

**University of Alberta**

**Nutrient Loading of Aspen, Jack Pine and White Spruce Seedlings for  
Potential Out-planting in Oil Sands Reclamation**

by

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## **Abstract**

Low nutrient availability and competing vegetation are some of the issues facing tree crop establishment for land reclamation in the oil sands region. A possible solution to both problems is to load seedlings with nutrients while they are in the nursery. Here, I report on results from a research project aimed at producing aspen, jack pine and white spruce seedlings that have nutrient reserves built up (or nutrient-loaded) in the nursery stage. Seedlings were either conventionally (C) or exponentially (E) fertilized or not (CK). The results showed that seedlings produced under a modified exponential fertilization regime have higher nutrient reserves than those produced conventionally with similar height and root collar diameter (RCD). The optimum rates in the modified nutrient loading model were 240, 500 and 450 mg N plant<sup>-1</sup> season<sup>-1</sup> for aspen, jack pine and white spruce, respectively.

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## Table of Contents

<b>Chapter 1. General Introduction and Literature Review .....</b>	<b>1</b>
1. Boreal forest ecosystems and oil sands reclamation .....	1
2. Ecological requirements of aspen, jack pine and white spruce .....	5
2.1. Trembling Aspen .....	6
2.2. Jack Pine.....	7
2.3. White Spruce .....	9
3. Nutrient loading: the concept and application.....	10
3.1. Concept .....	10
3.2. Application .....	14
3.2.1. Container size.....	15
3.2.2. Fertilization rate.....	19
3.3. Nutrient loading models for nursery fertilization.....	20
3.3.1. Exponential model.....	20
3.3.2. Modified exponential model.....	21
3.3.3. Model parameters for aspen, jack pine and white spruce.....	22
4. Summary .....	23
5. Objectives.....	24
6. Thesis structure .....	24
7. References .....	26
<b>Chapter 2. Nutrient Loading of Aspen, Jack Pine and White Spruce Seedlings Using an Exponential Model .....</b>	<b>38</b>
1. Introduction .....	38
2. Materials and Methods .....	42
2.1. Plant material and growing conditions .....	42
2.2. Fertilization Models .....	44
2.3. Plant sampling and chemical analysis .....	45
2.4. Statistical analysis .....	46
3. Results and Discussion.....	47
3.1. Aspen.....	47
3.2. Jack pine.....	49
3.3. White Spruce .....	51

4. Conclusions .....	52
5. References .....	54
<b>Chapter 3. Nutrient Loading of Aspen, Jack Pine and White Spruce Seedlings Following a Modified Exponential Model .....</b>	<b>71</b>
1. Introduction .....	71
2. Materials and Methods .....	75
2.1. Plant material and growing conditions .....	75
2.2. Fertilization model .....	76
2.3. Plant sampling and chemical analysis .....	78
2.4. Vector diagnosis .....	79
2.5. Statistical analysis .....	81
3. Results and discussion.....	81
3.1. Aspen.....	81
3.1.1. Seedling growth and nutrition.....	82
3.1.2. Identifying the optimum nutrient loading rate.....	84
3.2. Jack pine.....	86
3.2.1. Seedling growth and nutrition.....	86
3.2.2. Identifying the optimum nutrient loading rate.....	87
3.3. White spruce.....	88
3.3.1. Seedling growth and nutrition.....	88
3.3.2. Identifying the optimum nutrient loading rate.....	89
3.4. Similar patterns of total nitrogen and available nitrogen in growth media and $\delta^{15}\text{N}$ data of seedlings with the three species.....	91
3.5. Discussion of model suitability .....	92
4. Conclusions .....	93
5. References .....	95
<b>Chapter 4. Synthesis of Results and Recommendations for Future Research .....</b>	<b>129</b>
1. Synthesis .....	129
2. Recommendations for future research.....	131
3. References .....	133

## List of Tables

<b>Table 1.1</b> Available cavatiy size of styroblocks for containerized seedlings production .....	34
<b>Table 1.2</b> The effects of container cavity size on seedling quality .....	35
<b>Table 2.1</b> Sampling schedule during the experiment .....	57
<b>Table 3.1</b> Sampling schedule during the experiment .....	99

## List of Figures

<b>Figure 1.1.</b> Relationships among nutrient supply, plant growth and tissue nutrient content and concentration. ....	36
<b>Figure 1.2.</b> Schedule of fertilizer additions by seedling age .....	37
<b>Figure 2.1.</b> The detailed fertilizer schedule for three species. ....	58
<b>Figure 2.2.</b> Aspen stem height under different treatments over the entire growth period in 2009 .....	59
<b>Figure 2.3.</b> Aspen component dry mass under different treatments over the entire growth period in 2009 .....	60
<b>Figure 2.4.</b> Nitrogen content data of aspen under different treatments over the entire growth period in 2009 .....	61
<b>Figure 2.5.</b> Relationships among nutrient supply, plant growth and tissue nutrient content and concentration for aspen at 12 <sup>th</sup> week in 2009. ....	62
<b>Figure 2.6.</b> Jack pine stem height under different treatments over the entire growth period in 2009 .....	63
<b>Figure 2.7.</b> Jack pine component dry mass under different treatments at different sampling times in 2009. ....	64
<b>Figure 2.8.</b> Nitrogen content data of jack pine under different treatments over the entire growth period in 2009 .....	65
<b>Figure 2.9.</b> Relationships among nutrient supply, plant growth and tissue nutrient content and concentration for jack pine at 18 <sup>th</sup> week in 2009 .....	66
<b>Figure 2.10.</b> White spruce stem height under different treatments over the entire growth period in 2009 .....	67
<b>Figure 2.11.</b> White spruce component dry mass with different treatments after 18 weeks of growth in 2009 .....	68
<b>Figure 2.12.</b> Nitrogen content data of white spruce under different treatments over the entire growth period in 2009 .....	69
<b>Figure 2.13.</b> Relationships among nutrient supply, plant growth and tissue nutrient content and concentration for white spruce at 18 <sup>th</sup> week in 2009 .....	70
<b>Figure 3.1.</b> The detailed fertilizer delivery schedule for three species .....	100
<b>Figure 3.2.</b> Interpretation of directional differences (vectors) in nutrient concentration, nutrient content, and dry weight between plants of contrasting different treatments .....	101
<b>Figure 3.3.</b> Aspen morphological data under different treatments over the entire growth period in 2010 .....	102
<b>Figure 3.4.</b> Aspen component dry mass under different treatments over the entire growth period in 2010 .....	103
<b>Figure 3.5.</b> Aspen N concentration under different treatments in the last collection (12 <sup>th</sup> week) in 2010 .....	104
<b>Figure 3.6.</b> Components nitrogen, phosphorus and potassium content of aspen	



under different treatments over the entire growth period in 2010 .....	106
<b>Figure 3.7.</b> Relationships among nutrient supply, plant growth and tissue nutrient content and concentration of aspen at 12 <sup>th</sup> week in 2010 .....	107
<b>Figure 3.8.</b> Component $\delta^{15}\text{N}$ of aspen seedlings under different treatments over the entire growth period .....	108
<b>Figure 3.9.</b> Vector nomogram of relative change in plant dry mass, N content, and concentration in aspen fertilized at different rates .....	109
<b>Figure 3.10.</b> Nitrogen information in styroblocks for aspen in 2010.....	110
<b>Figure 3.11.</b> Pine morphological data under different treatments over the entire growth period in 2010 .....	111
<b>Figure 3.12.</b> Pine component dry mass under different treatments over the entire growth period in 2010 .....	112
<b>Figure 3.13.</b> Pine N concentration under different treatments in the last collection (in the 21 <sup>st</sup> week) in 2010.....	113
<b>Figure 3.14.</b> Component nitrogen, phosphours and potassium content of pine seedlings under different treatments over the entire growth period in 2010 .....	115
<b>Figure 3.15.</b> Relationships among nutrient supply, plant growth and tissue nutrient content and concentration of jack pine at week 21 in 2010 .....	116
<b>Figure 3.16.</b> Component $\delta^{15}\text{N}$ of pine seedlings under different treatments over entire growth period .....	117
<b>Figure 3.17.</b> Vector nomogram of relative change in plant dry mass, N content, and concentration in jack pine fertilized at different rates.....	118
<b>Figure 3.18.</b> Nitrogen information in styroblocks for pine in 2010 .....	119
<b>Figure 3.19.</b> Spruce morphological data under different treatments over the entire growth period in 2010 .....	120
<b>Figure 3.20.</b> Spruce component dry mass under different treatments over the entire growth period in 2010.....	121
<b>Figure 3.21.</b> Spruce N concentration under different treatments in the last collection (23 <sup>rd</sup> week) in 2010 .....	122
<b>Figure 3.22.</b> Components nitrogen, phosphours and potassium content of spruce seedlings under different treatments over the entire growth period in 2010 .....	124
<b>Figure 3.23.</b> Relationships among nutrient supply, plant growth and tissue nutrient content and concentration of spruce at week23 in 2010.....	125
<b>Figure 3.24.</b> Component $\delta^{15}\text{N}$ of spruce seedling under different treatments over entire growth period .....	126
<b>Figure 3.25.</b> Vector nomogram of relative change in plant dry mass, N content, and concentration in white spruce fertilized at different rates .....	127
<b>Figure 3.26.</b> Nitrogen information in styroblocks for spruce in 2010 .....	128

## **Chapter 1. General Introduction and Literature Review**

### **1. Boreal forest ecosystems and oil sands reclamation**

Boreal forest ecosystems, predominantly evergreen forests in the cold-temperate region, are some of the most outstanding terrestrial ecosystems due to their large forested land area and wide distribution in the world (Bruenig, 1987). In Canada, the boreal forest is one of the largest ecosystems, covering approximately 58% of the land base (Anielski and Wilson, 2005). The Alberta oil sands region is located in the Cold Lake, Peace River and Athabasca regions in the North America Boreal Plain and cover approximately 142,200 km<sup>2</sup>. Currently, approximately 856,000 barrels of bitumen per day are produced in the mineable portion of the Athabasca oil sands region. From 1967 to 2006, the cumulative disturbance for oil sands development to boreal forests was 47,832 ha. To date only 0.2% of the total land base disturbed by mining has been certified as reclaimed land by the Government of Alberta (Rowland et al., 2009). In addition, open-pit mining leaves a reconstructed landscape of overburden dumps and tailings deposits that require reclamation targeting self-sustaining and locally common ecosystems. Thus, there are still a very large area of disturbed land left in the oil sands region that requires reclamation (Fung and Macyk, 2000).

Reclamation is defined as the stabilization, contouring, maintenance, conditioning or reconstruction of the surface of land with the aim of restoring disturbed land to its productive stage. In Alberta, mining companies are required

to reclaim the land after mine closure. Reclamation is an essential component of responsible oil sands development (Gentes et al., 2006; Grant et al., 2008).

Reclamation materials available for use as cover soil include LFH (a thin organic layer on the forest floor consisting of litter (L), fibric (F) and humic (H) material), peat-mineral mix, upland surface soil and transitional soil (BGC Engineering Inc., 2010). A transitional soil is a soil with properties intermediate between two different soils and genetically related to them. Cover soils are either stored in a stockpile for use at a later date or directly placed onto areas to be reclaimed. The biological properties and nutrient capital in upland surface soils and transitional soils make them valuable reclamation materials for use as a cover soil (Strong and Laroi, 1983; Fisher and Binkley, 2000; Mackenzie and Naeth, 2010).

The majority of the landscape within the mineable oil sands region is comprised of organic soils. Because of the widespread availability of organic soils, cover soils comprised of peat or peat-mineral mix have been and will continue to be the dominant reclamation material used on post-disturbance landscapes for most mines (Barbour et al., 2007). Peat is commonly used as an amendment to overburden materials for reclamation by Syncrude in Fort McMurray (Land Resources Network Ltd., 1993). There are many advantages of using peat as an amendment for oil sands reclamation (Logan, 1975; Land Resources Network Ltd., 1993). It can improve physical conditions of the soil and provide humus and modest nutrient sources. In addition, peat is longer lasting

than other organic amendments. Peat or peat-mineral mix provides plants with nutrients and increases the soil water-holding capacity and thus alters the physical, chemical and biological properties of the reclaimed soil.

Amendment of the extracted sand with peat or peat-mineral mix may also alter the pH of the reclaimed soil, which would ultimately affect nutrient availability (Danielson et al., 1983). Low nutrient availability in peat based materials is a major concern as it can restrict plant growth (Arii and Turkington, 2002). Nitrogen (N) and phosphorus (P) are generally present in the peat in organic forms that are slowly available to plants (Puustjarvi and Roberson, 1975). Of all the major plant nutrients, N is often the most important determinant of plant growth and crop yield. Plant growth and crop yield usually increase when N is added, despite the presence of N in the soil. This is because most of the N in the soil is stored within the soil humus in forms that plants cannot access (Lee et al., 1983). Thus, the low N availability in reclamation material will significantly affect plant growth. Organic soils contain only low amounts of available micronutrients. Deficiencies of copper, zinc, manganese and boron were common in crops grown entirely on peat (Brady, 1990). In a study conducted by Logan (1978) that examined the use of several types of peat to amend tailings sand, it has been found that all treatments required additions of fertilizers to increase plant growth, regardless of peat type. In addition, in a comparison of different reclamation treatments to natural ecosystems, Rowland et al. (2009) concluded

that treatments adding fertilizers to peat-mineral mix and/or to underlying subsoil over clean overburden could help ecosystem processes of reclaimed forests to mimic those of the natural boreal forest because peat-mineral mix and underlying subsoil have low nutrient availabilities.

