Effect of Nitrogen-Based Biosolid Application on Tree Growth, Water Quality, and Soil Phosphorus in Hybrid Poplar Plantations

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ABSTRACT: A three-year study applied four N-based biosolid treatments to hybrid poplar trees over a two year period with spring and fall applications. All biosolid treatments had a faster rate of diameter and biomass growth (but not height) than the control, but the High and Medium application rates had the fastest rate of growth, and were not significantly different. There was no difference in foliar nitrogen and phosphorus levels between any of the treatments. Water quality data from suction lysimeters generated nitrate values below 20 ppm for the low treatment, which is similar to levels found in a cornfield. Four wells surrounding the site reported nitrate levels within the range of values found associated with other eastern shore cropland. P levels in soil samples escalated immediately after broadcast application to levels above the threshold in the state nutrient management regulation. The use of nitrogen based biosolid application in Maryland or other states with similar phosphorus-based nutrient management regulations is not feasible on the soil types tested.

KEYWORDS: hybrid poplar, nitrogen, phosphorous, forest system, biosolids, water quality, soil

INTRODUCTION

Agricultural land to apply biosolids has become increasing difficult to find for many reasons, leading to the need for more beneficial techniques to utilize the nutrient value of biosolids. Landfilling of nutrient rich material and trucking long distances with the associated environmental impacts are not desirable options.

The use of short rotational woody crops (SRWC) such as hybrid poplar show great promise due to their ability to take up large amounts of nutrients and produce significant increases in growth and biomass that can be used for pulp, bioenergy, co-fired generation, compost and other purposes (Van Ham 2000). The use of biosolids in hybrid poplar forest systems is included as part of the bioenergy policy in the final report of the Governor's Commission on Protecting the Chesapeake Bay through Sustainable Forestry (Sutherland et al. 2006).

The development of deep row application using hybrid poplar trees is an operational technique being done successfully on abandoned gravel spoils in Southern Maryland (Felton, Kays, et al. 2005; Kays, Felton, & Flamino 2008; Buswell, Felton, Kays & Flamino 2006), however, it is problematic in areas with high water tables such as the Eastern Shore. Surface application of biosolids to hybrid poplar plantations that matches the trees nitrogen uptake capabilities has been a mainstay in the Pacific Northwest (PNW) since the early 1990's and provides much of the raw material for the pulp industry (NBMA 2004). However, there is no phosphorous-based nutrient management regulation in the PNW. Therefore, it is critical to determine the effect of different agronomic rates of nitrogen on the P-index. The technique has the potential for widespread application on Maryland's Eastern Shore where utilization of biosolids on grain crops is problematic due to public concerns. The Lower Eastern Shore Tri-County Council has expressed interest in the concept of hybrid poplar tree farms to utilize the over 70,000 wet tons of biosolids generated each year from Worchester, Somerset and Wicomico counties that is presently landfilled (Kays, Felton, & Flamino, 2006). What is lacking is an on-ground demonstration/research study to educate local decision makers about the technique.

The standard uptake capability of hybrid poplar trees is 392 kg/N/ha/yr [350 lbs/N/ac/yr] (Plant Available Nitrogen or PAN) when the trees fully occupy the site (National Agroforestry Center 2000). However, research at Cary Island in British Columbia has found significant growth response of hybrid poplar trees using a one-time application rate of up to 3,989 kg/N/ha/yr [3,562 lbs/N/ac/yr] with minimal impacts on long term water quality (Van Ham 2006). The impact of biosolid applications on the phosphorous index may limit nitrogen application; however, more research is needed to develop regulatory guidelines for silvicultural applications using hybrid poplar.

This research will help provide a better understanding of how SRWC plantations in combination with surface application of biosolids, can provide another beneficial reuse option for biosolids on Maryland's eastern shore, and yield biomass for energy production or other uses.

METHODOLOGY

Site Location & Description

The site is located at the University of Maryland Wye Research and Education Center which is approximately 6 miles south of Queenstown (Figure 1). The site is in the coastal plain physiographic regional province. A 0.61 ha [1.5 ac] research plantation was established by planting steckings of OP367 hybrid poplar clones in spring 2006 (Figure 2). The trees were planted on 3.7 m X 3.7m [12ft X 12ft] spacing and divided into 12 plots with 20 trees per plot (4 rows X 5 rows). There is one row of trees between each internal plot to provide a buffer and one buffer row on the outside of the plot. Vegetation management was used to eliminate or reduce vegetation growth in the tree rows. Goal herbicide is sprayed each winter to create a six-foot dirt strip around the trees. The between-tree-alleys are colonized with fescue grass and is mowed about 3 times per year.

Figure 1.University of Maryland Wye Research & Education Center marked by red star

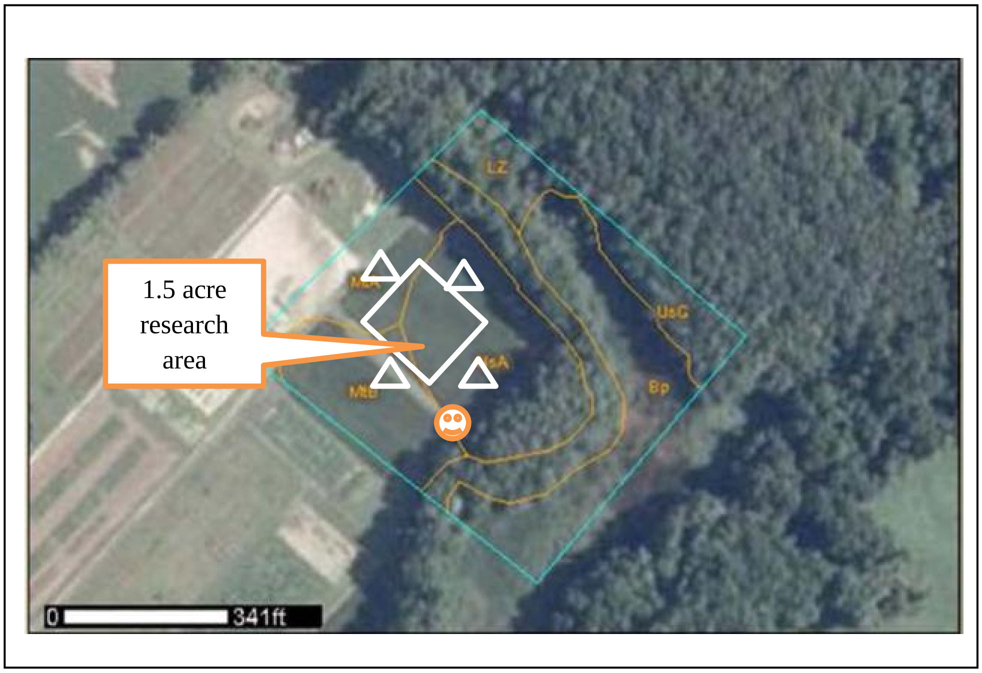

Figure 2. Aerial view of research site. Four well locations marked by triangles and flume marked by smiley face.

The site consists of a mildly sloping field about 3.0 – 7.6 m [10-25 ft] above the water table. The field was in corn and soybeans prior to tree planting. The research area is approximately 18.3-22.9 m [60-75 ft] from mature woodland that buffers a marsh (Figure 2). The study area consists of two soils types. The most abundant soil is a Nassawango silt loam, 0-2% slope (NsA) with very low available water content and is well drained. This soil comprises approximately 80% of the study area. The second soil is the Mattapex silt loam, again 0-2% slope (MtB), with very low available water content and is well- drained. This soil comprises the remaining 20% of the study area.

Research Design &Treatments

The research was set up using a complete randomized block design with three blocks, each including four biosolid application treatments that were based on the nitrogen-based prescribed rate of 392 kg/N/yr/ha [350 lbs. N/ac/yr] (Figure 3). They are: Treatment 1 – control – no biosolid application; Treatment 2 – Low rate, half of the nitrogen based prescribed rate; Treatment 3 – Medium rate, the full nitrogen based treatment; and Treatment 4 – High rate, the twice the nitrogen-based prescribed rate. Actual application rates are expressed as kg/N/ha/yr [lbs/N/ac/yr]. The four treatments were randomly assigned within each of the three blocks.

Applications were applied for two growing seasons (2009 – 4th growing season and 2010 – 5th growing season). The total site area is approximately 0.61 ha (1.5 ac), but application rates (Table 1) are provided on a per acre basis. The total biosolid application for any year were split equally into two separate applications (50% in spring and 50% in the fall) - usually March and October, respectively.

Statistical Analysis Systems (SAS) was used to analyze the data.

