.06

Means within a row with different superscripts differ (P < 0.05).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Fiber Yield

Nitrogen rates impacted (*p* = 0.04) fiber yields (Table 13). Fiber yields did not differ when 236 kg N/ ha, 157 kg N/ ha or 79 kg N/ ha were applied; however fiber yields were improved when 236 kg/ ha was applied (Table 13). Pre-emergent herbicide did not impact (*p* = 0.13) fiber yield (Table 14).

Table 13. Effect of Nitrogen Fertilizer Rate on Industrial Hemp Fiber Yield

Variables	Nitrogen rate (kg/ ha)				SEM[1]	*P* - value	
	0	79	157	236		N*H	N
	kg/ ha						
Harvest	2170^{b}	2960^{ab}	2540^{ab}	3612^{a}	384	0.65	0.04

Means within a row with different superscripts differ (P < 0.05).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Table 14. Effect of Herbicide on Industrial Hemp Fiber Yield

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	kg/ ha				
Harvest	3090	2550	290	0.65	0.13

Means within a row with different superscripts differ (P < 0.05).
[1] Standard Error of the Mean
N*H = interaction between nitrogen and herbicide

Experiment Two

Stand count

Nitrogen timing applications did not impact stand counts (*p* = 0.12) at 34 DAP (Table 15). At harvest, only 157 kg/ ha nitrogen (p < 0.01) at establishment differed statistically from the control (Table 15). Pre-emergent herbicide reduced stand counts by 21% (*p* < 0.01) at 34 DAP as compared with the control (Table 16). At harvest, pre-emergent herbicide reduced stand counts by 26% (*p* < 0.01) as compared with the control (Table 16).

Table 15. Effect of Nitrogen Fertilizer Timing on Industrial Hemp Stand Count

Variables	Nitrogen Application[1]				SEM[2]	*P* - value	
	C	E	SD	S		N*H	N
	plants/ ha x 1000						
6 July 2017	1621.73	1589.30	1467.00	1707.70	256.1	0.69	0.12
Harvest	1055.7[a]	717.41[b]	873.187[ab]	863.66[ab]	95.6	0.49	<0.01

[1]C = Control, E = 157 kg/ ha at establishment, SD = 157 kg/ ha at side dress, and S = 79 kg/ ha at establishment and 79 kg/ ha at side dress.

[2] Standard Error of the Mean
Means within a row with different superscripts differ (P < 0.05).
N*H = interaction between nitrogen and herbicide

Table 16. Effect of Herbicide on Industrial Hemp Stand Count

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	plants/ ha x 1000				
6 July 2017	1787.83[a]	1405.26[b]	203.1	0.69	<0.01
Harvest	1006.5[a]	748.42[b]	77.4	0.49	<0.01

[1] Standard Error of the Mean
Means within a row with different superscripts differ ($P < 0.05$).
N*H = interaction between nitrogen and herbicide

Plant Height

Nitrogen timing application did not have an effect on plant height, except at the 27 July evaluation date. Split application of 79 kg/ ha at establishment and 79 kg/ ha side dressed, 157 kg/ ha at establishment, and control did not differ; however split application of 79 kg/ha at establishment and 79 kg/ ha side dressed (*p* = 0.048) resulted in greater plant height than when side dressed (Table 17). Pre-emergent herbicide reduced plant height (p < 0.01) by 21% at 34 DAP as compared to control (Table 18). Plant height did not differ at later evaluation dates.

Table 17. Effect of Nitrogen Fertilizer Timing on Industrial Hemp Plant Height

Variables	Nitrogen Application[1]				SEM[2]	*P* - value	
	C	E	SD	S		N*H	N
	meters						
6 July 2017	0.383	0.375	0.347	0.403	0.027	0.67	0.11
27 July 2017	2.091[ab]	2.142[ab]	2.059[b]	2.252[a]	0.089	0.46	0.048
Harvest	2.101	2.247	2.186	2.234	0.138	0.94	0.63

[1]C = Control, E = 157 kg/ ha at establishment, SD = 157 kg/ ha at side dress, and S = 79 kg/ ha at establishment and 79 kg/ ha at side dress.