To eliminate potential nutrient deficiencies that could limit the success of reclamation, broadcast fertilization is often used to supply limiting nutrients to reclaimed sites. However, fertilization not only benefits the target crop species, it also benefits the non-target or competing species and most often in fact creates a greater stress (e.g., through increased competition) to the crop species (Timmer, 1997), as the competing species may benefit more than the planted tree species as newly planted tree seedlings have limited root development and limited ability to take up the applied nutrients. For example, it has been found that early seedling growth can be affected by the competition of nutrients and moisture in the field (Anderson et al., 2001). Moreover, a study conducted in the boreal mixedwood forests of Saskatchewan by using two white spruce and two jack pine experimental sites also proved that interspecific competition for soil N was the principle cause of reduced white spruce and jack pine seedling growth during the early establishment phase under conditions of adequate soil moisture (Hangs et al., 2004). The negative effects of interspecific competition on forest trees have been investigated in many different studies (Stewart et al., 1984; Walstad and Kuch, 1987; Staples et al., 1999; Robinson et al., 2001). In the interior of British

Columbia, vegetative competition such as shrubs can seriously hinder regeneration of white spruce (Eis, 1981). In addition, the applied nutrients may also be lost from the system through leaching and other processes with the limited ability of seedlings to take up the nutrients. Competition for nutrients can be severe, and tree growth can be limited.

In order to solve the low nutrient availability and competition problems in oil sands reclamation, a technique that can be used is the nutrient loading technique that increases the internal nutrient reserve in seedlings in the nursery phase, to provide the seedlings an advantage in gaining fast establishment and better survival under competing environments (Malik and Timmer, 1998; Salifu et al., 2009), as the extra nutrients stored in the nutrient loaded seedlings can be retranslocated to the new growth to meet the growth requirement and such nutrients are only available to the target trees and not the competing species.

## **2. Ecological requirements of aspen, jack pine and white spruce**

Trembling aspen (*Populus tremuloides*), jack pine (*Pinus banksiana*) and white spruce (*Picea glauca*) are major tree species native to and routinely planted in the oil sands region (Alberta Environment, 2011); moreover, more than 40% of the replanted area is covered by white spruce and jack pine as they are important commercial tree species in the boreal forest in Canada (B. Hadden (Canadian

Forest Service), personal communication). Due to the importance of these species in the oil sands region, if nutrient loaded seedlings of these species can be proven advantageous when outplanting on reclaimed sites in the oil sands region, there could be large gains in efficiency, survival rate, and initial growth rates. Thus, the ecological requirements for each species need to be identified.

## **2.1. Trembling Aspen**

Trembling aspen, the most widely distributed tree species in North America, is a vital component of almost every forest ecosystem in western North America. Trembling aspen accounts for 37% of the gross merchantable volume in the productive forests of the three Prairie Provinces (Manitoba, Saskatchewan and Alberta) in Canada and has also become increasingly important to the forest products industry in the region in the past 20 years (Canadian Council of Forest Ministers, 1997). In northern Alberta, aspen is not only widely distributed but also has economic importance. It is a tall, fast growing deciduous tree, usually with a height of 20-25 m and a diameter of 20-80 cm at maturity. The regeneration of aspen is abundant in the boreal forest. Aspen is extremely intolerant of shade and it reproduces primarily by vegetative sprouting from the pre-existing roots of parent trees. Aspen requires full sunlight to thrive, allowing it to grow very well in areas cleared by fire. However, this trait also makes it very sensitive to competition from shade tolerant species such as spruce and fir in mixed species

forests. It reproduces primarily by root suckering throughout most of its western range. Aspen seedlings require bare mineral soil and constant moisture to survive (McDonough, 1979). Aspen have a short growing season (barely 80 days long) with a wide climatic range, with temperatures varying from -60 degrees in winter to over 38 degrees in summer (Shepperd et al., 2006). They can survive in low soil nutrient conditions and can photosynthesize in low light and alter leaf morphology depending on moisture availability. Optimum pH level for growing aspen is between 5 and 7 (DesRochers et al., 2003). It typically grows on upland sites with balsam poplar occupies sites with a high water table (Shepperd et al., 2006).

There is an increasing interest in establishing aspen plantations on marginal agricultural land or reclamation lands because of the importance of this species in North America (Van den Driessche et al., 2003). Since it is the dominant species on upland forest sites in the oil sands region – it is desirable to use it for reclamation. Early study (Coyne and Vancleve, 1977) showed that height and diameter of aspen would be increased substantially by applying N, P and K fertilizers.

## **2.2. Jack Pine**

In Canada, jack pine is a widely distributed boreal forest species with its range extending from Nova Scotia to northern British Columbia. It is an important



commercial timber species in the United States and Canada. Jack pine is a small- to medium-sized, evergreen, boreal conifer that ranges from 9-22 m in height. It grows well in subarctic and cool temperate climates. It can grow in dry, acidic sandy soils with medium to dry moisture conditions, but also in loamy soils, thin soils over bedrock, peat, and soils over permafrost (Klinka et al., 1999). Jack pine does not grow on alkaline soils and grows poorly on calcium-rich soils (Klinka et al., 1999). The optimum soil pH level for this species is between 5.0 and 7.5. This pine is an early successional species commonly found on sandy, nutrient-poor sites. Thus, it is a suitable reclamation species for the oil sands mining areas (Franklin et al., 2002). In the oil sands region, jack pine is a natural component on well to rapidly drained sites.

In order to successfully produce jack pine seedlings for outplanting, a high (larger than 24 mg N plant<sup>-1</sup> season<sup>-1</sup>) and optimal nutrient regime (at 24 mg N plant<sup>-1</sup> season<sup>-1</sup>) is usually applied during stock production to improve seedling growth rate and shorten the production cycle (Tan and Hogan, 1997). When outplanting in a drought environment, a modified nutrient regime may be necessary to produce stocks capable of reasonable initial growth rates while maintaining an enhanced level of drought tolerance. Consideration should be given to matching the level of fertilization with the nature of drought on the site when outplanting jack pine seedlings (Tan and Hogan, 1997).

### **2.3. White Spruce**

White spruce is widely distributed over Canada and is an important tree species both economically and ecologically for Alberta. It is important commercially for pulpwood and construction lumber production. White spruce is a medium-sized evergreen conifer that ranges from 15 to 40 m in height. White spruce generally occurs on well-drained soils in alluvial and riparian zones with a pH of 4.0-5.5 (Klinka et al., 1999). It grows well on loams, silt loams, and clays, but rather poorly on sandy soils, and also grows poorly on sites with high water tables. The suitable soil pH for growing white spruce is 4.7 to 7.0. Soil properties such as fertility, temperature, and structural stability are partial determinants of the ability of white spruce to grow in the extreme northern latitudes. White spruce tolerates shade, but grows best in full sunlight. In boreal forests, white spruce often grows in mixtures with other species, mostly with trembling aspen. In these areas, white spruce typically occurs in the understory and aspen usually dominates the canopy layer for an extended period of time (Filipescu and Comeau, 2007). White spruce grows well on medium and rich sites provided a moderate humus formation exists. This humus form can favor the establishment and growth of shade-tolerant white spruce after wildfires or clearcutting (Klinka et al., 1999).

In silviculture, it always takes several years for transplanted white spruce nursery stock to have greater height than that of the surrounding brush. Moreover, survival and growth of planted white spruce trees are often low (Margolis and

Brand, 1990). Initial nutrient deficiency is often a factor contributing to early reduction of white spruce seedlings regardless of many other possible causes of plantation failure. This would indicate the importance of increasing initial white spruce seedling's nutrient storage in the nursery phase by using a suitable method (Mullin and Bowdery, 1977; Sutton, 1992).

### **3. Nutrient loading: the concept and application**

#### **3.1. Concept**

Seedling survival and growth after outplanting can be influenced by nursery culture. Although many factors influence growth, the main contributors are adequate sunlight, ample water supply, proper nutrition and appropriate temperature and growing season length (Ingestad, 1979). Improved nutrient availability after germination can result in high root growth and water uptake, which may enhance seedling resistance to other stresses (Krammer, 1969; Larcher, 1980).

For containerized seedlings under greenhouse culture, they always have two major growing periods in their production cycle, an intensive fertilization period during the rapid exponential growth phase to meet nutrient consumption demands and an extended period of growth when fertilization is reduced during hardening to prepare seedlings for field planting (Ritchie and Dunlap, 1980; Miller and Timmer, 1994).

Nursery-grown seedlings often exhibit nutrient deficiency soon after planting because slow regeneration and extension of the root system restrict exploitation and absorption of soil nutrients (Burdett et al., 1984; Burdett, 1990; Van den Driessche, 1991). With the purpose of improving outplanting performance, nutrient loading was suggested to increase internal nutrient reserves of nursery-grown seedlings. The internal nutrient reserve of the seedlings after the hardening period is highly related to the growth after planting and the accumulated internal nutrients can be retranslocated and reutilized to reduce the risk of nutrient deficiency (Chapin et al., 1990).

For conventional (traditional) fertilization, fertilizers are often supplied in equal doses at fixed intervals over the growing season. The operational dose applied to black spruce container stock was around  $10 \text{ mg N plant}^{-1}$  season<sup>-1</sup> (Timmer and Aidelbaum, 1996). For this practice, it has been found that high nutrient inputs in the early growing season not only reduce survival of fungal inoculums but also limit the capacity to initiate further infection (Ruehle and Wells, 1984; Gagnon et al., 1991). Furthermore, as nutrient additions are poorly synchronized with the relative growth rate and nutrient demand of seedlings during the exponential growth phase of the greenhouse rotation, constant addition rate schedules may also result in a surplus of nutrients at the beginning of the culture and a deficiency by the end of the nursery production period (Timmer et al., 1991; Imo and Timmer, 1992).

Nutrient loading, or the so called exponential fertilization practice, is a fertilization practice in the nursery that increases the plant's internal nutrient reserve by inducing luxury consumption of nutrients during its growth without altering seedling size (Imo and Timmer, 2002). In comparison to non-loaded seedlings, loaded container-grown seedlings exhibit the capacity to retranslocate nutrients for current growth rather than conserving them in old tissue (Malik and Timmer, 1995). It has also been found that the nutrients that accumulate in response to exponential loading are more readily available for retranslocation than is the case for conventional seedlings (Timmer, 1997). Nutrient loaded seedlings exploit greater internal nutrient reserves to promote early establishment success of outplanting conifers and hardwoods (Salifu and Timmer, 2003b; Birge et al., 2006). The higher nutrient reserves in the seedlings would lead to greater partitioning of nutrients to current needles, promoting photosynthesis and nutrient uptake (Nambiar and Fife, 1991). Compared with the conventional treatment, nutrient loaded seedlings can stimulate ectomycorrhizal development as the applied fertilization level of this technique is lower than that for conventional in the early growing season (Quoreshi and Timmer, 1998). For instance, Quoreshi and Timmer (1998) pointed out that exponential fertilization stimulated ectomycorrhizal development (49-85%) when compared with conventional fertilization (22-26%). While seedlings produced with the conventional method always has the problems with decreasing internal nutrient concentration during

the growth period, the nutrient loaded seedlings can avoid this problem because it has greater plant nutrient reserves and residual fertilizer in the growing media (Miller and Timmer, 1997).

Exponential fertilization also better synchronizes nutrient supply with seedling demand during its growth period in the nursery by inducing luxury consumption of nutrients (Timmer, 1997; Birge et al., 2006). In the exponential fertilization regime, steady state nutrition was created for the *Picea mariana* seedlings by matching nutrient supply to the exponential seedling growth in the nursery (Timmer and Munson, 1991; Timmer et al., 1991). It was also found that such a regime matches the nutrient uptake of naturally growing vegetation. These theories were combined together in nursery practice to improve the performance of seedlings after outplanting (Malik and Timmer, 1998; Salifu and Timmer, 2003a).

The proposed conceptual model (Figure 1.1) illustrates relationships between growth and nutrient conditions of the plant and nutrient supply (Timmer, 1997). By applying fertilizer in an exponential manner, nutrient supply can more closely match plant demand and improve nutrient uptake and use efficiency. The luxury consumption in this fertilization method is intended to ensure that seedlings store nutrients as reserves for later use when they are outplanted.

### 3.2. Application

Nutrient loading may improve seedling resistance to nutrient and moisture stress after outplanting by promoting root production (Van den Driessche, 1985; Timmer and Munson, 1991) and improving seedlings' survival against competing vegetation (Malik and Timmer, 1996). Management of competing hardwoods for successful establishment of young seedlings is critical as severe competition from naturally regenerating hardwoods may hamper seedlings' establishment (Brumelis and Carleton, 1988). Although there are many methods (such as mechanical, chemical and biological) to control weeds, these methods always need to be considered on a site-specific basis since tree and weed responses to treatments usually vary among sites (Burger and Pierpoint, 1990). In addition, these methods only reduce weed competition to the extent that they relieve significant interference with trees based on some specified economic or biological criteria. Thus, these methods are not that suitable for all sites. However, nutrient loading has been found to be very effective on sites with high-competition in terms of growth and nutritional responses by comparing with other methods for black spruce (Imo and Timmer, 1999).

Successful outplanting results of nutrient loaded seedlings in many studies that have been conducted in recent years with various species such as black spruce (*Picea mariana*), white pine (*Pinus monticola*), holm oak (*Quercus ilex* L.) as well as northern red oak (*Quercus rubra*) and white oak (*Quercus alba*) have

all supported the application of this technology in various sites of North America to improve seedlings' potential to extend new roots and to survive the competing vegetation (Timmer, 1997; Imo and Timmer, 2001; Dumroese et al., 2005; Thorpe and Timmer, 2005; Birge et al., 2006; Oliet et al., 2009; Salifu et al., 2009).

To apply the nutrient loading method in nursery culture, some limiting factors such as container size and fertilizer rate that limit seedling growth and nutrient availability should be considered.

### **3.2.1. Container size**

In the past several decades, the production of containerized seedlings has dramatically increased. The primary function of using containers to grow seedlings is to provide water, air, and mineral nutrients in the growing medium for seedlings. Containerized seedling production is typically done under greenhouse conditions, allowing intensive management and year round production. The containers can be used to reduce root coiling and encourage root pruning at the base of the cell to favor a more fibrous root system (NeSmith and Duval, 1998). In comparison with bareroot stocks, containerized seedlings survived and grew better on poor sites after outplanting (Winter, 1980; Sloan et al., 1987). In addition, containerized seedlings offer more flexibility. For example, seedlings can be held in containers while waiting for suitable conditions for planting. Handling of containerized seedling is generally easier as seedlings are



less bulky and less prone to root desiccation.