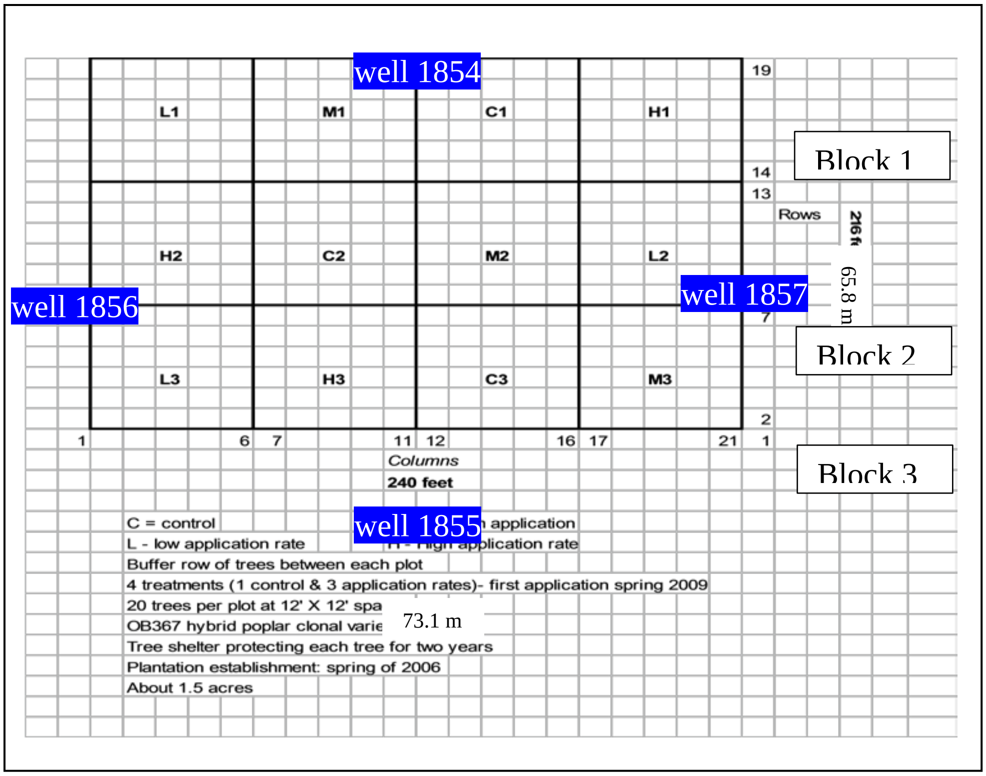

Figure 3. Plot layout. The intersection of each line marks a tree location. The black lines are the buffer rows between treatments. The complete randomized block design has three blocks with four treatments per block. The four well locations and number are marked with a blue box.

Table 1. Target nitrogen application rates on an annual basis. The annual rate is split equally between the spring and fall applications as noted.

	Treatments in kg/N/ha [lbs/N/ac]			
	Control	Low	Medium	High
Plant Available N applied kg/N/ha [lbs/N/ac]	0	196 [175] 99.6 spring - 99 fall	392 [350] 196 spring - 196 fall	588 [525] 296 spring– 296 fall
Total N applied kg/N/ha [lbs/N/ac]	0	560 [500] 280spring- 280 fall	1120 [1000] 560 spring- 560 fall	1680 [1500] 840 spring- 840 fall

Tree Measurements

After each growing season diameter and height measures were taken to assess growth, survival, and biomass production of the trees. Foliar nutrient levels were taken in mid-August of each year according to accepted protocols to determine the fertility status of the trees (nitrogen, phosphorous, potassium, and other nutrients N, P, K, etc) and how it relates to growth measures.

Water Quality Sampling

Lysimeters

Suction lysimeters were installed about 61 cm [24 in] deep in the center of each of the 12 plots on March 12, 2009 to allow monitoring of water quality. Due to damage to the equipment by mowing the first sample was not taken until June, 2009, after the first biosolid application in March. The lysimeters will collect water moving through the soil profile and it will be sampled about once a month and analyzed for pH, chloride, sulfate, NH4, NO3, and Total N.

Wells

Four monitoring wells were installed by a licensed well drilling company to an approximate depth of 7.6 meters [25 ft] just outside the perimeter of each side of the research plot as indicated on plot layout (Figure 1). The wells were screened from 1.5-7.6 m [5 to 25 ft]. Well samples are taken twice a year at six month intervals and analyzed for pH, NH_4, NO_3, and Total N. A sample was taken before the initial application of the biosolids in Year 1 to provide a background level.

Surface Water

Surface water monitoring is achieved by collecting samples from a small flume at the southeast corner of the plot where the topography drains water off the plot area. A berm was constructed down the southwest border of the research area to exclude any surface off from the adjacent field and upslope location and route runoff from the study area to the flume. Grab samples were taken once a year for three years when water is actually moving offsite during a rain event. Samples were analyzed for NH4, NO3, total N, orthophosphate, and Total phosphorous.

Biosolids Sampling

Prior to each application, a sample of biosolids was taken from the storage area at the Kent Island Waste Water Treatment Plant (WWTP) and analyzed for moisture content, pH, total solids, TKN, ammonia, nitrate, total P, plant available P, and K. Sample collection was completed about one week prior to the biosolid delivery to allow the analysis to be used to calibrate the application equipment.

Foliar Leaf Collection and Analysis

Foliar leaf samples were taken to determine the nutritional status of the trees. A rubber-tired articulating boom lift was used to travel up the tree rows and take samples from the terminal leaders. About 12 trees in each of the 12 plots were sampled to form a composite sample, providing 12 data points, one for each plot, or three samples per treatment. Each composite sample was put in a paper bag, air-dried, and sent to a reputable commercial laboratory for analysis. The sampling protocol was as follows (VanHam, 2006): 1) sample time during the peak of the growing season – early to mid-August; 2) sample the first fully expanded leaf, which is usually 5-7 leaves down from the terminal leader of the central vertical branch. Sampling of leaves that are still actively expanding means the leaf will be pulling nutrients from the tree and give unrealistic value. Conversely, if the leaf is past the expanding stage, it will be in a process of decay and a will be net exporter of nutrients. Therefore, the resulting values may be low; 3) do not sample leaves on leaders with deer browsing or extensive insect damage; 4) sample near mid-day when actively growing; and 5) sample 7-10 trees in each plot if possible and make a composite sample.

The protocol had to be altered in some cases due to the high incidence of deer browsing and lack of samples. Rather than a regular sampling scheme across each plot, it was necessary to take leaves from available trees without deer browsing, which resulted in about 6-7 leaf samples from each plot that were combined into a composite sample.
Each application rate – tree density combination is replicated three times. Therefore, there were only three foliar measurements on which to base the statistical tests for any treatment.

Soil Sampling:
In order to assess changes in available soil P (Mehlick 3 technique) and other nutrients with increasing application rate of biosolids, soil samples were taken in the upper 15.2 cm. [6 in] of the soil profile in winters prior to application (2007 & 2008). More intensive soil sampling was completed in September 2009, September 2010, December 2010, August 2011, December 2011, and August 2012, to determine the direction and cumulative effects of applications. The following composite soil samples were taken in each treatment replication in September 2009: 1) 0-15.2 cm [0-6 in] in the tree row; 2) 15.2-30.5 cm [6-12 in] in the tree row; 3) 0-15.2 cm [0-6 in] in the between-tree-alley (grass strip); and 4) 15.2-30.5 cm [6-12 in] in the between-tree-alley (grass strip). Samples in August 2010 and following were restricted to the depth of 0-15.2 cm. [0-6 in] in the tree row and between-tree-alley.

Application Methods and Challenges

Spring 2009 Application – March 26, 2009
The first application in spring 2009 was completed using a conventional manure spreader that only dropped the biosolids behind the spreader on the between-tree-alley (grass strips) and not in the tree row (herbicide strips) where the tree roots could more effectively access the nutrients (Figure 4). Several calibrations were completed using tarps and measuring the application rate at different speeds.

Figure 4. Overview of research area after first application in spring 2009. High application treatment in foreground. Manure spreader deposited biosolids only in grass strip.

Deer rubbing of the tree trunks was significant problem during the winter of 2008-2009 (Figure X) and caused serious damage to the woody trunks. However, by August 2009 (Figure 7) almost all the trees had recovered and mortality was minimal. It is more difficult to determine the impact the damage had on tree growth but the abundant rainfall during the 2009 growing season likely minimized significant mortality or growth impacts. Hybrid poplar is a prolific grower and the wounds were overgrown and growth appeared vigorous.

Every effort was made to minimize buck rubbing during the winter of 2009 and 2010. An electrically charged two-wire polywire fence was installed in November 2009 and baited to attract and shock the deer, thereby deterring them from the area (Kays 2003). The original tree shelters were reinstalled loosely around each of the trees to minimize damage from deer that may comprise the fence.

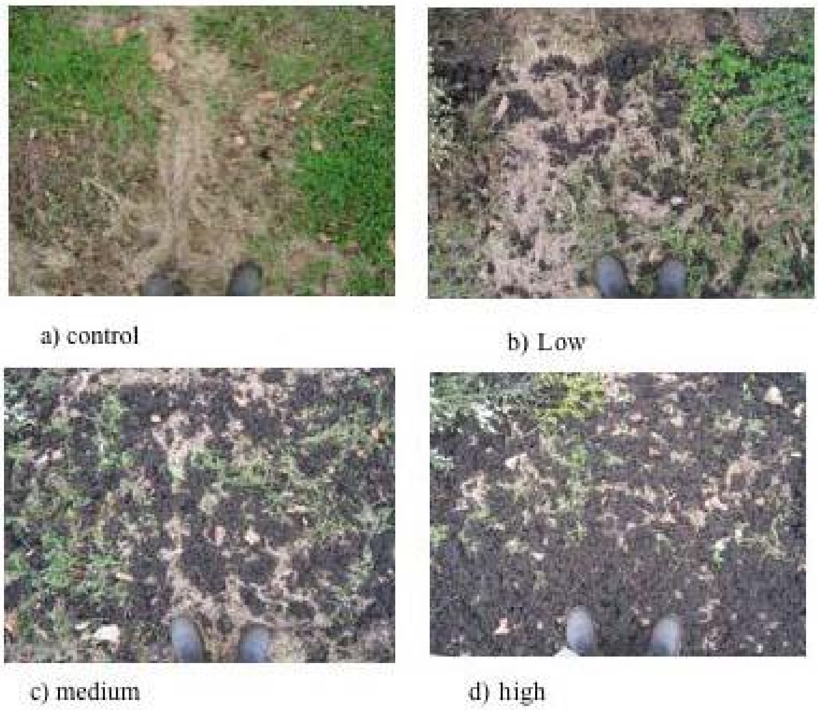

Figure 5. Qualitative view of four biosolid application rates looking straight down from eye level

The following photographs were taken in each of the biosolid treatments from a height of 1.5 meters [5 ft] looking straight down to give a visual impression of ground coverage in each treatment application. Note photographer's boots for perspective.