[2] Standard Error of the Mean
Means within a row with different superscripts differ (P < 0.05).
N*H = interaction between nitrogen and herbicide

Table 18. Effect of Herbicide on Industrial Hemp Plant Height

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	meters				
6 July 2017	0.422[a]	0.332[b]	0.85	0.67	<0.01
27 July 2017	2.100	2.171	0.08	0.46	0.13
Harvest	2.227	2.156	0.11	0.94	0.39

[1] Standard Error of the Mean
Means within a row with different superscripts differ ($P < 0.05$).
N*H = interaction between nitrogen and herbicide

Biomass

Nitrogen timing applications (*p* > 0.29) did not impact fresh or dried biomass (Tables 19, 21). Pre-emergent herbicide did not impact (p > 0.07) fresh or dried biomass weights (Tables 20, 22).

Table 19. Effect of Nitrogen Fertilizer Timing on Industrial Hemp Fresh Biomass

Variables	Nitrogen Application[1]				SEM[2]	*P* - value	
	C	E	SD	S		N*H	N
	kg/ ha						
Harvest	20590	22900	22270	23170	1990	0.54	0.33

[1]C = Control, E = 157 kg/ ha at establishment, SD = 157 kg/ ha at side dress, and S = 79 kg/ ha at establishment and 79 kg/ ha at side dress.

[2] Standard Error of the Mean
Means within a row with different superscripts differ (P < 0.05).
N*H = interaction between nitrogen and herbicide

Table 20. Effect of Herbicide on Industrial Hemp Fresh Biomass

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	kg/ ha				
Harvest	22990	21450	1720	0.54	0.13

[1] Standard Error of the Mean
Means within a row with different superscripts differ (P < 0.05).
N*H = interaction between nitrogen and herbicide

Table 21. Effect of Nitrogen Fertilizer Timing on Industrial Hemp Dry Biomass

Variables	Nitrogen Application[1]				SEM[2]	*P* - value	
	C	E	SD	S		N*H	N
	kg/ ha						
Harvest	8810	8750	9253	9344	680	0.38	0.29

[1]C = Control, E = 157 kg/ ha at establishment, SD = 157 kg/ ha at side dress, and S = 79 kg/ ha at establishment and 79 kg/ ha at side dress.

[2] Standard Error of the Mean
Means within a row with different superscripts differ (P < 0.05).
N*H = interaction between nitrogen and herbicide

Table 22. Effect of Herbicide on Industrial Hemp Dried Biomass

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	kg/ ha				
Harvest	9299	8573	544	0.38	0.07

[1] Standard Error of the Mean
Means within a row with different superscripts differ (P < 0.05).
N*H = interaction between nitrogen and herbicide

Seed Yield

Nitrogen timing application (*p* = 0.6) did not impact seed yield (Table 23). Pre-emergent herbicide application increased seed yield (p < 0.01) by 24% (Table 24).

Table 23. Effect of Nitrogen Fertilizer Timing on Industrial Hemp Seed Yield

Variables	Nitrogen Application[1]				SEM[2]	*P* - value	
	C	E	SD	S		N*H	N
	kg/ ha						
Harvest	839.1	953.1	874.0	955.2	141.6	0.4	0.6

[1]C = Control, E = 157 kg/ ha at establishment, SD = 157 kg/ ha at side dress, and S = 79 kg/ ha at establishment and 79 kg/ ha at side dress.
[2] Standard Error of the Mean
Means within a row with different superscripts differ (P < 0.05).
N*H = interaction between nitrogen and herbicide

Table 24. Effect of Herbicide on Industrial Hemp Seed Yield

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	kg/ ha				
Harvest	826[b]	1023[a]	117.2	0.4	<0.01

[1] Standard Error of the Mean
Means within a row with different superscripts differ (*P* < 0.05).
N*H = interaction between nitrogen and herbicide

Fiber Yield

Neither nitrogen timing applications (p = 0.36) nor pre-emergent herbicide (p = 0.09) impacted fiber yield (Tables 25, 26).