At present, styroblocks are most commonly used in Canadian nurseries for containerized seedling production. A range of volumes from 8 to 3200 cm<sup>3</sup> can be used for specific greenhouse production and for meeting outplanting requirements. The commercially available cavity sizes for containerized seedling production are listed in Table 1.1.

In producing containerized seedlings, container types need to be considered in order to improve seedling growth and development. Even though there are containers with many different cavity sizes, many nurseries prefer to use containers with small cavity sizes as they are less expensive and the nursery can grow more seedlings per unit greenhouse space. However, there is a trade-off between the number of seedlings produced per unit greenhouse space and seedling size for most species (Tinus, 1979).

In a nursery environment, seedling development can be influenced by the growing regime as the physical restriction of the root system affects the availability of both water and nutrients (McConnaughay and Bazzaz, 1991).

McConnaughay and Bazzaz (1991) suggested that large container cavity volumes would result in greater water and nutrient availability, and more space for root development although differences in responses can occur among species. Various studies have been conducted to support that hypothesis (Table 1.2). In a research conducted by Dumroese et al. (2011), increasing container cavity volume from 50

to 656 mL yielded 200% more height and stem diameter growth for *Acacia koa* seedlings (Dumroese et al., 2011). In another study, the 270 mL block-type was selected for Indian sandalwood (*Santalum album* L.) production as it had additional advantages, such as ease of handling and lower cost (Annapurna et al., 2004). Another study showed superior growth of deciduous species such as Chinese pistache (*Pistacia chinensis*) and coniferous trees such as deodora cedar (*Cedrus deodara*), loblolly pine (*Pinus taeda*), Japanese black pine (*P. thunbergi*), red pine (*P. resinosa*), Scotch pine (*P. sylvestris*), and Afghan pine (*P. eldarica*) when they were seeded into the largest volume (680 mL) propagation container and the positive effects remained evident through the second growing season for all species (Dillon and Whitcomb, 1981). For species with a strong taproot, such as *Quercus* spp., the depth of container cavity need also be considered. For species with heavy lateral roots, narrow and long containers are not suitable (Dominguez-Lerena et al., 2006). For most Mediterranean species, such as *P. pinea*, tissue nutrient concentration, transplanting survival rate and growth all increased with container cavity volume (Dominguez-Lerena et al., 2006).

In general, seedling growth during the greenhouse production phase increases with increasing container volume between lower and upper volume limits. White spruce and jack pine seedling growth was restricted and was generally substandard in containers with cavity volumes less than 30 cm<sup>3</sup> (mL) (Sutherland and Day, 1988). Although no upper limit was found for these species, it was

suggested that larger container cavity volumes would be better suited for seedlings grown for two seasons in the greenhouse, and for container seedlings that were outplanted (Sutherland and Day, 1988). Dominguez-Lerena et al. (2006) suggested that the largest stone pine seedlings were produced with container cavity volumes of 300–400 mL with a depth/diameter ratio of 4. Under these criteria, production costs were also acceptable for the nursery. In addition, most Spanish nurseries grow Mediterranean species in container volumes larger than 200 mL (Villar-Salvador et al., 2005). A container volume of 336 mL was commonly used to grow aspen, jack pine and white spruce seedlings as it has been practiced in the Smoky Lake Forest Nursery in Alberta to produce seedlings for oil sands reclamation, while a container volume of 250 mL for aspen, and 125 mL for jack pine and white spruce were used for reforestation in other sites (L. Lafleur (Smoky Lake Forestry Nursery), personal communication). Thus, 336 mL was selected to be the cavity size in this project for growing aspen, jack pine and white spruce seedlings.

Containers with large cavity size will not only improve the success for seedling growth in the nursery but also create better seedling growth and survival post-planting. In past studies, researchers found that the height of white spruce seedlings grown in large volume containers (120 mL) were taller than those grown in smaller containers (40 mL) (McMinn, 1981; Walker and Ball, 1981). There was also an 8-10% increase in white spruce survival when container size

tripled in size (McMinn, 1981). For *P. pinea*, containers with larger rooting volume resulted in seedlings with larger height and diameter, greater nutrient content, and better field performance (Dominguez-Lerena et al., 2006).

### **3.2.2. Fertilization rate**

Fertilization in the nursery can influence seedling morphology and physiology and transplanting performance. N is usually the most important nutrient for seedlings in the nursery (Nambiar and Sands, 1994). High N fertilization rate using the nutrient loading method can promote seedling growth, photosynthetic rate, nutrient concentration and root growth capacity (Van den Driessche, 1988). The optimal amount of fertilizer to be applied in nursery seedling production is defined as the amount needed to grow a seedling that maximizes its nutrient content and stress resistance without causing morphological imbalances. Excessive N fertilization will reduce frost and drought hardiness and transplanting performance (Van den Driessche, 1988).

Generally, good transplanting performance is obtained when N fertilization rate is greater than 70 mg plant<sup>-1</sup> for most species (Van den Driessche, 1988). However, the exact amount for each species may vary. For red oak and white oak seedlings, the sufficiency and optimum rates were determined to be 0.84 and 1.68 g N per seedlings per season, respectively (Birge et al., 2006). For holm oak 100 mg N plant<sup>-1</sup> season<sup>-1</sup> has been found to be the optimal fertilization rate (Oliet et

al., 2009). Seedling biomass was maximized at sufficiency ( $30 \text{ mg N plant}^{-1} \text{ season}^{-1}$ ) and tissue N content peaked at the optimum loading rate ( $64 \text{ mg N plant}^{-1} \text{ season}^{-1}$ ) for black spruce (Salifu and Timmer, 2003b). Although the optimal fertilization rate will vary for different species, the fertilization rates from these studies can all set as reference rates for future studies. Since container size and fertilization rate all need to be considered in nutrient loading, it is crucial to examine the relationships between container size and fertilization rate. Even though there is no optimum relationship that can fit all species, many studies suggested that within each fertilizer rate, seedlings generally grew larger as container volume increased (Broschat, 1981; Van den Driessche, 1988; Pinto et al., 2008; Dumroese et al., 2011). This relationship has been shown for a variety of species.

### **3.3. Nutrient loading models for nursery fertilization**

The two main models used in the nutrient loading method are the exponential model and the modified exponential model (as shown in Figure 1.2). Details for these models are described below.

#### **3.3.1. Exponential model**

Exponential functions for each species to match nutrient supply with seedling growth (Timmer, 1997; Salifu and Timmer, 2003b) use the following model:

$$N_T = N_S (e^{rt} - 1) \quad (1)$$

where  $r$  is the relative addition rate required to increase  $N_S$  (N content, mg plant<sup>-1</sup> season<sup>-1</sup>), in seed or the initial N content) to a final N content ( $N_T + N_S$ ), and  $N_T$  is the desired amount to be added in a number ( $t$ ) of fertilizer applications. This fertilizer application then spread over the entire growth period of seedlings in the nursery.

The quantity of fertilizer to be applied on a specific day ( $N_t$ ) over the growth period can be computed using Eq. (2):

$$N_t = N_S (e^{rt} - 1) - N_{t-1} \quad (2)$$

where  $N_{t-1}$  is the cumulative amount of N added up to the previous application.

### 3.3.2. Modified exponential model

Timmer and Armstrong (1987) and Timmer et al. (1991) have shown the possibility of applying the exponential model to soil-like cultures. However, there is a problem with reduced nutrient accessibility by small root systems after germination. The exponential model involves the addition of very small amounts of nutrients initially, which may be immobilized in the soil substrates in contrast to nutrient-solution cultures where there is little restriction on nutrient mobility and availability to roots. Thus, it is necessary to modify exponential nutrient additions to compensate for these differences in the growth media to improve seedling's early root development (Timmer et al., 1991). Overall, by comparing

the pure exponential model, this model increases initial rates of fertilizer addition to facilitate nutrient supply in juvenile growth and avoid over-fertilization by inducing potential nutritional imbalances near the end of the season (Timmer et al., 1991).

For this model, an amount of N called N compensation ( $N_c$ ) is initially subtracted from the last two applications calculated from eq. (2). Then  $N_c$  is delivered exponentially within the compensation period to correspond with the exponential expansion of the root system based on Eq. (3):

$$N_c = N_0 (e^{-rt} - 1) \quad (3)$$

where  $r$  is the relative addition rate required to increase  $N_0$  to  $N_c$ ;  $t$  was the compensation period;  $N_0$  is the final amount of nutrient added in the compensation period (Imo and Timmer, 1992; Jacobs and Timmer, 2005).

### **3.3.3. Model parameters for aspen, jack pine and white spruce**

In these two models,  $N_s$  can be easily determined by measuring the initial seedling's N concentration and initial biomass of seedlings. Since there is no publication available for optimal nutrient loading rate for aspen, jack pine and white spruce seedlings, all the exponential treatments'  $N_T$  for each species were assigned according to the nursery (Smoky Lake Forestry Nursery) fertilization rate. The compensation period was determined by using the result from Birge et al. (2006) for loading white oak and red oak as no information is available for aspen,

jack pine and white spruce. The growth period was established by using the nursery protocols.

#### **4. Summary**

As nutrient limitation is one of the major problems for early tree establishment for oil sands reclamation, it is important to develop methods to improve regeneration success. The development of new nursery cultural practices to improve seedling quality for outplanting is a possible option. N plays such an important part of seedling nutrition and growth; it would seem a logical place to start. Nutrient loading was found to be an alternative to current nursery fertilization practices, as fertilizing seedlings at rates of luxury consumption has been successfully accomplished for conifers, mesquite and other species to produce significant improvements in outplanting success (Imo and Timmer, 1992; Imo and Timmer, 2001; Dumroese et al., 2005; Thorpe and Timmer, 2005; Birge et al., 2006; Olier et al., 2009; Salifu et al., 2009). However, the optimal N level for the exponential fertilization method (or loading) has not been determined for species such as aspen, jack pine and white spruce that are commonly distributed in oil sands reclamation areas. Thus, the ability of exponential fertilization during nursery culture to maximize nutrient reserves and the result that this practice will have on outplanting success needs to be explored.



## **5. Objectives**

This study was conducted with the following objectives:

1) To determine target nursery fertilization rates and models for effective nutrient loading, so as to increase seedling nutrient storage at the time of harvest for aspen, jack pine and white spruce.

2) To understand relationships among nutrient supply, plant growth, tissue nutrient content, and concentration for aspen, jack pine and white spruce with the model in Figure 1.1. Such understanding will guide the nutrient loading research for future improvement.

3) Store nutrients in seedling plugs to benefit outplanting.

## **6. Thesis structure**

Two closely related experiments were conducted in two separate years on aspen, jack pine and white spruce seedlings to accomplish these objectives. The first experiment (Chapter 2) was conducted in 2009 to identify the optimal fertilization rates for three species using the exponential model as well as to test suitability of the model for these species. The experiment reported in Chapter 3 was conducted in 2010. The modified exponential model was used in this experiment to overcome some shortcomings of the 2009 experiment and a much wider range of exponential rates were used in order to determine the rate of which toxicity occurs in these three species. Quantification of seedling morphology,

seedling vector diagnosis, and nutrient analysis of seedling components were used to evaluate the response of both the seedlings and growth medium to the different exponential and conventional fertilization rates. The final chapter (Chapter 4) provides a summary of research findings from the two experiments and recommends areas for future research.

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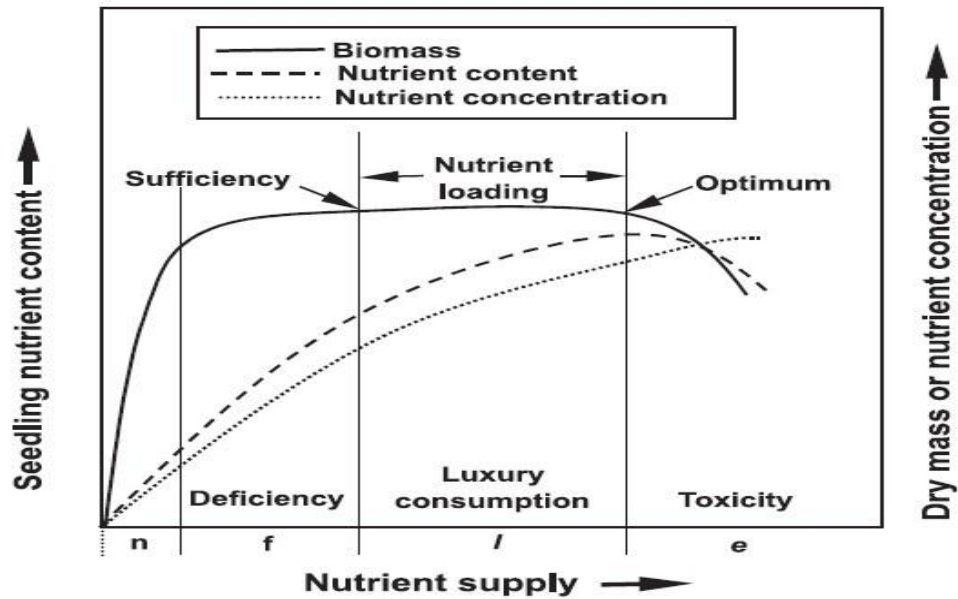
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**Table 1.1.** Available cavity size of styroblocks for containerized seedling production (Stuewe and Sons, 2011)