A used Knight 8114TR Side Slinger Manure Spreader was purchased in August 2009 to allow for more even distribution of biosolids during application. The manure spreader was used for the last three applications of the study: Fall 2009; Spring 2010 and Fall 2010.

Figure 6. Knight Sideslinger manure spreader provided broadcast coverage of site.

Application Data

A sample of the biosolids to be applied was analyzed prior to field calibration and application (Table 2). The spreader was calibrated based on the lbs of biosolids needed to reach the target lbs/N on a 3.0X6.1 m [10ft X 20ft] tarp (Table 3). Three tarps were laid out in a test area so that each run

Table 2. Nutrient content of biosolids as determined from WWTP samples.

Biosolids Analysis for All Application				
Contents	Spring 2009	Fall 2009	Spring 2010	Fall 2010
% Solids	18.92	20.9	17.47	18.59
Dry Tons				
% TKN	6.04	4.11	6.24	5.65
% NH4	2.05	0.43	1.24	1.78
% P	2.89	3.69	2.59	3.27
% K	0.49	0.36	0.53	0.32
Wet Tons				
% TKN	1.14	0.86	1.09	1.05
% NH4	0.39	0.09	0.22	0.33
% P	0.55	0.77	0.45	0.61
% K	0.09	0.08	0.09	0.06

Table 3. Actual biosolid application rates and errors.

Biosolid Application Rate of N					
	Nutrient	Treatment	Wet Mg/ha [Wet tons/ac]	Kg/N/ha [lbs/N/ac]	Target Application Rate kg/N/ha (% over/under applied)
Spring 2009	Nitrogen	Low	221 [9.9]	272 [243]	280 (-3%)
		Medium	43.9 [19.7]	454 [486]	560 (-3%)
		High	66.0 [29.6]	816 [729]	840 (-3%)
Fall 2009	Nitrogen	Low	34.4 [15.4]	387 [346]	280 (38%)
		Medium	68.7 [30.8]	775 [692]	560 (38%)
		High	103 [46.2]	1163 [1038]	840 (38%)
Spring 2010	Nitrogen	Low	25.4 [11.4]	222 [198]	280 (-21%)
		Medium	50.8 [22.8]	444 [396]	560 (-21%)
		High	76.3 [34.2]	665 [594]	840 (-21%)
Fall 2010	Nitrogen	Low	26.5 [11.9]	255 [228]	280 (-9%)
		Medium	53.1 [23.8]	510 [456]	560 (-9%)
		High	79.6 [35.7]	745 [666]	840 (-9%)

provided three replications. The biosolids were collected from the tarp and weighed. During each of the trials, settings such as ground speed, PTO speed, spreader gate opening was adjusted to reach the target rate. During application to the research plots, the tarps used during the calibration were installed to collect actual application rate data and confirm that the settings were appropriate.

Over-application in Fall 2009 occurred due to the difficulty of applying biosolids on small 18.3m X 21.9m [60ftx72ft] plots with machinery (Knight Side Slinger) developed for application over large acreages. The Knight Side Slinger did have advantage of a more even distribution across the tree rows and between-tree-alleys compared to the conventional manure spreader.

The spreader was calibrated to provide the low treatment rate 280 kg/N/ha [250 lbs/N/ac] with one pass. To obtain medium and high rates on a plot, the spreader was stopped and the tractor backed up, followed by either one or two additional forward passes to get the desired application rate. Initially, the technique attempted made multiple applications from opposing sides of the tree row but this quickly led to the overapplication due to confusion on how rates were calculated on an area basis. After careful analysis, it was determined that applications must be completed with the tractor applying biosolids only going in one direction in the tree rows and backing up and going forward again to make multiple applications to a plot.

Table 4. Variation In Application Of Biosolids - Fall, 2010

Target Rate is 28.1 kg [62 lbs] of biosolids on the tarp					
Calibration Prior To Application					
PTO speed	Km/hr [mph]	Gate Height Cm [in]	# of Runs	Average kg [Lbs] on tarp	Range kg [lbs]
800	2.4 [1.5]	15.2 [6]	2	22 [49]	12-36 [27-80]
800	2.4 [1.5]	30.5 [12]	2	36 [79]	32-38 [71-85]
800	3.1 [1.9]	30.5 [12]	2	23 [51]	23-36 [27-80]
800	2.7 [1.7]	30.5 [12]	1	26 [57]	25-30 [42-68]
Each run consists of three replicates					
In-Field Application					
	MPH	Gate Height	# of Runs	Average Kg [Lbs] on tarp	Range Kg [lbs]
800	2.7 [1.7]	30.5 [12]	4	27 [60]	11-46 [26-102]
Two replicates taken at four locations during application					

One objective of the study was to develop operational procedures for this type of agroforestry technique for engineered hybrid poplar plantations. After dealing with the issue of reaching an acceptable average target level, the other issue of concern was the variability in the application rate across the research area. During each application there was wide variation in application rate in many cases. To quantify the variation, the calibration and output of the spreader for the Fall 2010 application was measured using many replicates and runs. Table 4 provides the highlights of this study. During the calibration trial, adjustments were made in the speed of the PTO, MPH, and gate height to reach the target rate of 28 kg [62 lbs]. The best settings provided an average of 25 kg [57 lbs] on the tarp but the different runs ranged from 19-30 kg [42 to 68 lbs]. Measurements taken during the infield application at this same setting provided an average of 27 kg [60 lbs], with values ranging from 11-46 kg [26-102 lbs].

There are many possible reasons for the wide variability, but this study demonstrates that targeting exact application rates with low variability is not possible with this type of conventional application technology. One factor among many was the low PTO speed used to limit the throw of the biosolids to about 4.6-6.1 m [15-20 ft]. Calibration was performed on level ground. It is likely that running up and down hills caused at least some of the variation.

Precipitation

Monitoring stations near the research site provided precipitation data. The monthly data is provided. Most precipitation during the growing season is thunderstorm type events which may produce runoff if the sandy loam soils can not handle the load.

Precipitation Record at Wye Research & Education Center, Queenstown, MD												
Monthly Rainfall in inches												
YEAR	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC
2009	2.74	0.61	2.38	4.83	4.29	5.94	2.93	5.9	3.74	5.87	4.45	8.15
2010	3.08	4.62	5.54	1.46	2.05	2.81	6.91	3.00	7.42	3.32	2.04	2.43
2011	2.46	2.44	4.65	4.29	2.66	2.40	2.87	15.70	8.85	2.50	3.15	5.48
2012	1.81	2.85	1.16	2.54	1.97	4.03	2.78	11.29	2.60	11.19	0.89	3.99
Average	2.52	2.63	3.43	3.28	2.74	3.80	3.87	8.97	5.65	5.72	2.63	5.01

Figure 5. Precipitation at research site for years of the study.

RESULTS

The research plots were planted in spring, 2006 with an overall survival rate of 96% that ranged from a low of 92% for the low treatment plots to 98% for the medium treatment plots. Some trees did die back after the first and second year and resprouted but were not very competitive with the main canopy. In this analysis all trees living in 2012 were included and those that were dead were eliminated. There was a buffer row of trees between each of the 12 replicate plots and these trees were not included in the analysis.

Tree Growth

Tree height, diameter at breast height (DBH) measurements and calculated biomass were analyzed to test for effects of treatment, year, and the interaction of treatment with year. The MIXED procedure of the SAS System (ver. 9.3. SAS Institute, Cary, NC 27513) was used to fit an analysis of covariance model (Figure 11 & 14). The model included block as a random effect and treatment, year, and the year by treatment interaction as fixed effects, as well as the baseline measurement (measured at the end of the growing season in the year before treatment application) of the response as a covariate. Significant year by treatment interactions were further investigated using pairwise comparisons of the slopes across year for each treatment. Model adequacy was assessed using plots of studentized residuals.

Tree Height Growth

Hybrid poplar clones are genetically programmed to grow at similar rates of height and diameter when they have the same level of fertility, available moisture, and environmental factors. This research is providing different levels of fertility through the treatment application of biosolids, although the nutrients in biosolids take longer to become available compared to inorganic fertilizer. There was no significant difference in the rate of height growth between any of the treatments (Figure 7). Most research of this kind find the lowest height for the control and increasing height with higher rates of fertilization. The lack of any difference in tree height is not well understood and may be related more to nutrient runoff from nearby treated plots, moisture stress during the growing season, physiological factors that promote root growth over shoot growth, or other factors.

Figure 7 provides the predicted values of tree height (HT) using the analysis of covariance model. HT did not increase across years (P=0.9881) and the rate of increase across years was not different among the treatments (P=0.9881). The equations describing the relationship are: Control HT = -16178 + 8.05*year + 0.329*baseline HT, Low treatment HT = -15748 + 7.84*year + 0.329*baseline HT, Medium treatment HT = -16083 + 8.01*year + 0.329*baseline HT, High treatment HT = -16575 + 8.26*year + 0.329*baseline HT.