Table 25. Effect of Nitrogen Fertilizer Timing on Industrial Hemp Fiber Yield

Variables	Nitrogen Application[1]				SEM[2]	*P* - value	
	C	E	SD	S		N*H	N
	kg/ ha						
Harvest	3566	3709	3076	3970	457	0.57	0.36

[1]C = Control, E = 157 kg/ ha at establishment, SD = 157 kg/ ha at side dress, and S = 79 kg/ ha at establishment and 79 kg/ ha at side dress.

[2] Standard Error of the Mean
Means within a row with different superscripts differ (*P* < 0.05).
N*H = interaction between nitrogen and herbicide

Table 26. Effect of Herbicide on Industrial Hemp Fiber Yield

Variables	Herbicide		SEM[1]	*P* - value	
	Control	Applied		N*H	H
	kg/ ha				
Harvest	3886	3275	381	0.57	0.09

[1] Standard Error of the Mean
Means within a row with different superscripts differ (*P* < 0.05).
N*H = interaction between nitrogen and herbicide

Discussion

Experiment 1

Industrial hemp is a crop being reintroduced that has agronomic recommendations in Europe and Canada, but not in the United States. This study was designed to investigate both an optimal rate of nitrogen fertilizer and the effects of herbicide application. During the early growing season, there was little impact from nitrogen being added but by harvest addition of nitrogen impacted fresh and dried biomass, seed, and fiber yield.

Nitrogen rates of 157 kg/ ha and 236 kg/ha resulted in greater fresh and dried biomass, and only the 236 kg ha $^{-1}$ application rate increased fiber yields. Rates of 157 and 236 kg N/ ha had an impact correlated with Canadian research that stated a range of up to 150 kg N/ ha nitrogen in one area was optimal but in another part of Canada 200 kg N/ ha nitrogen was needed for maximum yield (Vera et al., 2009, Aubin et al., 2015).

Herbicide application at planting had negative effects on stand counts at 34 DAP and at harvest. Herbicide application reduced plant height on 34 DAP but had no significance as the growing season continued. Previous research in Bowling Green, KY and Lexington, KY indicated that pendimethalin had no significant injury to the crop. Pendimethalin also had no significant negative impact on biomass or seed yield at either location (Maxwell, 2016). In this study, herbicide had no effect on biomass, fiber, or seed yields which suggests that after the initial injury from the herbicide, hemp recovered and overcame the early phyotoxicity and stand loss.

Experiment 2

This study was designed to determine an ideal timing application of nitrogen fertilizer and the effects herbicide application has on industrial hemp. By harvest, stand counts were reduced by nitrogen applied at establishment compared to control. This finding correlated with research from Manitoba, Canada that nitrogen fertilizers should be added as side dressed, or in between rows (Manitoba, 2018).

Fertilizer timing did not have an effect on plant height 34 DAP but by 55 DAP only side dressed and split applications differed statistically from one another. However, by harvest no significant difference was noted between any of the treatments for plant height, biomass, seed, or fiber yields. Herbicide reduced stand counts by 21% 34 DAP. Although stand counts were reduced early, hemp adjusted with more branching through the growing season and this, along with reduced weed competition, likely increased seed yield. Herbicide had no impact on fiber yield or biomass and positively impacted seed yield compared to control plots which suggests that pendimethalin could be a viable herbicide option should future pesticide registration be possible.

CONCLUSION

These experiments examined nitrogen rates and timings that could potentially be utilized by farmers growing industrial hemp. The experiments also examined the effects of pendimethalin on hemp. Pendimethalin had an early negative effect on hemp stand counts and plant heights in both experiments. However, over time plants were able to overcome the effects of herbicide treatment; therefore, herbicide did not have a lasting effect on fiber, seed, or fresh and dried biomass yields.

Increased nitrogen fertilizer rates increased fresh and dried biomass which correlated with an increase in fiber yields. Different fertilizer timings affected stand counts early in the growing season but by harvest no differences in yield were seen from application timing. In future research, higher nitrogen rates should be applied to determine a maximum rate of fertilizer that could be used on hemp before yield plateaued. For rate and timing applications, research should be repeated to compare data for multiple years.

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