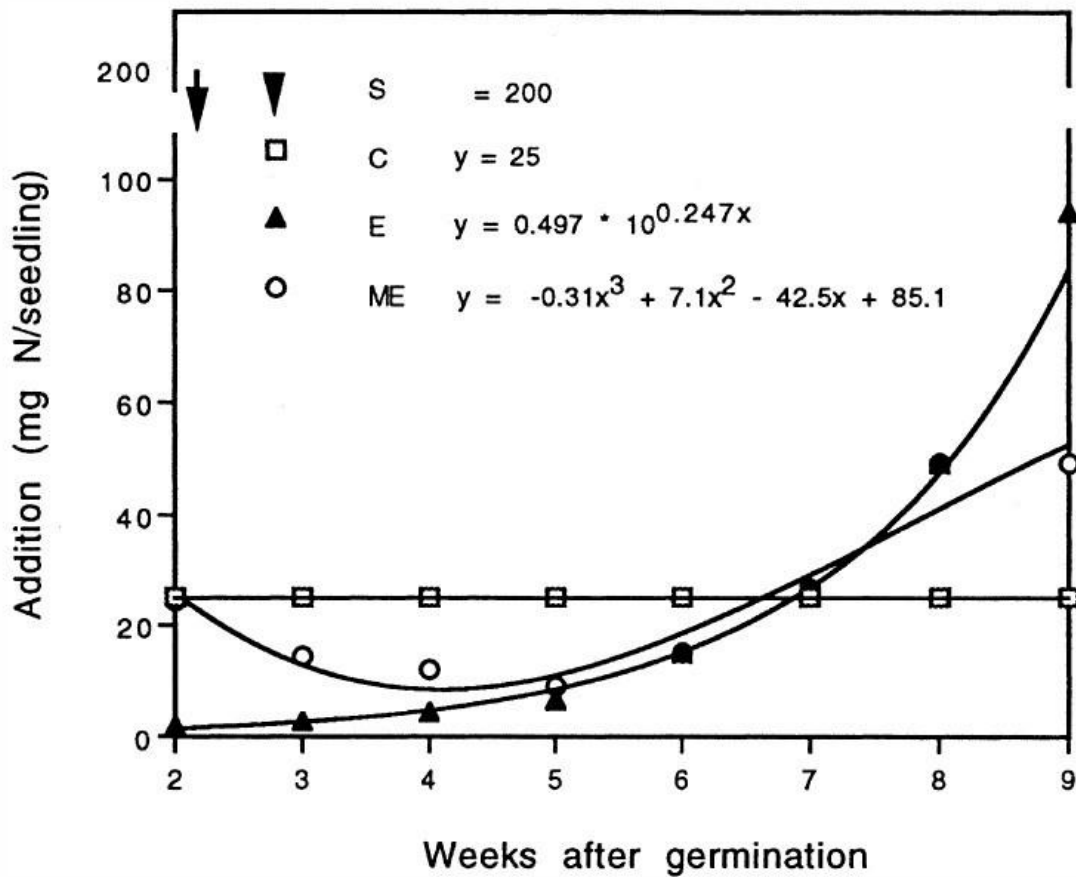
<b>ID code</b> <b>Cavities/ml</b>	<b>Metric</b> <b>number</b>	<b>Cavities per m<sup>2</sup></b>	<b>Volume per</b> <b>cavity (mL)</b>	<b>Cavity depth (cm)</b>
540/8	105A	2,557	8	5.1
448/17	207A	2,121	18	7.0
240/40	211A	1,135	40	11.4
240/50	213A	1,135	49	13.0
209/40	310A	996	40	10.4
198/40	310C	936	40	10.4
198/50	312A	936	48	12.1
198/60	313A	936	60	13.3
180/60	309A	852	60	9.5
180/70	313C	852	66	13.3
160/60	310B	756	54	10.4
160/65	313B	756	66	12.7
160/90	315B	756	90	15.2
160/120	323A	756	120	22.8
144/80	411B	681	80	10.9
144/95	313D	681	95	13.3
128/85	411B	553	84	10.9
112/80	410A	530	80	10.4
112/95	412B	530	95	11.7
112/105	415B	530	108	15.0
91/130	415C	430	130	15.2
77/80	410B	364	80	10.3
77/125	412A	364	125	11.7
77/170	415D	364	164	15.2
60/220	512A	284	220	12.0
60/250	515A	284	250	15.2
45/340	615A	213	336	15.2
45/450	620A	213	440	20.3
35/440	815C	166	440	15.1
28/500	623A	132	520	22.8
24/700	815A	113	700	15.2
20/535	815B	95	535	15.2
20/700	723A	95	710	22.8
15/1000	1015A	71	1,000	15.0
8/3000	1318A	38	3,200	17.9

**Table 1.2.** The effects of container cavity size on seedling quality

<b>Species</b>	<b>Optimal container size per cavity (mL)</b>	<b>Effects on seedling quality</b>	<b>Reference</b>
Indian sandalwood	270	Good quality seedlings >20 cm high, >3.0 mm in collar diameter	Annapurna et al., 2004
Mediterranean species	>200	Transplanting survival rate and growth all increased	Villar-Salvador et al., 2005
Italian stone pine	300-400	Taller seedlings with larger diameter and biomass	Domingues-Lerena et al., 2006
Acacia koa	656	Greater height and stem diameter growth	Dumroese et al., 2011



**Figure 1.1.** Relationships among nutrient supply, plant growth and tissue nutrient content and concentration. Conventionally, fertilizer (f) is added to supplement native supply (n) to maximize plant growth to its sufficiency level by preventing nutrient deficiency. Optimum nutrient loading is achieved by adding fertilizer (l) that induces luxury consumption to build up seedling nutrient reserves for outplanting. However, excessive fertilization (e) would inhibit growth due to the toxicity (Salifu and Timmer, 2003b).



**Figure 1.2.** Schedule of fertilizer additions by seedling age when applied as a single dose (s), constant top dressing (conventional practice) (c), pure exponential (e) and modified exponential (me) fertilization regimes during greenhouse culture. In each regime, a total of 200 mg N per seedling for mesquite seedlings was delivered as a complete nutrient solution starting 2 weeks after germination (Imo and Timmer, 1992)

## **Chapter 2. Nutrient Loading of Aspen, Jack Pine and White Spruce Seedlings Using an Exponential Model**

### **1. Introduction**

Oil sands mining has evolved over more than forty years in the mineable oil sands region of Alberta. Open pit mining leaves a reconstructed landscape of overburden dumps and tailings deposits that requires reclamation targeting self-sustaining and locally common ecosystems. Reclamation materials available for use as cover soil include LFH (a thin organic layer on the forest floor consisting of litter (L), fibric (F) and humic (H) material), peat-mineral mix, upland surface soils, and transitional soils (BGC Engineering Inc., 2010). A transitional soil is a soil with properties intermediate between two different soils and is genetically related to them. Cover soils are either stored in a stockpile for use at a later date or directly placed onto areas to be reclaimed. The biological properties of and nutrient capital in upland surface soils and transitional soils make them valuable reclamation materials for use as a cover soil (Strong and Laroi, 1983; Fisher and Binkley, 2000; Mackenzie and Naeth, 2010).

The majority of the landscape within the mineable oil sands region is comprised of organic soils. Because of the widespread availability of organic soils, cover soils comprised of peat or peat-mineral mix have been, and will continue to be, the dominant reclamation material used on post-disturbance

landscapes for most mines (Hemstock et al., 2010). Peat or peat-mineral mix provides plant nutrients and increases the soil water-holding capacity and thus alters the physical, chemical and biological properties of the reclaimed soil. Amendment of the extracted sand with peat or peat-mineral mix may also alter the pH of the reclaimed soil, which would ultimately affect nutrient availability (Danielson et al., 1983). Although peat or peat-mineral mix may contain high amounts of N, most of the N is not readily available. Furthermore, concentrations macro- and micronutrients are relatively low in the peat-based cover soil as compared with the upland surface soil. Thus, the ability of organic soils to provide nutrients for planted trees and other vegetation is often poor.

Low nutrient (especially N and P) availabilities can hinder the development of forest stands (Kaye and Hart, 1997) and lead to higher costs for land reclamation. To eliminate potential nutrient deficiencies that could limit the success of reclamation, fertilization is often used to supply the limiting nutrient to reclaimed sites. However, fertilization not only benefits the target crop species, but also benefits the non-target or competing species and most often that creates a greater stress (e.g., through increased competition) to the crop species (Timmer, 1997). Moreover, the competing species may benefit more than the planted tree species as newly planted tree seedlings have limited root development and limited ability to take up the applied nutrients. In addition, the applied nutrients may be more prone to be lost from the system through leaching and other processes.



One way to deal with the potential negative impact of fertilization is to use nutrient loading in the nursery phase to increase the nutrient reserve in seedlings, which would provide outplanted seedlings an advantage in gaining fast establishment and better survival under competing environments (Malik and Timmer, 1998; Salifu et al., 2009), as the extra nutrients stored in the nutrient loaded seedlings can be retranslocated to new as required moreover such nutrients are only available to the target trees and not competing species.

Nutrient loading, or the so called exponential fertilization, is a practice in the nursery that increases the plant's internal nutrient reserve by inducing luxury consumption of nutrients during early growth without altering seedling size (Imo and Timmer, 2002). Nutrient loaded seedlings can have many advantages over conventionally produced seedlings for outplanting in the field. First, nutrient loaded seedlings may have greater internal nutrient reserves to promote early establishment success of outplanted conifers and hardwoods (Salifu and Timmer, 2003; Birge et al., 2006). Second, nutrient loaded seedlings can improve their resistance to nutrient and moisture stress after outplanting by promoting root production (Vandendriessche, 1985; Timmer and Munson, 1991). Furthermore, nutrient loaded seedlings have better abilities to survive against competing vegetation (Malik and Timmer, 1996), again because of the greater internal nutrient reserve available. The successful outplanting reported in many studies (Timmer, 1997; Imo and Timmer, 2001; Dumroese et al., 2005; Salifu et al., 2009)

conducted in recent years with various species such as black spruce (*Picea mariana*), white pine (*Pinus monticola*) as well as northern red oak (*Quercus rubra*) and white oak (*Q. alba*) have also supported the utility of this technology in North America. Some nurseries such as the Smoky Lake Nursery do tissue tests throughout nursery production and maintain tissue nutrient concentration to a pre-set value through fertilization. The nutrient loading method is quite different from such conventional fertilization techniques. For a conventional fertilization practice, Imo and Timmer (1992) found that it would cause a surplus of nutrients at the early stage of the culture and a deficiency by the end of the growing season, which would cause growth dilution of nutrient concentrations (Imo and Timmer, 1992). Exponential fertilization better synchronizes nutrient supply with seedling growth or demand for nutrients during its growth period by inducing luxury consumption of nutrients (Timmer, 1997; Birge et al., 2006). On the other hand, conventional (or traditional) fertilization practices supply fertilizers in more or less equal doses at fixed intervals over the growing season.

Trembling aspen (*Populus tremuloides*), jack pine (*Pinus banksiana*) and white spruce (*Picea glauca*) are three of the major tree species native to and routinely planted for reclamation in the oil sands region. Even though some research (Imo and Timmer, 2002; Salifu and Timmer, 2003; Thorpe and Timmer, 2005) have been conducted in eastern Canada to understand the relationships among nutrient supply, plant growth, tissue nutrient content and concentration in

black spruce using a conceptual model (shown in Figure 1.1), very little research on those aspects has been conducted on aspen, jack pine and white spruce. If nutrient loaded seedlings of these species can be proven advantageous when outplanted on reclaimed sites in the oil sands region, there could be large gains in efficiency, survival rates, and initial growth rates.

This study was conducted with the following objectives:

- 1) To determine target nursery fertilization rates and models for effective nutrient loading, so as to increase seedling nutrient storage at the time of outplanting for aspen, jack pine and white spruce.
- 2) To understand relationships among nutrient supply, plant growth, tissue nutrient content, and concentration for aspen, jack pine and white spruce with the model in Figure 1.1. Such understanding will guide future nutrient loading research.

## **2. Materials and Methods**

### **2.1. Plant material and growing conditions**

In summer 2009, aspen, jack pine and white spruce seedlings were reared in styroblocks (615 A, 45 cavities per block, 340 mL per cavity) filled with a uniformly mixed peat moss and perlite (9:1, v/v) mixture at the Smoky Lake Forest Nursery (54° 05" N, 112° 14" W) in Alberta. Lime (500 g for each m<sup>3</sup> of peat moss) was added to the growth media for growing aspen seedlings in order

to adjust the pH level to between 5 and 6. The pH level for jack pine and white spruce was between 5 and 5.5. The growth periods for aspen, jack pine and white spruce were 12, 16 and 16 weeks, respectively.

Prior to seeding, styroblocks packed with the growth media described above were watered and allowed to drain freely for 24h and that process was repeated for 3 days to achieve full saturation. Five stratified seeds for each species were sown per cavity, germinating in about 3 weeks. Germinants were thinned during week 5 to leave one seedling per cavity. Seedlings were fertilized using a 20-20-20 commercial water-soluble fertilizer (20:20:20 plus micronutrients) from Plant Products Co. Ltd. (Brampton, ON) at this time once a week until the end of each species' growth period (see the growth periods for each species and year described above). The application rate was calculated based on N for the seven treatments as described in Timmer (1997) and Birge et al. (2006).

For each species, seven treatments (detailed treatment information is described in section 2.2) were randomly assigned to 28 trays, that were arranged in a completely randomized design with four replicates (seven treatments  $\times$  four replications = 28). The styroblocks were placed on raised benches in a heated and ventilated greenhouse in Smoky Lake Forest Nursery where the temperature was maintained between 18 and 30 °C. Conditions were maintained at 65-85% humidity and 20-h photoperiod. The extended photoperiod was maintained with natural light supplemented with sodium vapor lamps at a light intensity of 250

$\mu\text{mol m}^{-2} \text{s}^{-1}$ . Styroblocks were rotated periodically (once every two weeks) to minimize edge effects. After completing the whole fertilization process, blackout curtains were used for reducing greenhouse photoperiod from 20 h to short-day length (8h) to artificially induce bud set (Bigras and Daoust, 1992).