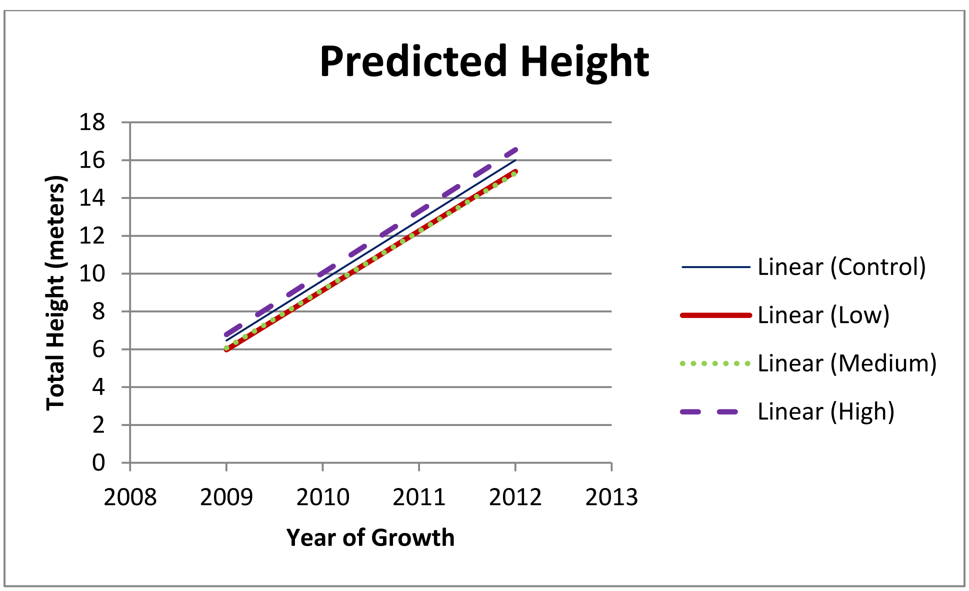

Figure 7. The relationship between total tree height (HT) and year of hybrid poplar trees treated with surface-applied biosolids in 2008 and 2009. Biosolid treatments were control or no biosolids, low rate of applied biosolids, medium rate of applied biosolids, and high rate of applied biosolids.

The least square means and confidence intervals for height for each of the treatments are provided for the 2012, but values for previous years can be read off the graph for the year of interest (Table 6).

Table 6. Least square means and confidence intervals (alpha=0.05) for 2012 tree height based on covariate model.

Treatment	LS Mean Estimate Height (m)	Confidence Intervals	
Control	15.59	13.84	17.35
Low	14.93	13.17	16.69
Medium	16.33	14.57	18.10
High	16.42	14.66	18.18

Tree Diameter

Figure 8 provides the predicted values of DBH using the analysis of covariance model. DBH increased across years (P=0.0001) and the rate of increase across years was different among the treatments (P=0.0345). The equations describing the relationship are: Control DBH = -6263 + 3.12*year + 0.300*baseline DBH, Low treatment DBH = -7009 + 3.49*year + 0.300*baseline DBH, Medium treatment DBH = -7178 + 3.58*year + 0.300*baseline DBH, High treatment DBH = -7851 + 3.91*year + 0.300*baseline DBH.

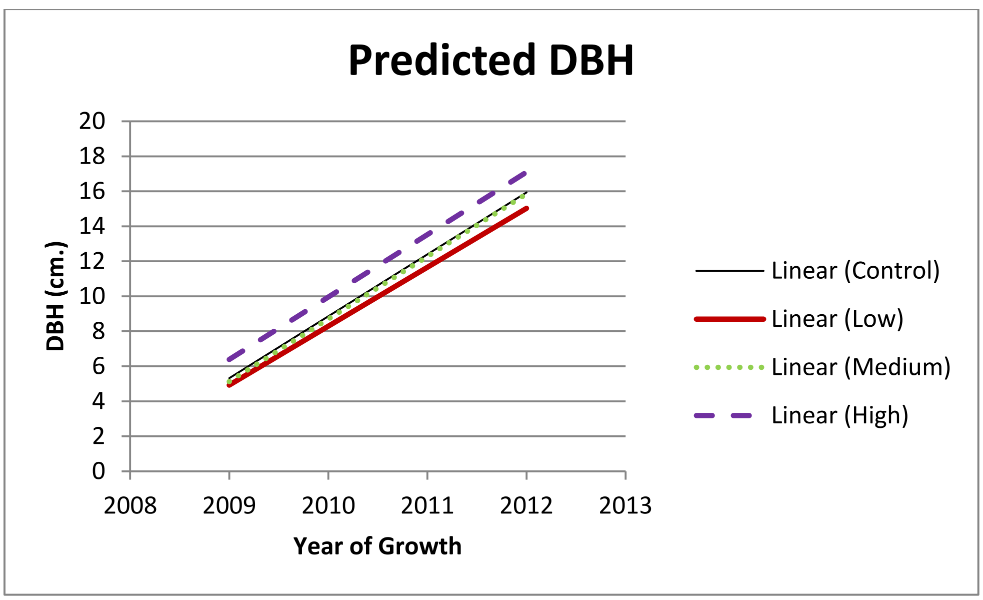

Figure 8. The relationship between diameter at breast height (DBH) and year of hybrid poplar trees treated with surface-applied biosolids in 2008 and 2009. Biosolid treatments were control or no biosolids, low rate of applied biosolids, medium rate of applied biosolids, and high rate of applied biosolids.

All three levels (Low, Medium, and High) of biosolids application resulted in significantly faster increases in diameter biomass than the Control treatment (P=0.0437, P=0.0168, P=0.0003). The

High treatment trees also increased in diameter faster than the Low treatment (P=0.0352) but not faster than the Medium treatment trees (P=0.0729). No difference in increase in diameter across years was detected between the trees in the Low treatment and those in the Medium (P=0.6390) (Figure 8).

The least square means and confidence intervals for diameter for each of the treatments are provided for the 2012, but values for previous years can be read off the graph for the year of interest (Table 7).

Table 7. Least square means and confidence intervals (alpha=0.05) for 2012 DBH based on covariate model.

Treatment	LS Mean Estimate DBH (cm.)	Confidence Intervals	
Control	14.54	13.21	15.87
Low	15.03	13.71	16.37
Medium	16.65	15.3	17.98
High	17.75	16.42	19.08

Biomass Production

Woody biomass production was calculated by using the following equations developed at the ERCO biosolids research site (Felix, et al., 2008).

Calculating Biomass of Hybrid Poplar

When diameter at breast height (DBH) was greater than 4 cm:

Dry woody tree biomass in kg (WB) = 2.6 * DBH (cm) – 9.64

When DBH was less than 4 cm:

Dry woody tree biomass in kg (WB) =0.5 * DBH (cm) – 0.35

Figure 7. Equation used to calculate dry aboveground biomass per tree based in dbh

Diameter at breast height (DBH) is the estimator of tree biomass (Figure 7) so that the significant differences found for tree diameter are directly related to those for tree biomass.

Figure 8 provides the predicted values of biomass using the analysis of covariance model. Biomass increased across years (P=0.0001) and the rate of increase across years was different among the treatments (P=0.0345). The equations describing the relationship are: Control BIOM = -15320 + 7.63*year + 0.062*baseline BIOM, Low treatment BIOM = -17251 + 8.59*year + 0.062*baseline BIOM, Medium treatment BIOM = -18142 + 9.03*year + 0.062*baseline BIOM, High treatment BIOM = -19274 + 9.60*year + 0.062*baseline BIOM.

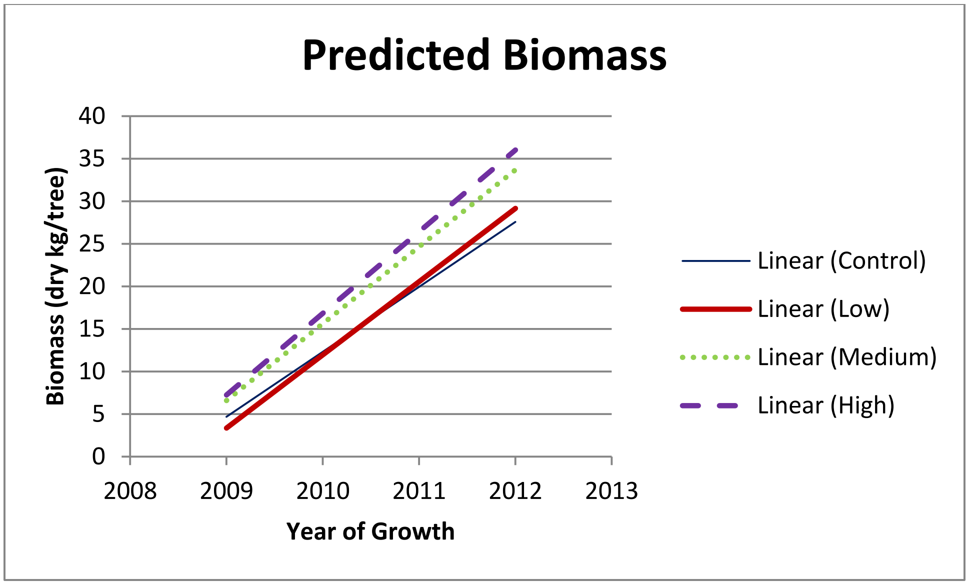

Figure 8. Total tree biomass (BIOM) over time by treatment and year of hybrid poplar trees treated with surface-applied biosolids in 2008 and 2009. Biosolid treatments were control or no biosoids, low rate of applied biosolids, medium rate of applied biosolids, and high rate of applied biosolids.

All three levels (Low, Medium, and High) of biosolids application resulted in significantly faster increases in biomass than the Control treatment (P=0.0493, P=0.0056, P=0.0002). The High treatment trees also increased in biomass faster than the Low treatment (P=0.0438) but not faster than the Medium treatment trees (P=0.2388). No difference in increase in biomass across years was detected between the trees in the Low treatment and those in the Medium (P=3570) (Figure 9).

The least square means and confidence intervals for biomass for each of the treatments are provided for 2012, but values for previous years can be read off the graph for the year of interest (Table 8).

Table 8. Least square means and confidence intervals (alpha=0.05) for 2012 biomass based on covariate model.

Treatment	LS Mean Estimate Biomass (dry kg/tree	Confidence Intervals	
Control	27.57	23.79	32.35
Low	29.17	25.39	32.95
Medium	33.67	29.87	37.47
High	36.03	32.25	39.80

All biosolid applications were effective in increasing the rate of diameter and biomass growth which would be expected due to the additional nutrients available from the biosolids. The High treatment, while increasing faster than the Low rate, was not different from the Medium rate. This indicates there is some type of threshold growth response for the higher application rates. The lack of a difference between the High and Medium treatments indicates that there is no real growth advantage to the higher application but using the medium rate would lower the risk of movement of nutrients from the site and subsequent environmental impacts. The Medium application rate was the prescribed rate of 392 kg/N/ha [350 lbs/N/ac] of plant available nitrogen, which is presented in the literature (National Agroforestry Center, 2000), and this research appears to support that rate.