## **2.2. Fertilization Models**

In 2009, the treatments used were CK, C120, C180, E60, E120, E180, and E300 for aspen; CK, C200, C300, E100, E200, E300, and E500 for jack pine; and CK, C300, C450, E150, E300, E450, and E750 for white spruce. Among those treatments, CK represents the control treatment with addition of water only, C denotes conventional nursery practice and E denotes exponential fertilization practice, and the value represents the total amount of N (mg per plant) applied in the entire growth period. The reason for choosing these fertility treatments for each species is explained below with aspen as an example.

For aspen, C120 was the nursery standard fertilization rate at the Smoky Lake Forest Nursery, whereas C180 was selected to see if an increased fertilization rate would improve plant growth under the conventional fertilization regime. The exponential rates were selected to match and surpass the conventional rates. The greater fertilizer application rate in the exponential than in the conventional fertilization treatments was tested in an attempt to identify the rate that will cause toxicity as described in Figure 1.1. The treatments followed exponential functions

for each species to match nutrient supply with seedling growth (Timmer, 1997; Salifu and Timmer, 2003) using the following model:

$$N_T = N_S (e^{rt} - 1) \quad (1)$$

where  $r$  is the relative addition rate required to increase  $N_S$  (N content in seed or the initial N content) to a final N content ( $N_T + N_S$ ), and  $N_T$  is the desired amount to be added in a number ( $t$ ) of fertilizer applications, with  $t=12, 16, 16$  for aspen, jack pine and white spruce, respectively.

The quantity of fertilizer to be applied on a specific day ( $N_t$ ) can be computed using Eq. (2):

$$N_t = N_S (e^{rt} - 1) - N_{t-1} \quad (2)$$

where  $N_{t-1}$  is the cumulative amount of N added up to the previous application.

The detailed fertilizer delivery schedules in 2009 for the three species are listed in Figure 2.1.

### 2.3. Plant sampling and chemical analysis

From seedling emergence and first leaf flush (representing the baseline or time 0) to the end of the growth period, three samplings for each species were conducted in 2009 (the detailed sampling time was listed in Table. 2.1). Five seedlings per styroblock (20 per treatment) were harvested and placed in coolers (2 °C) for further processing at the University of Alberta. Samples were washed with deionized water following lab standard procedure and measured for stem

height in 2009. Then, samples were separated into stem, leaf and root. After the samples were oven-dried for 72 h at 70 °C, dry mass of different parts were weighed. Plant samples were subsequently ground and sent for chemical analysis. The combustion method was used to determine the total N concentration (both organic and inorganic compounds) using a CE440 Elemental Analyzer (Exeter Analytical Inc, North Chelmsford, MA, USA) (Kalra and Maynard, 1991).

#### **2.4. Statistical analysis**

Morphological and nutritional data were analyzed by repeated measures analysis of variance for each species based on the linear model as described in eq. 4 (Steel et al., 1996) using SAS (SAS, 2001):

$$Y = \mu + B + D + T + B(D + T + D*T) + e$$

(4)

where Y is the measured seedling response,  $\mu$  is the overall mean, B is the blocking effect that block out the environmental gradient and D is the fixed factor of the sampling date, T is the fixed effect of the fertility treatment and e is the error (random effect) associated with measured response from replicates.

Significant treatment means were compared according to the Waller-Duncan's multiple range test at  $\alpha=0.05$ . The types of covariance matrix used were CS (compound symmetry with 2 parameters) and AR(1) (autoregressive(1) with 2 parameters).

### 3. Results and Discussion

#### 3.1. Aspen

From Figure 2.2, by comparing data in week 12, it can be found that seedlings under exponential fertilization practices had less height growth than those under the conventional ones in the 12<sup>th</sup> week. However, all the seedlings had reached the minimum outplanting height of 25 cm for aspen seedlings according to nursery protocols (B. Woods (Smoky Lake Forest Nursery), personal communication).

Seedling dry weight doubled between week 4 and 8 and again between week 8 and 12 (Figure 2.3), demonstrating the quick increase of dry mass of components for aspen seedlings over their growth period. Even though no significant difference existed between the conventional and exponential treatments in terms of leaf dry mass, the root and stem biomass were much higher in the conventional treatment, suggesting that the low fertilization rate at the beginning of experiment in the exponential treatment limited root growth and subsequently stem growth for aspen seedlings. Thus, the modified model illustrated in Birge et al. (2006) with successful results in loading *Quercus rubra* and *Quercus alba* seedlings should be tested to resolve this problem. The modified exponential model was used to increase initial nutrient additions to facilitate nutrient exploitation by small root systems early in the season and to reduce application near the end of the season and avoid over-fertilization, which



could induce potential nutritional imbalances close to the end of nursery culture (Imo and Timmer, 1992).

In week 12, higher amounts of leaf nutrients (per seedling) were stored in E180 and E300 than in C120 and C180 (Figure 2.4). However, the same amount of total nutrients (per seedling) was stored in the seedlings between the exponential and conventional treatments due to the lower root and stem biomass in the former treatment.

According to the theoretical basis presented in Figure 1.1, it can be concluded that E180 was the optimum loading rate as the biomass production maximized out at that rate and factoring in the total amounts of fertilizer that could be realistically applied without increasing the cost of nursery seedling production (Figure 2.5). However, the loading rate that causes toxicity was unknown at this stage as the maximum rate used did not cause toxicity. Thus, the nutrient loading rates should be modified (extended) in 2010 to identify the excessive rate for aspen and to verify that E180 is the optimum loading rate. These results are in general in agreement with published information (Salifu and Timmer, 2003). However, the rates used for aspen are higher than those used for other species.

The general similarity of experimental data with trends illustrated in the conceptual model demonstrates model suitability for rationalizing and quantifying optimal fertilizer prescriptions for producing nutrient loaded tree seedlings for field planting. Nutrients stored in the seedlings through luxury uptake in aspen

seedlings would not be lost through leaf fall because of resorption (Aerts, 1996; Yuan et al., 2005). This important nutrient conservation mechanism by deciduous tree species can recover about 50-90% of the nutrients from senescing leaves, which are conserved in stem and root tissues for later utilization (Aerts, 1996; Yuan et al., 2005). Thus, outplanted aspen seedlings with higher internal nutrient reserves as conditioned by the nutrient loading technique may readily draw on these stored resources for growth.

Under the conventional fertilization regime, aspen seedlings typically have excessive height growth, and it is a general practice in the Smoky Lake Nursery to cut the top of the seedling to control height growth in the conventional fertilization regime. As a result of the lower height growth in the exponential fertilization regime, the aspen seedlings produced under that regime had lower biomass per seedling, but had similar amounts of nutrients stored per seedling, producing seedlings with higher nutrient storage per unit biomass.

### **3.2. Jack pine**

In week 18, pine seedlings under exponential fertilization had lower height growth than those under the conventional treatment (Figure 2.6). Furthermore, only seedlings in the conventional treatment had reached the required outplanting height (25-35 cm for pine seedlings) (Duryea and Landis, 1984).

Results showed that pine seedling dramatically increased biomass over the

growth period (Figure 2.7). The same situation to that of aspen seedlings occurred for pine seedlings, suggesting that the modified nutrient loading model and an extended growth period were also needed for the production of pine seedlings.

At week 18, leaf N and total N content in the whole seedling were all higher in the conventional than in the exponential treatments (Figs. 2.8), indicating that the nutrient loading model for pine seedlings used in this study was problematic.

No optimum rate can be recommended and no toxicity occurred as the total biomass, total N concentration and total N content all increased with increasing fertilizer application rate for the exponential treatment (Figure 2.9), suggesting again that the fertilization model needs to be revised.

These results suggested that jack pine was not successfully loaded using the pure exponential model. The main reason for this was that the growth medium had relatively low available N if no fertilizer was added, which hindered the early root growth of seedlings and then caused nutrient deficiency at the end of the growth period. Thus, a modified nutrient loading model should be used to determine the optimum loading rate. A modified exponential function is needed to increase initial nutrient additions in order to facilitate nutrient exploitation by small root systems early in the season and to reduce the rate of application near the end of the season to avoid over-fertilization, which could induce potential nutritional imbalances close to the end of the nursery production period (Imo and Timmer, 1992; Timmer, 1997; Birge et al., 2006).

### 3.3. White Spruce

At week 18, although the seedlings under the conventional fertilization practice were taller than observed for the exponential treatments, none of them reached the required outplanting height (25 cm) (Figure 2.10), indicating that the growth period (16 weeks) for spruce was much too short. Conventional nursery practice at Smoky Lake forestry nursery uses a 24-28week growing period for spruce (B. Woods (Manager of Smoky Lake Forest Nursery), personal communication). Thus, the growth period for spruce should be extended in a new experiment.

Because of the smaller size of the spruce seedlings produced, the component biomass of white spruce was less than that of pine and aspen seedlings (Figure 2.11). The modified nutrient loading model should also be used for this species as the very low total biomass by using pure nutrient loading model. This was another indication of seedling size not meeting the required outplanting size.

At week 18, the amount of N stored in the leaf (per seedling) in the seedlings was similar between exponential and conventional treatments (Figure 2.12a). However, greater amounts of N (per seedling) were stored in the seedling in the conventional treatment than in the exponential treatment (Figure 2.12b) due to greater root and stem biomass in the former than in the latter.

The E450 treatment could be regarded as the optimum rate for nutrient loading, but again I did not find the toxicity level in this experiment (Figure 2.13).

Meanwhile, seedling growth rates under exponential treatments were inferior to those in the conventional treatments. Thus, increasing fertilizer application rates with the modified nutrient loading model will need to be tested in order to verify that E450 is the optimum rate.

The same problem (all exponential treatments did not reached the required outplanting height and had lower component dry mass than conventional treatments) to that of jack pine occurred for spruce, suggesting that coniferous species cannot be successfully loaded by using the exponential model as the low nutrient availability in the growth medium can hinder the early root growth for this species. Therefore, a modified exponential model should be considered and tested for this species.

Total plant dry mass, as well as leaf and stem components, increase with fertilization when compared with controls (Figure 2.3 for aspen; Figure 2.7 for jack pine and Figure 2.11 for white spruce), which clearly demonstrate the benefits of supplemental nutrient application for promoting seedling growth. Similarly, dry mass production increased over time and differed ( $P < 0.05$ ) between treatments by the end of the growing season for all three species. This trend is also supported by the nutrient response data for each species.

#### **4. Conclusions**

In conclusion, with the exponential fertilization model, aspen had similar

amounts of nutrients stored per seedling as observed for the conventional model and had reduced height growth, producing seedlings with higher nutrient storage per unit biomass. For jack pine and white spruce seedlings, the nutrient loading method did not achieve the expected result in this experiment. The main reasons were as follows: 1) Growth periods were too short for the two species (16 weeks for both species compared with 22 and 26 weeks for jack pine and white spruce, respectively, with the normal nursery practice); 2) The lack of nutrient supply in the early growth period in nursery production might have limited the success of the nutrient loading process.

A modified nutrient loading regime and extended growth period needs to be studied to improve our understanding of nutrient supply, nutrient requirement, and seedling growth for aspen, jack pine and white spruce in the nursery in 2010. As the lowest exponential fertilization rate ( $60 \text{ mg N plant}^{-1} \text{ season}^{-1}$  for aspen,  $100 \text{ mg N plant}^{-1} \text{ season}^{-1}$  for jack pine and  $150 \text{ mg N plant}^{-1} \text{ season}^{-1}$ ) for three species resulted in nutrient deficiency for seedlings, the rates could be modified in order to fit the model in Figure 1.1 for these three species. As no significant differences exist between the two conventional rates in the three species and no toxicity levels were found, the higher conventional rate could be replaced by an exponential rate to extend the nutrient loading level to try to induce toxicity in the new study.

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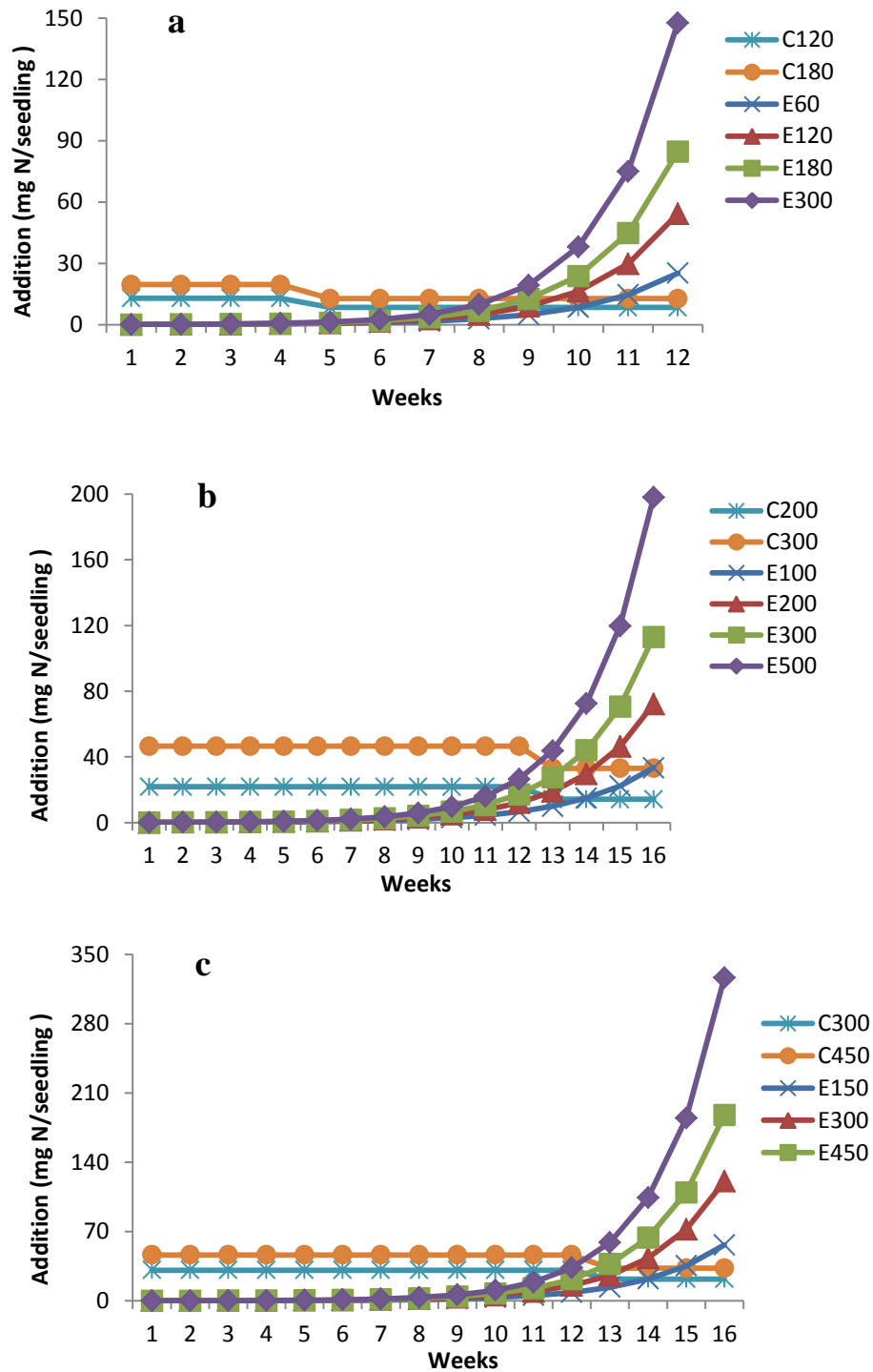
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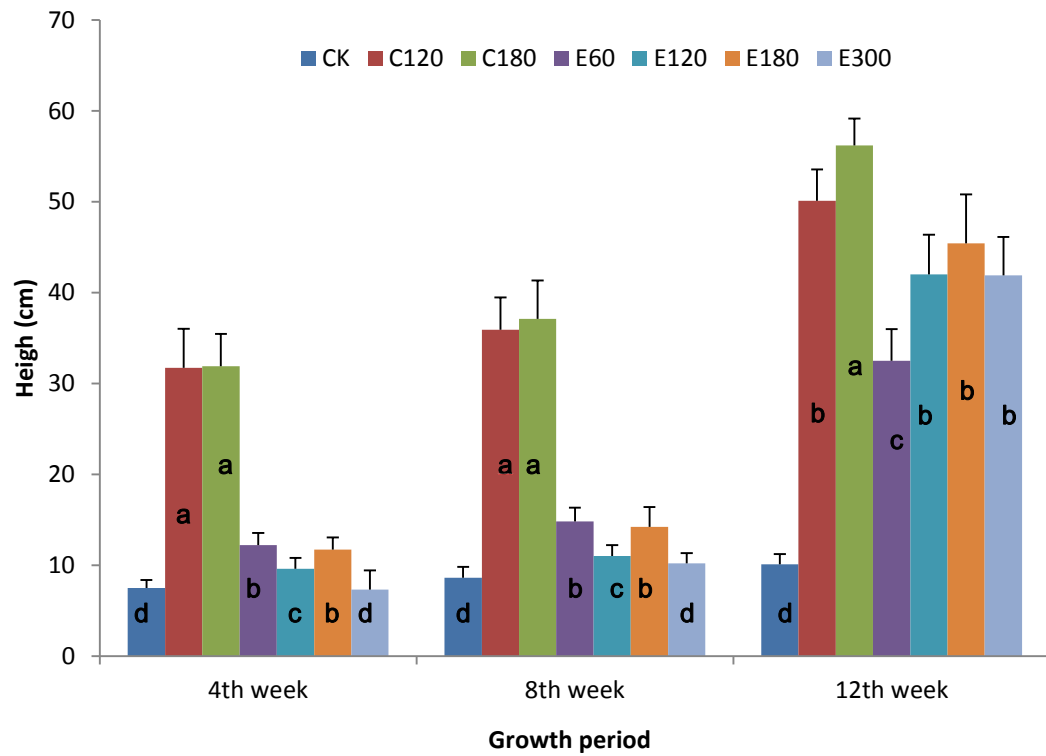
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**Table 2.1.** Sampling schedule during the experiment

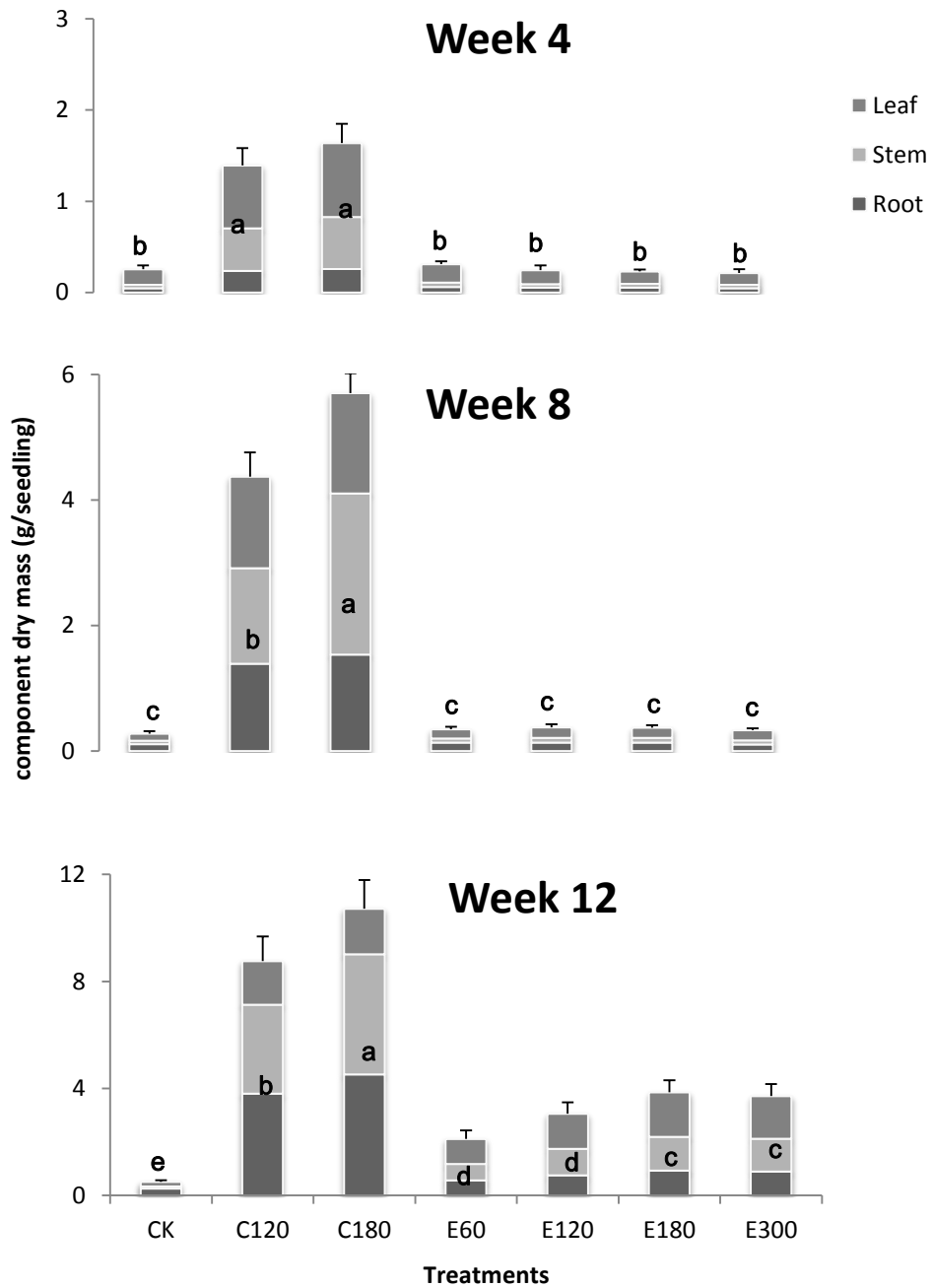
Species	Sampling time		
Aspen	week 4	week 8	week 12
Jack pine	week 5	week 10	week 18
White spruce	week 6	week 13	week 18



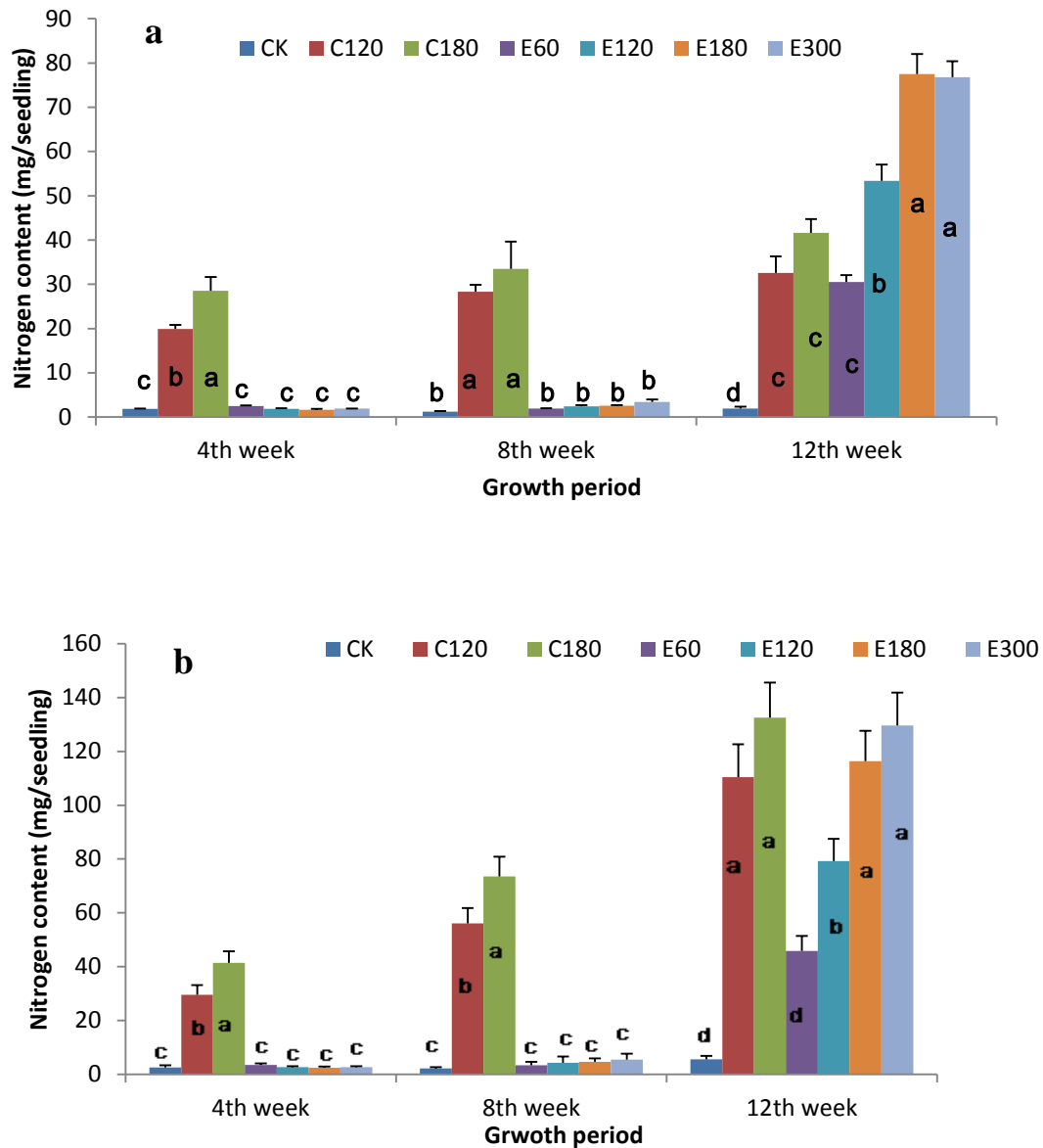
**Figure 2.1.** The detailed fertilizer schedule for three species. **a.** Aspen; **b.** Jack pine; and **c.** White spruce.