The nutrients present in biosolids is mostly in organic forms and takes time to mineralize and become available for plant uptake. This study demonstrates that the multiple applications of biosolids to the plots continued to provide available nutrients after the applications ceased, due to breakdown of the biosolids. The rate of increase in diameter and biomass may have been greater if the first application in spring 2009 had been broadcast across the plots, instead of applied only between the tree rows.

Foliar Nutrient Levels

The collection of foliar leaf samples of hybrid poplar trees is an accepted method to assess the uptake of available nutrients by the trees and the impact of various treatments on tree growth (Hansen, E.A 1994; Hansen, E.A. and D.N. Tolsted 1985). Changes in foliar leaf concentrations for N and P have been correlated with changes in growth of hybrid poplar (Zabek 1995; McIntosh 1984). To establish a baseline for the foliar nutrient uptake and enable trends to be identified in the future, foliar nutrient samples were collected during the second growing season (August, 2004) using an accepted protocol (VanHam 2003).

There are no comprehensive studies correlating levels of foliar nutrient concentrations and poplar growth; however, higher foliar nutrient levels are usually correlated with faster growth. In separate studies on hybrid poplar clones, both Hansen (1988) and Heilman and Xie (1993) suggest that maintaining a 3% foliar N concentration was desirable for fast growth. Clonal studies by Zabek (1995) on the eastern side of Vancouver Island found maximum growth occurred at foliar concentrations of 3.6% N and 0.42% P.

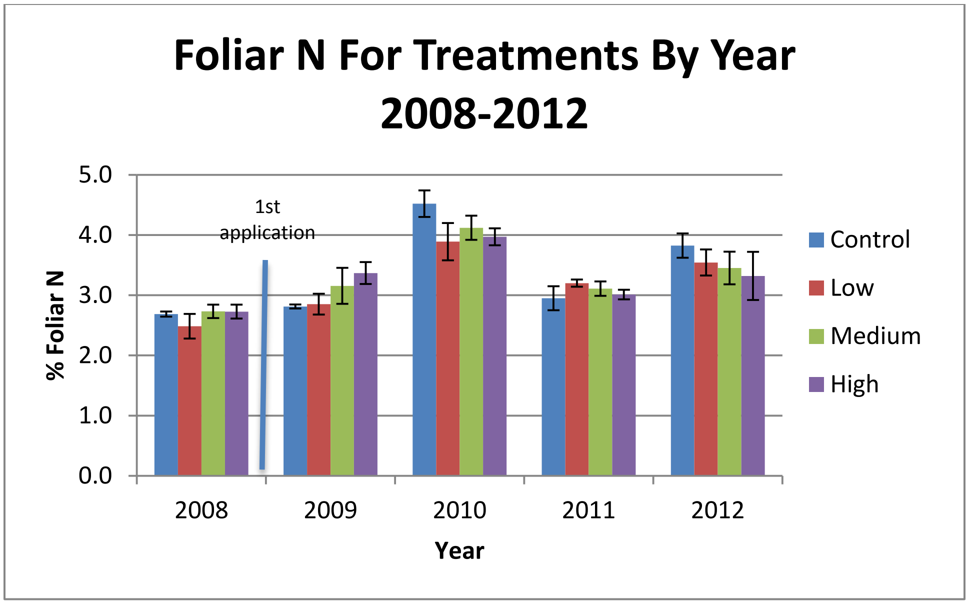

Figure 9. % Foliar N for Each Treatment by year (2008-2012).

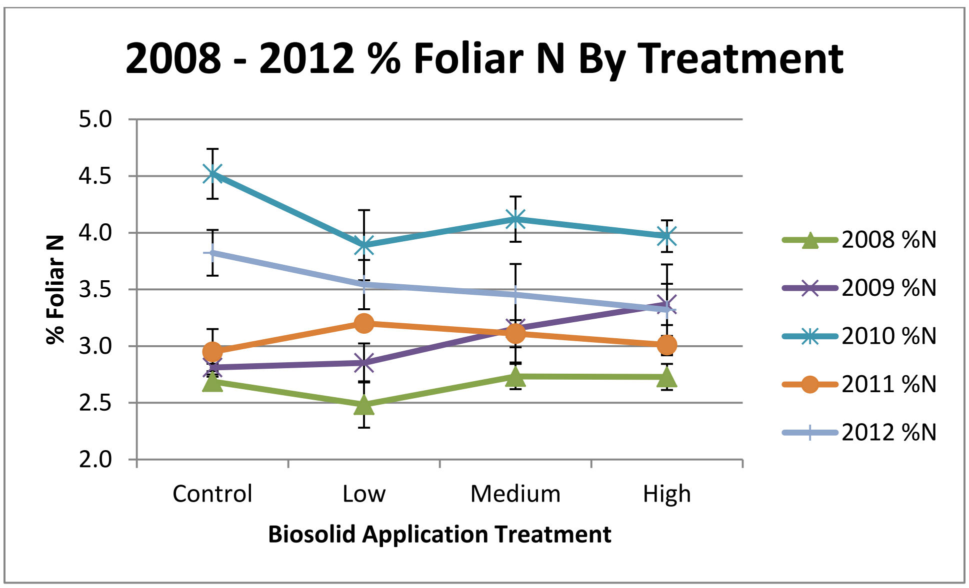

Figure 10. % Foliar N by treatment.

Composite foliar samples were taken in each of the 12 research plots from 2007 to 2010, but only those from the year prior to application (2008) are provided. In 2008, the %N and %P foliar nutrient levels were 2.66%N and 0.25%P, respectively. As expected, a statistical analysis found there were no differences between treatments because no biosolids were applied until 2009. The %N and %P levels were well below the 3% N and 0.35% P level needed for fast growth.

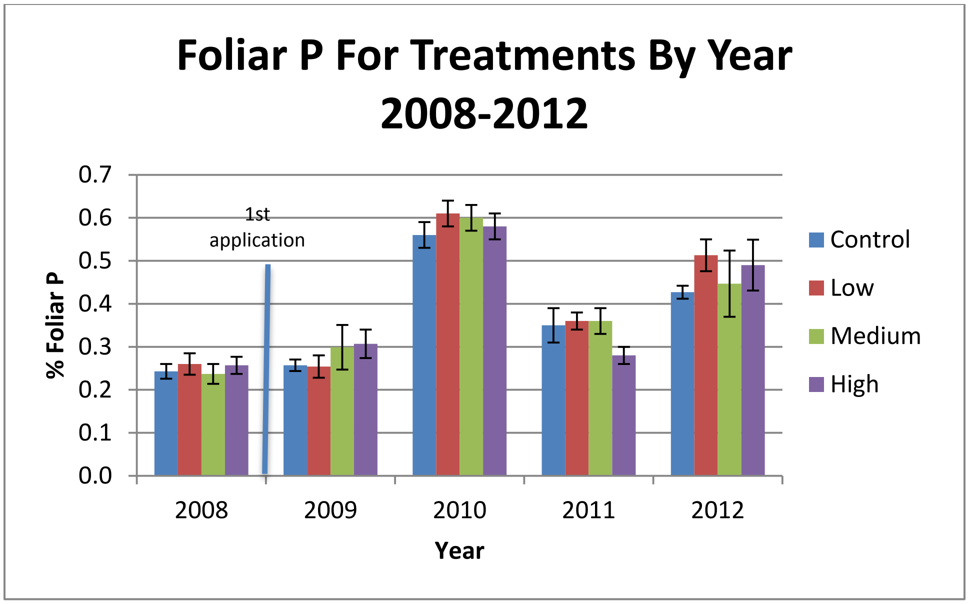

Figure 11. % foliar P for treatments by year.

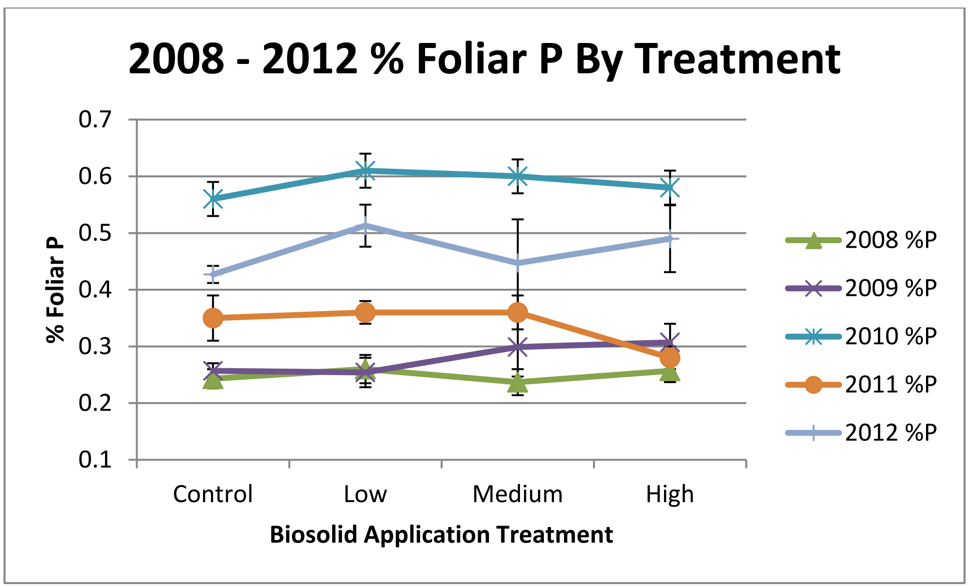

Figure 12. % foliar P by treatment

Figures 10 & 12 present the %N and %P levels for each treatment for 2008 through 2012 using a line graph. The different color line for each year demonstrate the relatively low nutritional status prior to application (2008), the slight increase in 2009, the dramatic increase in 2010, the subsequent fall in 2011, and the increase again in 2012.

Percent N and P increased greatly in 2010 to levels that are known to maximize growth, averaging 4.1% N and 0.59% P for all treatments. In 2010, %N ranged from 3.8-4.5%N for all treatments, levels indicating strong uptake of available nutrients and maximum growth. In the case of %P, values more than doubling from 2009 to 2010, ranging from 0.56 to 6.0. These are foliar nutrient levels not seen widely in the literature.

What was unexpected was the lack of significant differences between any of the treatments for foliar nutrient levels of N and P for the 2009, 2010, 2011, and 2012 growing season. Most research shows that amending soils with biosolids results in increases in growth that is matched by higher foliar nutrient levels of N and P. This study confirms the faster increases in growth in diameter and biomass (but not tree height) for the biosolid treatments compared to the control, without the expected increasing trend in foliar nutrient levels.

The higher rates of diameter and biomass growth for medium and high treatments have the same foliar nutrient status as the control and low application rates, which have lower rates of growth. Apparently, the elevated nutrient status of the two treatments with lower growth, are not being converted into above-ground biomass, while the two treatments with higher growth are responding with more above-ground biomass production.

A spike in foliar nutrient levels in 2010 for the treatments with biosolids applied was expected as nutrients become available from the biosolids. What was unexpected was high foliar level of N and P for the control plots. No biosolids were applied to control plots but they had the highest nutritional status of all treatments in 2010. The high nutrient levels across all treatments compared to the pretreatment levels in 2008 indicates that there was some kind of nutrient transport across the research area. Some possible explanations are provided in the discussion section.
****A possible explanation is that in October 2009 biosolids were overapplied by 38% to the entire research area (Table 3). The precipiThe small size of the plots and the heavy rains that fell after application could have resulted in overland or subsurface movement of nutrients into the control and low treatment plots. However, the nutrients were not effective in providing a growth response in diameter or biomass. Another factor is mid- to late-summer drought on the site that can affect the available water. The plots with medium to high application rates not only have more available nutrients but the biosolids also act as mulch layer of sorts to capture and keep rainfall that is produced from quick summer downpours. The control and low treatment plots lack this ability to retain moisture and the grass strip also utilized a lot of soil moisture. So, while the control and low treatment plots may have a good nutritional uptake, it may have lacked the available moisture to produce biomass.
***
The high nutritional status for N and P reported in 2010 only lasted one year. In 2011, %N and %P fell to levels similar to those found in 2009. The rapid drop in foliar nutrients for all treatments in 2011 suggests that whatever fertility was available from the previous biosolid applications had been utilized or immobilized and growth would be reduced unless additional biosolids were provided. However, the levels of N and P increased again in 2012, which was probably due an increase in the nutrient pool generated by favorable moisture and temperature conditions for biosolid decomposition during the 2012 growing season.

Water Quality

Lysimeters were installed March of 2009. The first samples (prior to any biosolids applications) were collected on March 31, 2009. Because of some modifications that had to be made to the system, no further sampling was possible for approximately 120 days. Lysimeters were then sampled monthly through December. Because of temperatures (water freezes in the tubes) sampling was then suspended until late February, 2010.

In June 2010, three lysimeters ceased yielding water for samples. The next month, July, ten of the twelve lysimeters did not yield water for samples. This is normal for dry soil, where the soil water pressure is lower than the vacuum applied to the lysimeters, so nothing was done until September, when it was clear that the soil water was in ample supply and only three lysimeters yielded samples. This was when it became clear that there was some operational problem with the lysimeters.

In December 2010, the nine lysimeters that were not functioning were field-tested. Four of those lysimeters were repaired in situ. Four more of those lysimeters were brought back to the lab for analysis, repair and/or replacement. This was completed in January 2011. Because of time constraints, one lysimeter was replaced in February, 2011.

During the site visit (December, 2010) a possible cause of failure for a number of lysimeters was the splices. The movement of the tubing hanging in the trees along with the wind likely caused a number of splices to leak. In some cases the splices came apart with minimal pulling on the tubing. Tubes with splices were replaced with continuous tubes. A second problem was the connection between the tube and the lysimeter, which is buried approximately one foot beneath the ground surface. In two cases, it was clear that this connection had failed. We hypothesize that the heavy traffic from the spreader may have caused the soil to shift and damaged the connection. Because we are not anticipating any more biosolids applications, we do not anticipate a repeat of this problem.

Two charts of results are presented below (Figures 13 and 14). The first chart (Figure 13) shows the individual lysimeter values. Both figures are color coded: the control is light blue, the low application rate is dark blue, the medium application rate is green, and the high application rate is magenta.

One lysimeter (#12) in figure13 was considerably higher than the other values. While this may be an artifact of installation or simply an outlier, this lysimeter was scrutinized for the next several months. The values in #12 were consistently high and this point must be considered a real, but suspect data point. All the other nitrate values were similar to each other, independent of biosolids application rate, up to May 2010. Nitrate in the control plots exceeded nitrate levels in the low application rate plots for the period Oct.-Nov. 2009.

After November of 2010, it was clear that the high and medium rates resulted in elevated nitrate levels in the suction lysimeters. Some lysimeters were higher than others, but all the high and medium lysimeters showed nitrate levels above the low and control levels. The purpose of fertilizing is to put nutrients, of which nitrate is one, into the root zone so that the plant can capture and utilize them. The treatments succeeded in this. However, nitrate levels above 60 mg/L suggest that not all nitrogen is being consumed by the plant (trees).

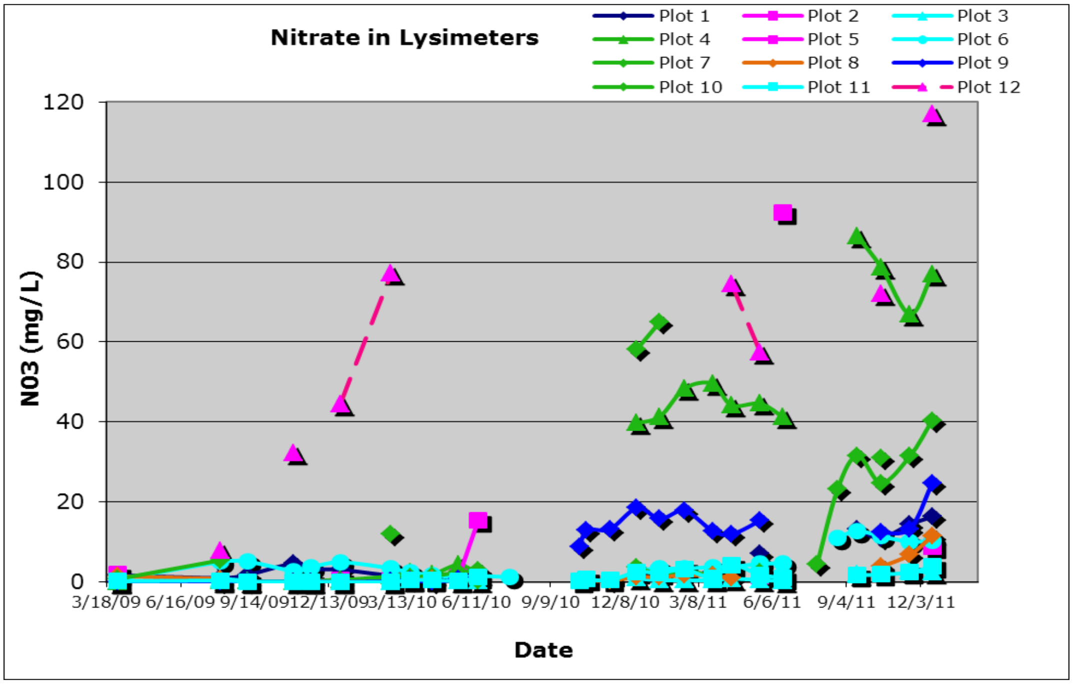

Figure 13. Nitrate in soil lysimeters.

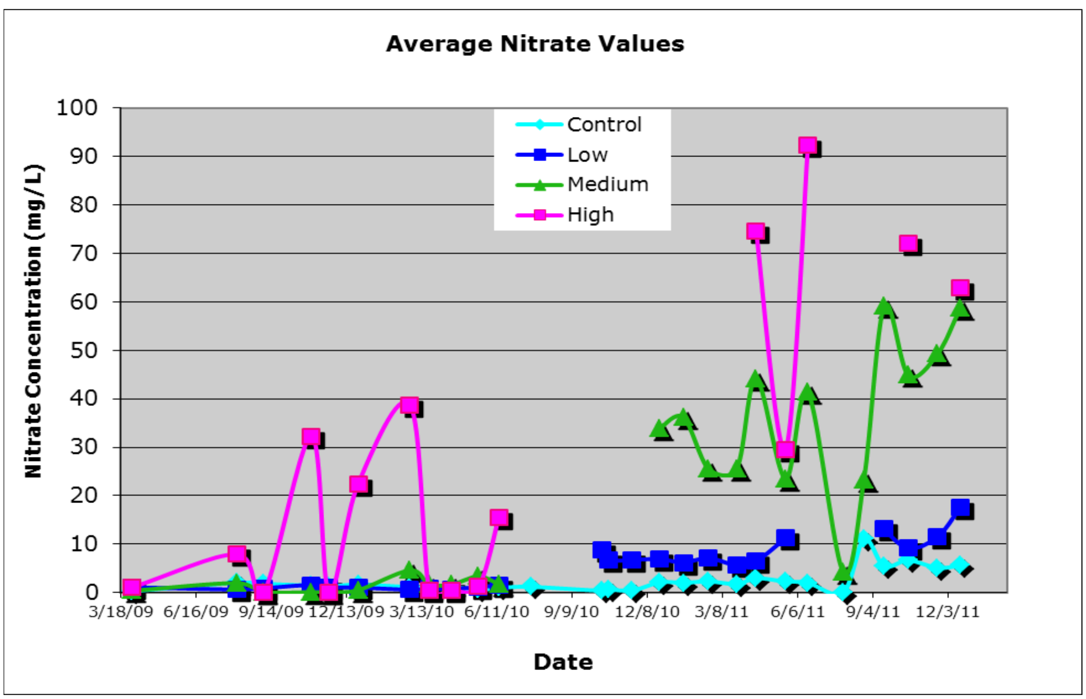

Figure14. Average nitrate values in soil lysimeters.