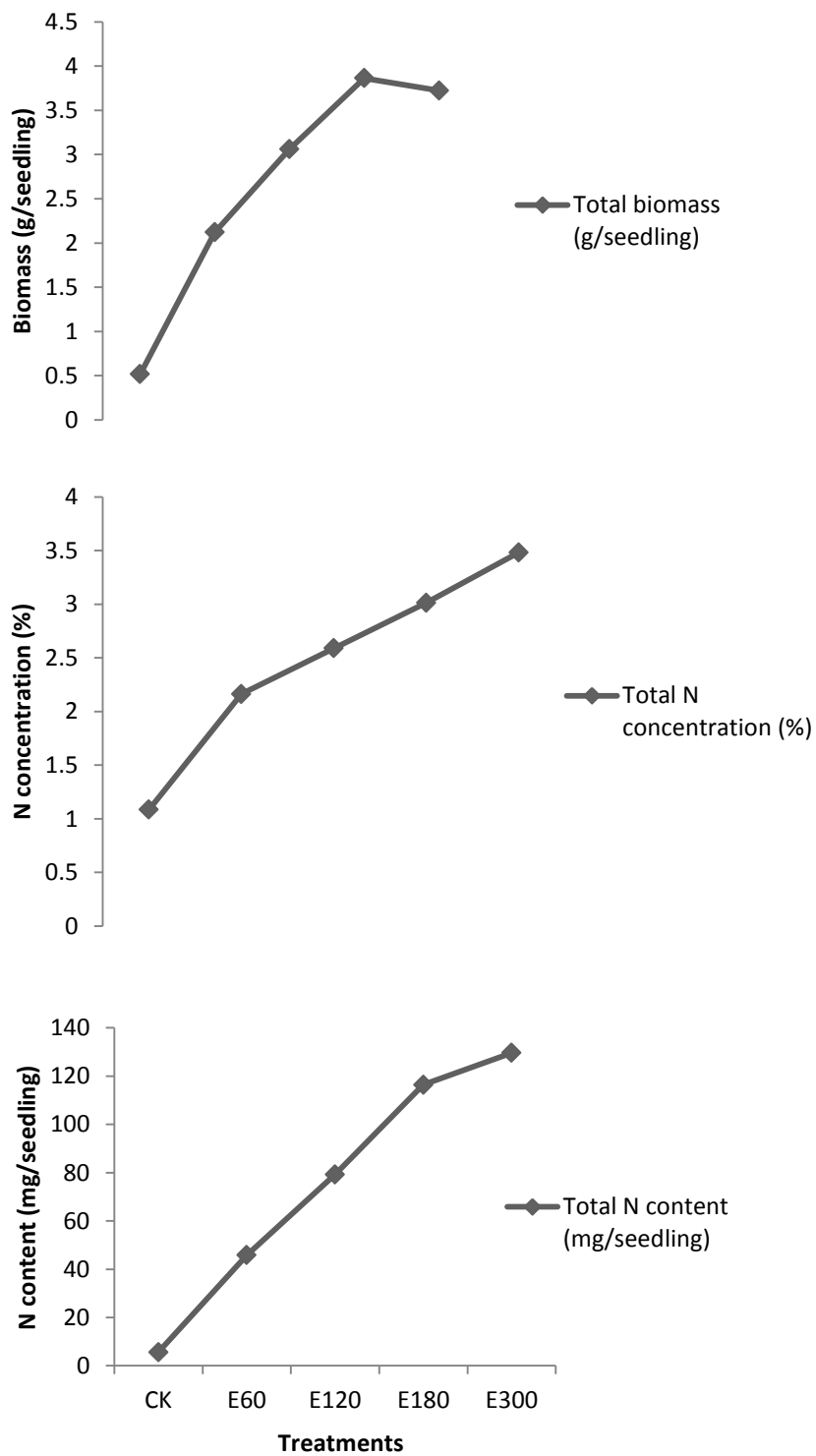
**Figure 2.2.** Aspen stem height under different treatments over the entire growth period in 2009. \* *Treatments followed by the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*



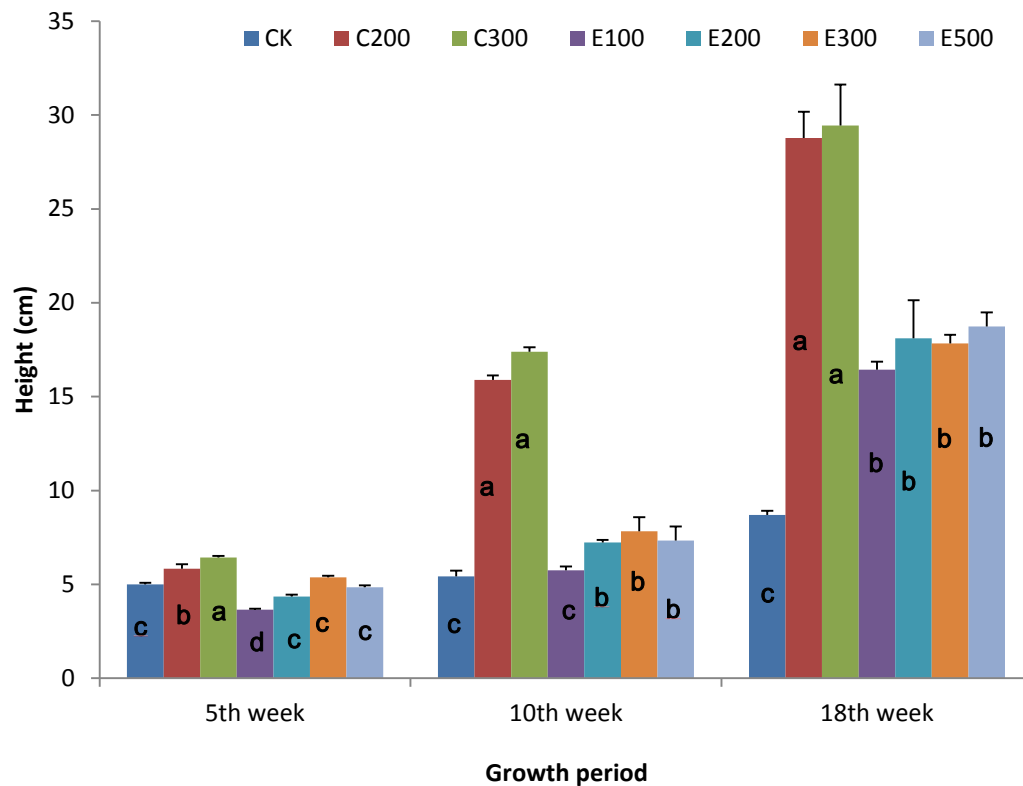
**Figure 2.3.** Aspen component dry mass under different treatments over the entire growth period in 2009.\* *Treatments followed by the same letter in the same sampling are not significantly different ( $p > 0.05$ ). Mean separation was based on the total biomass per seedling. Error bars are standard deviations of the means.*



**Figure 2.4.** Nitrogen content data of aspen under different treatments over the entire growth period in 2009. **a.** Aspen leaf nitrogen content under different treatments over the entire growth; and **b.** Aspen total nitrogen content under different treatments over the entire growth period. *\*Treatments followed by the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*

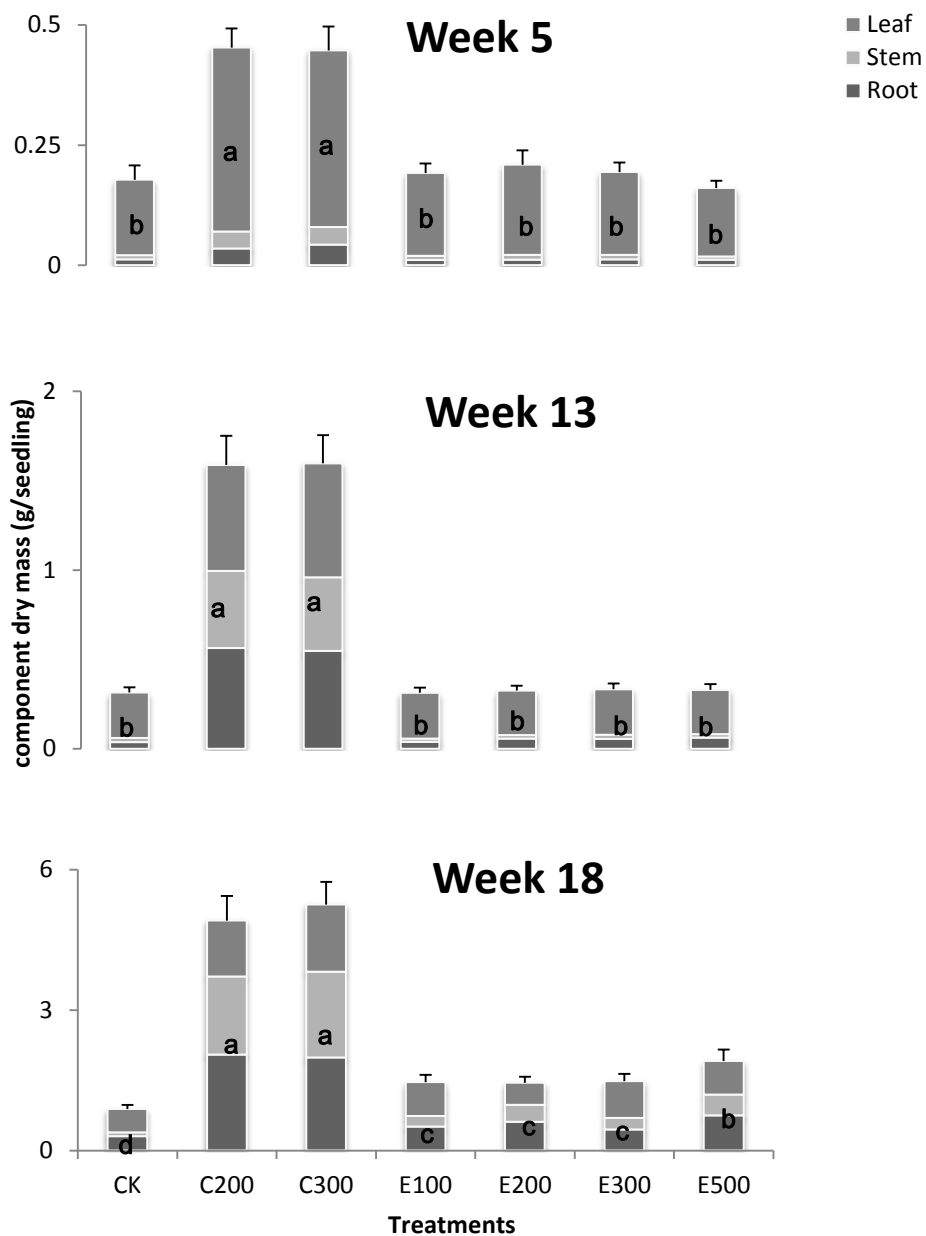


**Figure 2.5.** Relationships among nutrient supply, plant growth and tissue nutrient content and concentration for aspen at 12<sup>th</sup> week in 2009.

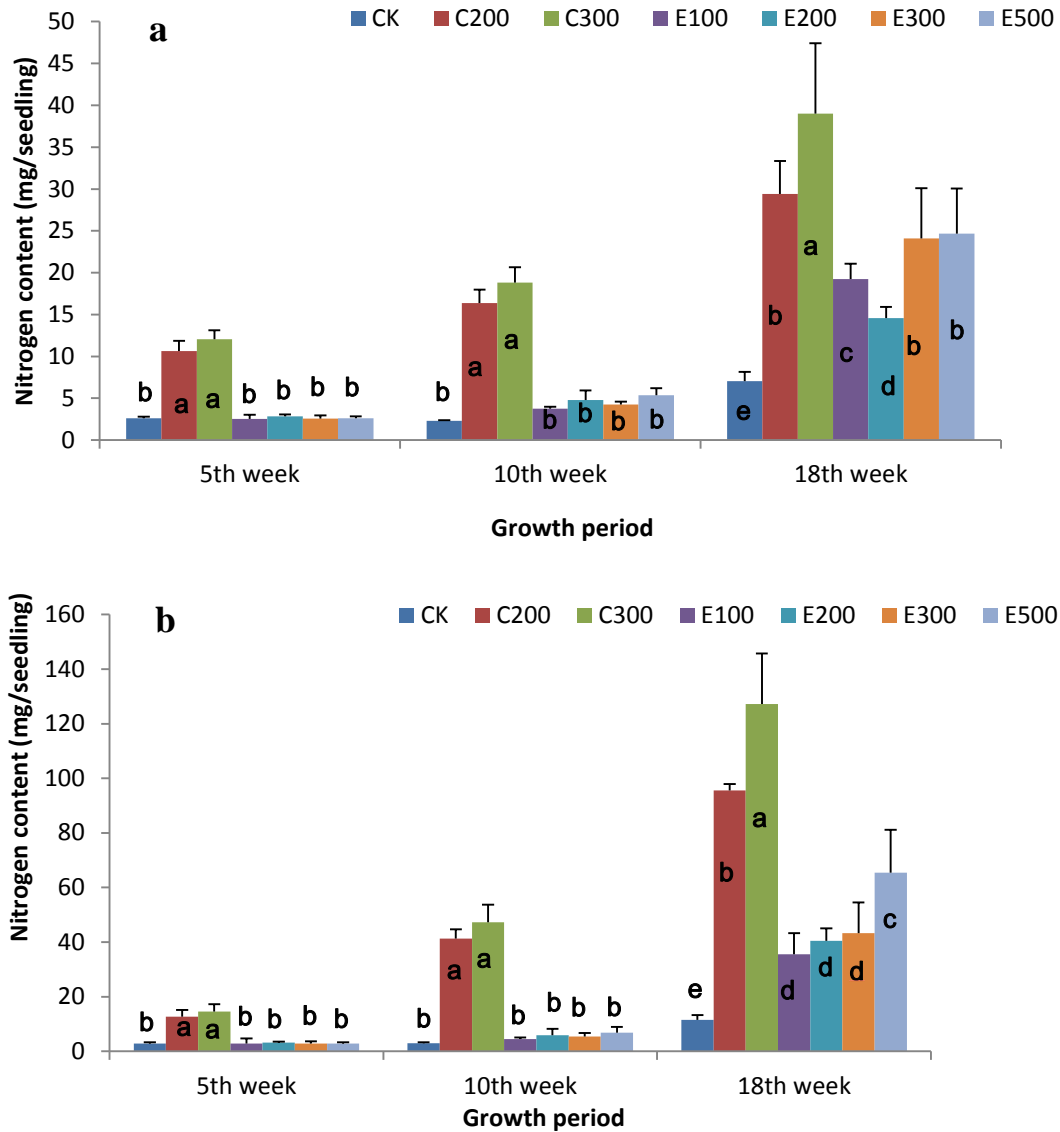


**Figure 2.6.** Jack pine stem height under different treatments over the entire growth period in 2009. *\*Treatments with the same letter at the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*