The elevated nitrate values in lysimeter 12 are the reason for the odd oscillations in figure 14 for the high application rate. The low values on the oscillating curve occurred when lysimeter 12 had no sample. Ignoring the oscillations, it is clear that beginning in about March of 2011, the average nitrate levels from the high and medium levels were getting too high for all the nitrate to be used and some was probably going to leach.

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Wells

The plot layout and well location is schematically represented in Figure 3. For the first well bailing, all wells were bailed to approximately three well bore volumes and then left to recharge for two hours. They refilled very rapidly. At well 1856, the first bailer full was clear, subsequent bailers increased in sediment load. At well 1855 there was sediment in every bailer. Even the ball in the bailer was coated with sediment. When the wells were sampled, Wells 1855 and 1856 had clear samples with almost no sediment. Wells 1854 and 1857 had a small amount of grey color to the water. Sediment in samples can alter nutrient contents, especially phosphorus. Similar procedures were performed at each well sampling event. During most samplings, fine roots were found in all four wells. It is assumed that the trees had sent roots down 10 feet or deeper to access the water table during the summer.

The wells were sampled twice a year, once in October and once in March, corresponding roughly to the low and high water table stages expected, based on the hydrologic year (hydrologic year begins October 1).

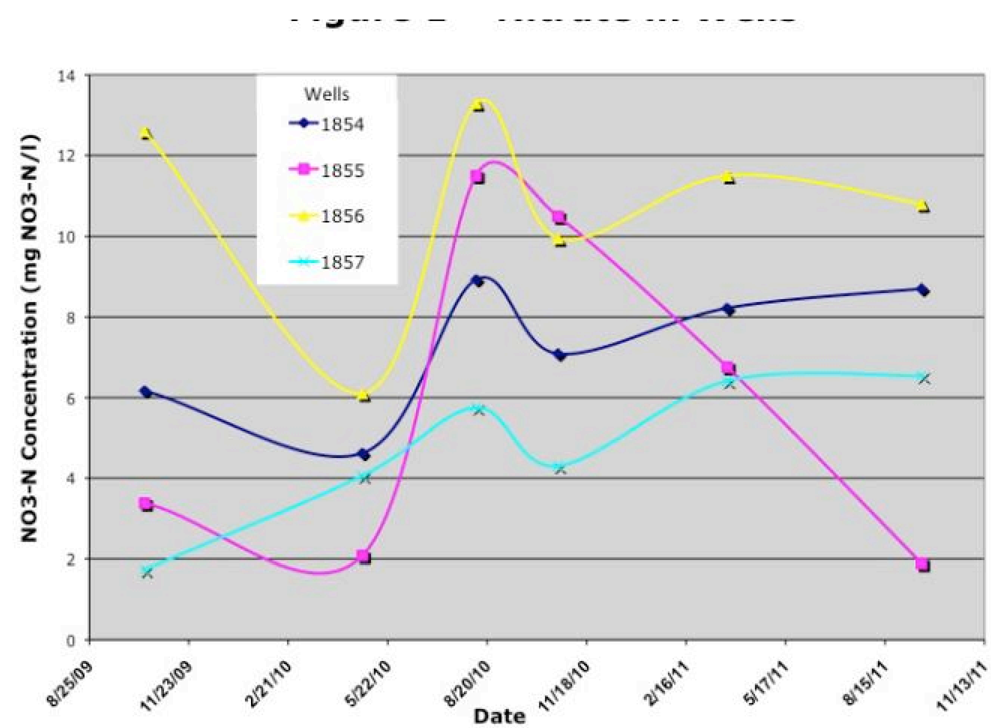

Figure15. Nitrate in well samples.

The four samples collected in the first sampling, ranged from 1.73 to 12.6 mg NO3-N /L (Figure 15). The 12.6 mg/L value was surprisingly high, except that ground water nitrate values on the shore often exceed 10 mg/L. Ammonia and TKN were all below the detection level. The analysis indicates that an annual cycle exists, with low nitrate levels occurring in April, when water is plentiful and higher nitrate levels occurring in the late summer when the water table has dropped and water for nitrate transport is scarce. Well 1855 exhibits a different pattern but that well lies in a different soil than others. While there is a lot of fluctuation there is an indication that average nitrate values for the four wells may have increased slightly, but they are within the range of 10 ppm, which is characteristic of eastern shore cropland soils (Figure 16).

Overall, the lysimeters in this study show high nitrate values in soil water for the medium and high treatments, but the well samples indicate it is not making it into the groundwater, even with very high application rates. The four wells surround the research plots so if large changes in nitrate did occur if would not be possible to determine the cause, but the higher application rates would be the logical suspect.

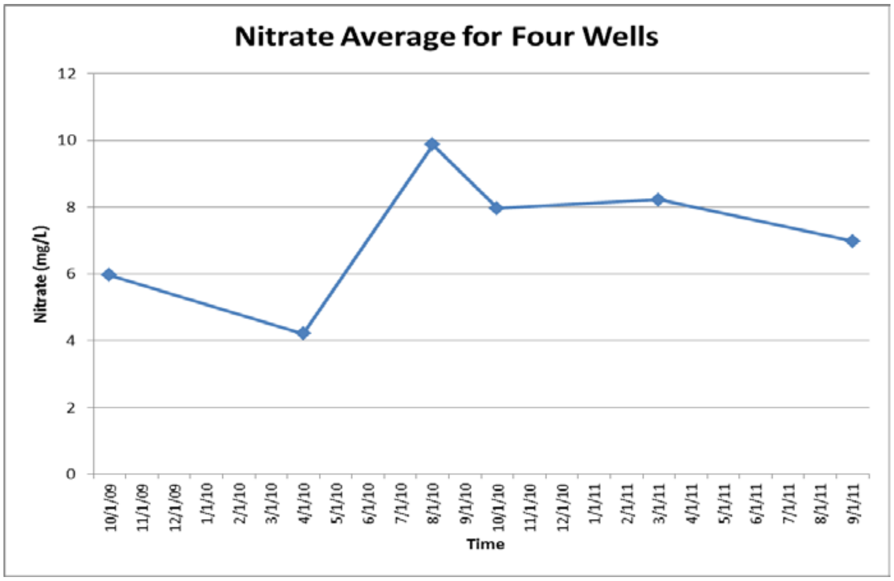

Figure 16. Average nitrate in four wells from research site. All are within 10 ppm.

Soil Phosphorous

A composite soil sample was taken from each of the 12 plots in December 2008 prior to any biosolid application. Means P values in the upper 6-inches of soil ranged from 67 to 84 ppm with a mean value of 76 ppm, with standard errors from 14 to 32 ppm. In general there were no large differences in soil P prior to the biosolid applications.

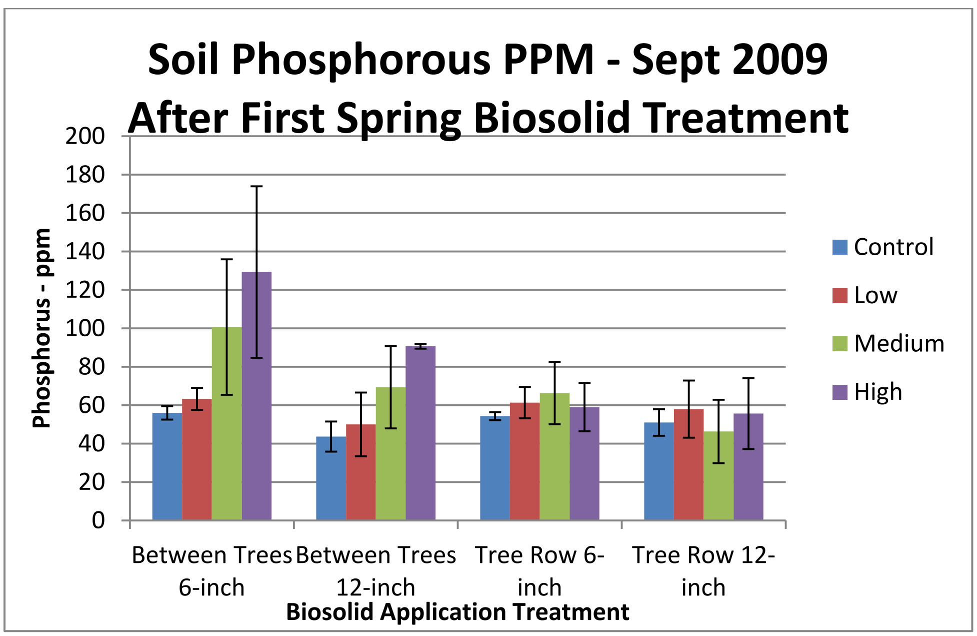

Figure17. Soil P after first 2009 spring biosolid application.

To determine the impact of the spring 2009 biosolid application on soil P, a composite soil sample was taken in September 2009 between-tree-alley rows and in the tree row (see Figure 4) at 0-6 inches deep and 6 to 12 inches deep (Figure 17). The between-tree-alley 6-inch samples were slightly higher than the pre-application samples, but the most noticeable change was the elevated P level for the medium application (101 ppm) and the even high higher value for the high application rate (129 ppm). A similar trend was seen for the 12-inch sample but the P ppm for the medium and high treatment was not as elevated, 69 and 91 ppm, respectively. The reason for the difference is due to the application method using the manure spreader, which deposited biosolids only in the between-tree-alley. The P in the low treatment was likely utilized by the grass found in the between-tree-alley, but the higher application rates of the medium and high treatments were moving through the soil profile. The lower P levels for the 6-12 inch samples indicates most of the P was residing in the soil surface, which is to be expected.

Figure 18 highlights the cumulative effect of the multiple biosolid treatments on soil P over the four growing seasons of application (2009 to 2012). The Maryland nutrient management regulations prohibited the application of manure or biosolids if the soil P is above 135 ppm (known as the Fertility Index Value or FIV in Maryland). It is clear that the first biosolid application had little impact on soil P due to the poor coverage, but once the broadcast applications were initiated in fall 2009, the soil P levels increased to levels well above the nutrient threshold that would effectively not allow additional applications.

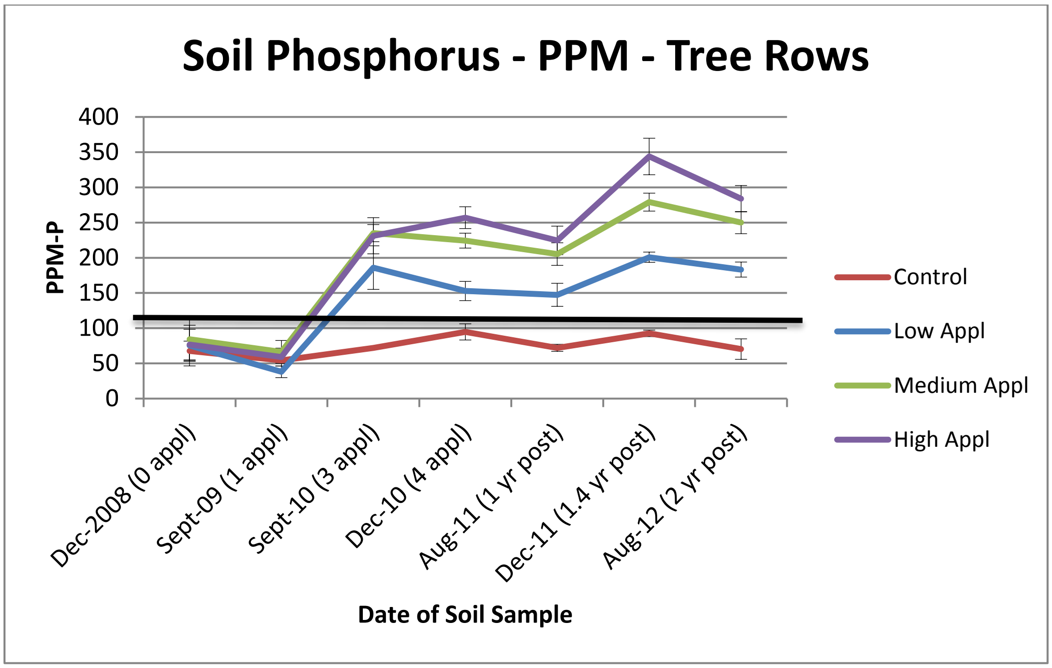

Figure 18. Level of soil P with continued application. Black line represents the soil P threshold value of 135 ppm above which additional applications would be prohibited or restricted.

DISCUSSION

The surface application of biosolids to a three year old hybrid poplar plantation on the Eastern Shore of Maryland was initiated to mimic nitrogen-based operational applications utilized in the Pacific Northwest. The medium rate represented the prescribed rate cited in research of 392 kg/N/ha [350 lb/N/ac], with the low rate half that amount, and the high rate double. Unlike industrial operations in the Pacific Northwest a grass strip was left between tree rows to minimize runoff.

Biosolid treatments had more rapid rate of diameter and biomass growth than the control, which is to be expected. All of the plots had grass strips between the tree rows and while herbicide was applied in the spring to clear the tree row, by mid-summer many weeds and vines had colonized the herbicide strips, capturing available nutrients and moisture. The production of biomass, the growth variable of greatest interest, has the fastest rate of growth with the Medium and High treatments, which were not significantly different. The lower rate of growth for the Low treatment can be partially attributed to the grass and other vegetation and that may have utilized available nutrients and moisture, leaving little for tree growth. It appears there is a threshold type of response to the treatments in this research, with only the higher rates of the High and Medium treatments able to be converted into tree diameter and above-ground biomass.

Nitrate found in soil lysimeters produced the expected trend with lowest levels of soil nitrate for the control and increasing levels with higher application rates. Using the high application rate resulted in levels of nitrate in soil water over 60 ppm but the medium rate was below that level. Hybrid poplar research has documented that the trees are capable of uptaking even higher levels of nitrate (VanHam, 2006). The fact that nitrate in soil water was not found in the wells indicates the trees or other vegetation is using the nitrate, and there may be no need for alarm with using the High treatment. However, if the maximum rate of biomass growth is desired, it makes sense to use the Medium rate since there is no apparent beneficial growth response with the high treatment but greater chance of runoff and environmental impacts.

The usual relationship between high foliar nutrient levels and high growth did not hold up across the treatments. It was expected that the control would maintain the lower low levels of foliar %N and %P reported for 2008, and the levels would increase in some stepwise fashion with higher application rates. However, the control plots had the highest %N and %P values in 2010 but a slowest rate of growth. It is obvious that nutrients are available to the trees in the control plots and other plots may have some cross plot movement of nutrients.

There are a few possible explanations for the high levels of %N and %P in the control plots and the lack of growth response. The 38% overapplication took place on October 14, 2009 very late in the season and it was followed by some rainfall events later in the month (Figure 5) that may have resulted in some runoff from adjacent plots or subsurface movement. The other possibility is that the small plot sizes allowed roots to cross over from control plots to treatment plots. Roots were found in the wells 10 feet and deeper so it is likely that all trees were trying to access the groundwater during the droughty summer months. The fall 2010 application was done in early September to provide time for decomposition and uptake by the trees and avoid potential problems. The lack of elevated levels of nitrate in the soil lysimeters would indicate that if nutrient transport occurred over the course of the study, it would have been near the surface, and did not reach down 0.61 m [2 ft] to the installed lysimeters. We have no way of knowing if root growth to adjacent plots was a factor but we do know control plots had to have some source of N and P to register such high values. Foliar nutrient levels in treatment plots could have been impacted as well.

Given that foliar N and P were present at high levels for the control, the lack of growth response may be related to the enhancement of soil physical properties provided by biosolids. McIntosh et. al (1984) found hybrid poplar steckings planted on soils, and in a climate similar to those of this study, ceased growth in July or August, but with compost applied growth continued through October. Organic wastes such as biosolids not only provide a slow release of nutrients, but improve the physical properties of the soil including porosity, water holding capacity, and bulk density (Gallardo-Lara & Nogales 1987). Hybrid poplar responds to moisture stress by allocating carbohyrates to root growth over shoot growth. Therefore, the biosolid treated plots likely had less moisture stress due to the layer of biosolids and continued growing through the droughty conditions found in the summer, allowing for increasing rates of growth in diameter and above-ground biomass. While the control plots did have access to nutrients from some source, the lack of an organic layer on the surface to alleviate drought stress may have caused the trees to direct carbohydrates produced to root growth over shoot growth, allowing access to groundwater. This study lacks the data to prove the cause for the foliar nutrient levels, but these are some possible scenarios.

A major question of this study was if nitrogen based surface applications using different application rates would be compliant with the Maryland nutrient management law which is based on phosphorus application. Treatments were split between spring and fall to allow the trees time to utilize the nutrients all year round. It is clear that soil P levels quickly increased to levels well above the acceptable Fertility Index Value (FIV) for P of 150, which corresponds 136 ppm. This occurred for all treatment levels but the absolute levels increased depending on application rate. Higher application rates had a faster increase in P levels and reached higher absolute levels. The threshold of 136 ppm was reached after the second application for all treatments, and may have exceeded it on the first if the application was broadcast across the plots.

This research documents that soil P levels will likely limit the application of biosolids to hybrid poplar plantations in locales that have nutrient management regulations based on P. It also alludes to the fact that operations using nitrogen-based application may already have extremely high soil P levels that may impact water.

The use of hybrid poplar plantations to utilize large amounts of biosolids in states with P-based nutrient management regulations is extremely unlikely. However, it does present the opportunity for additional research using lower application rates, less frequency of application, different types of biosolids, and other combinations to try to create an application system that will stay within the constraints of P-based nutrient management.

CONCLUSIONS

The research did highlight some practical problems that others should be aware of before implementing other studies.

- The suction lysimeters were not reliable and many had to be fixed in the field or excavated and re-installed. The major cause of the damage is believed to be the traveling of heavy equipment tractors and manure spreaders up and down the rows. Even though the lysimeters were in the middle of the rows the ground movement appears to pull on the plastic lines and break the vacuum. In general, lysimeters have been very reliable in many other studies, so something unique to this experiment was responsible for the problems with the lysimeters.

- Getting a consistent application of biosolids across small research plots with the Knight Side Slinger is a problem that cannot be easily solved. Careful calibration and experimental procedures have been used but application is highly variable. Any future research will need to consider the purchase of more specialized equipment for small plot application.
- To minimize the access of nutrients by tree roots from adjacent plots and movement of nutrients overland or subsurface, future studies will need to utilize at least two buffer rows of trees.
- If split applications are used, fall application in areas similar to this study should be applied in later August or very early September to allow time for the trees to utilize the nutrients and reduce the possibility of nutrient movement offsite.

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