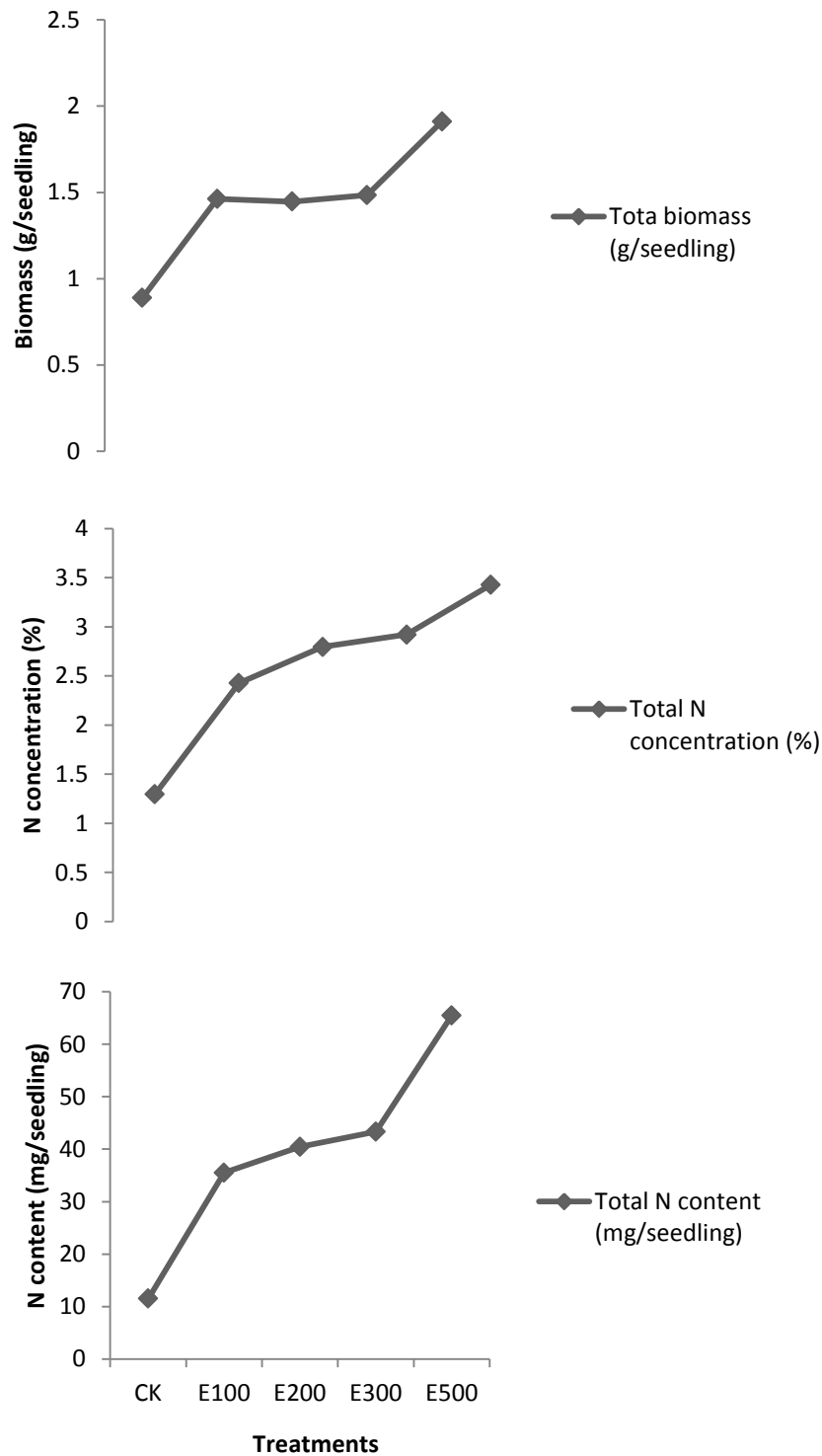




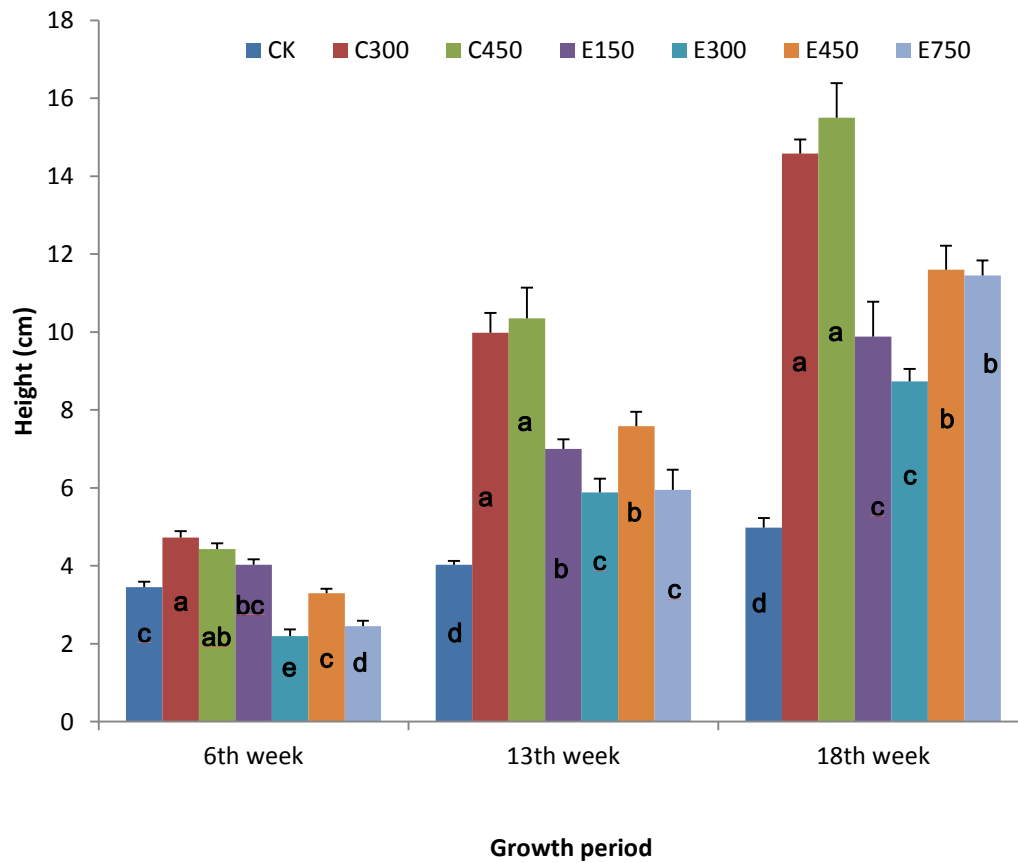
**Figure 2.7.** Jack pine component dry mass under different treatments at different sampling times in 2009. \*The total seedling biomass for treatments followed by the same letter in the same sampling are not significantly different ( $p>0.05$ ). Mean separation was based on the total biomass per seedling. Error bars are standard deviations of the means.



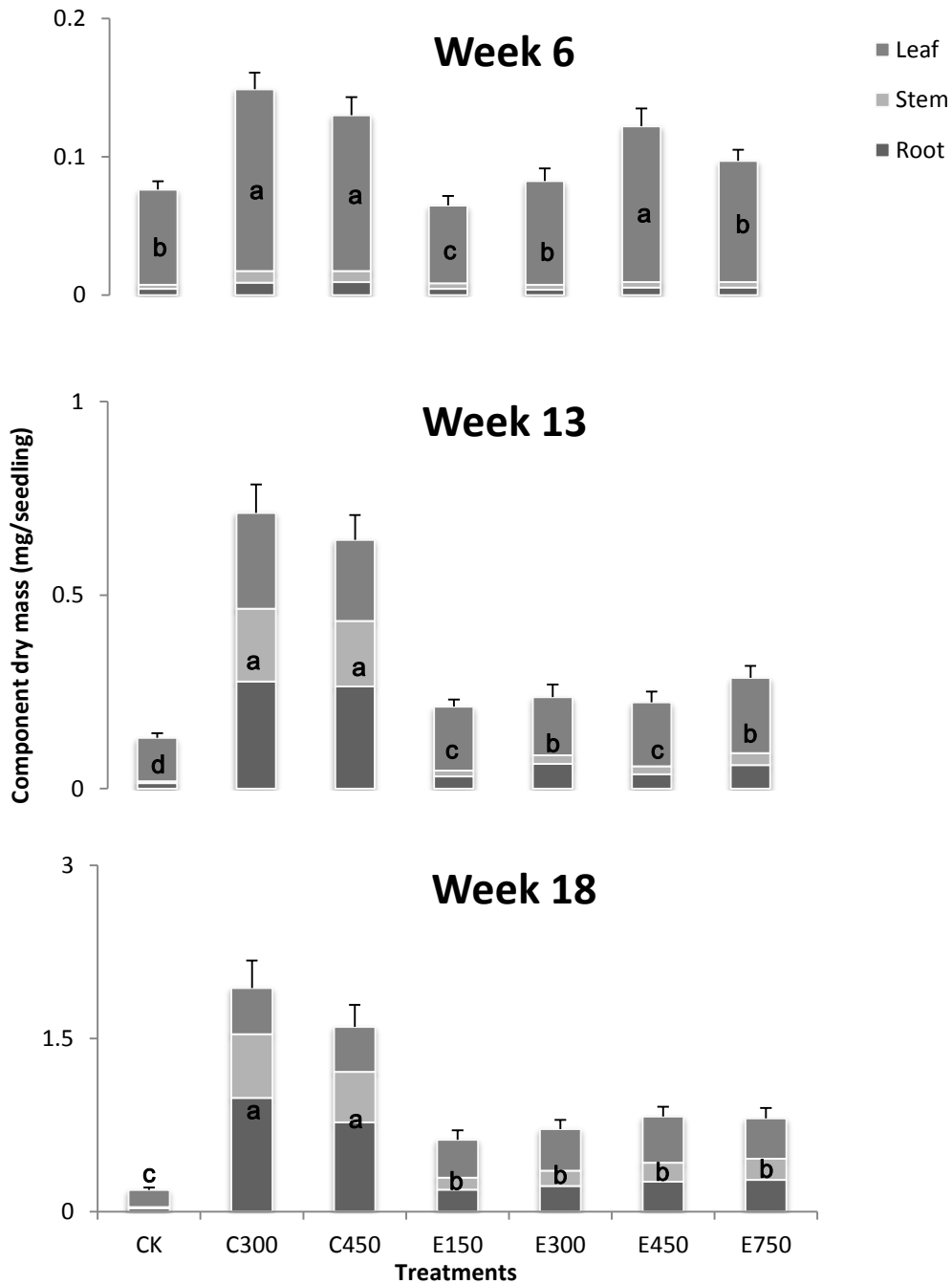
**Figure 2.8.** Nitrogen content data of jack pine under different treatments over the entire growth period in 2009. **a.** Jack pine leaf nitrogen content under different treatments over the entire growth; and **b.** Jack pine total nitrogen content under different treatments over the entire growth. *\*Treatments with the same letter at the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*



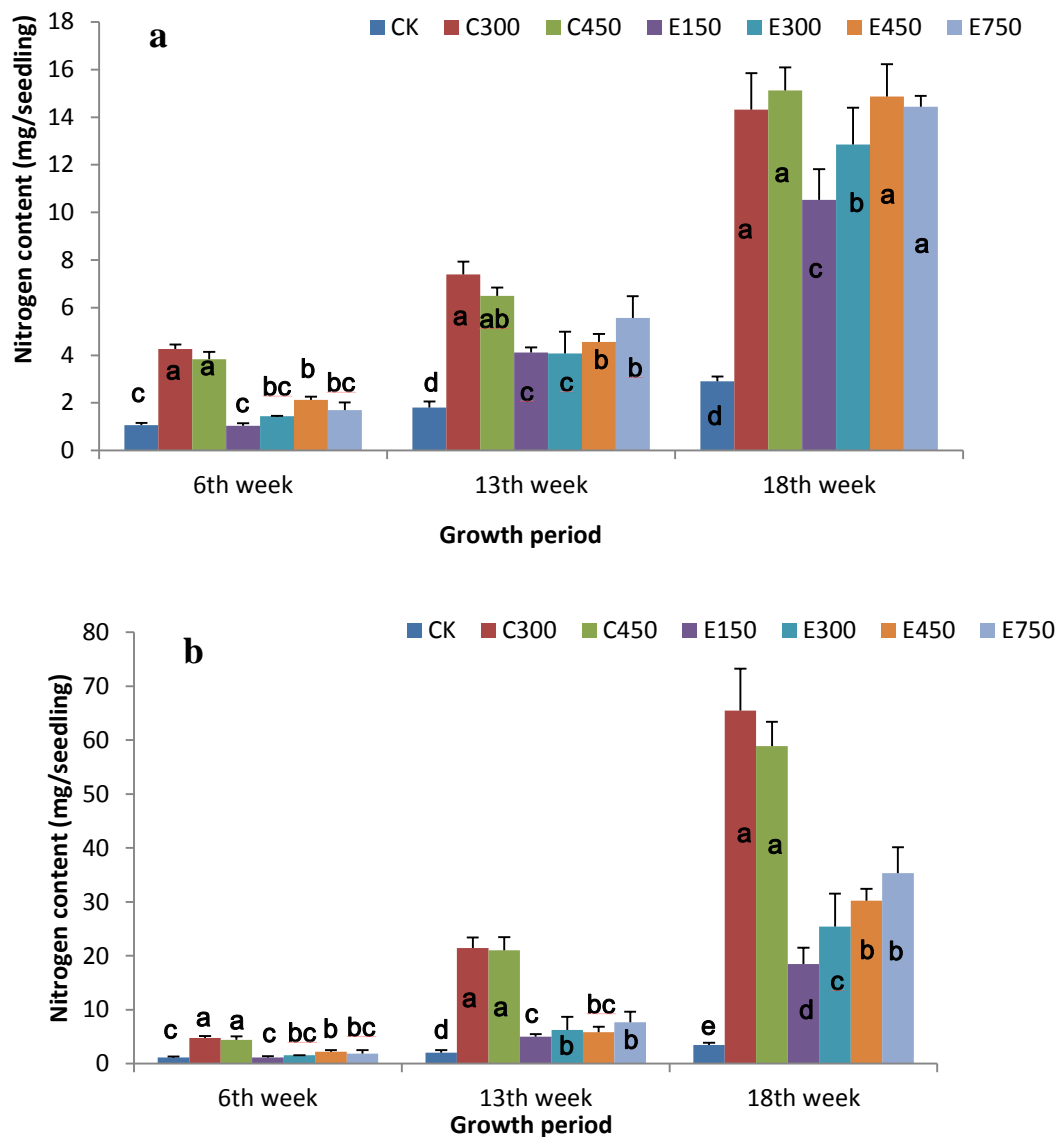
**Figure 2.9.** Relationships among nutrient supply, plant growth and tissue nutrient content and concentration for jack pine at 18<sup>th</sup> week in 2009.



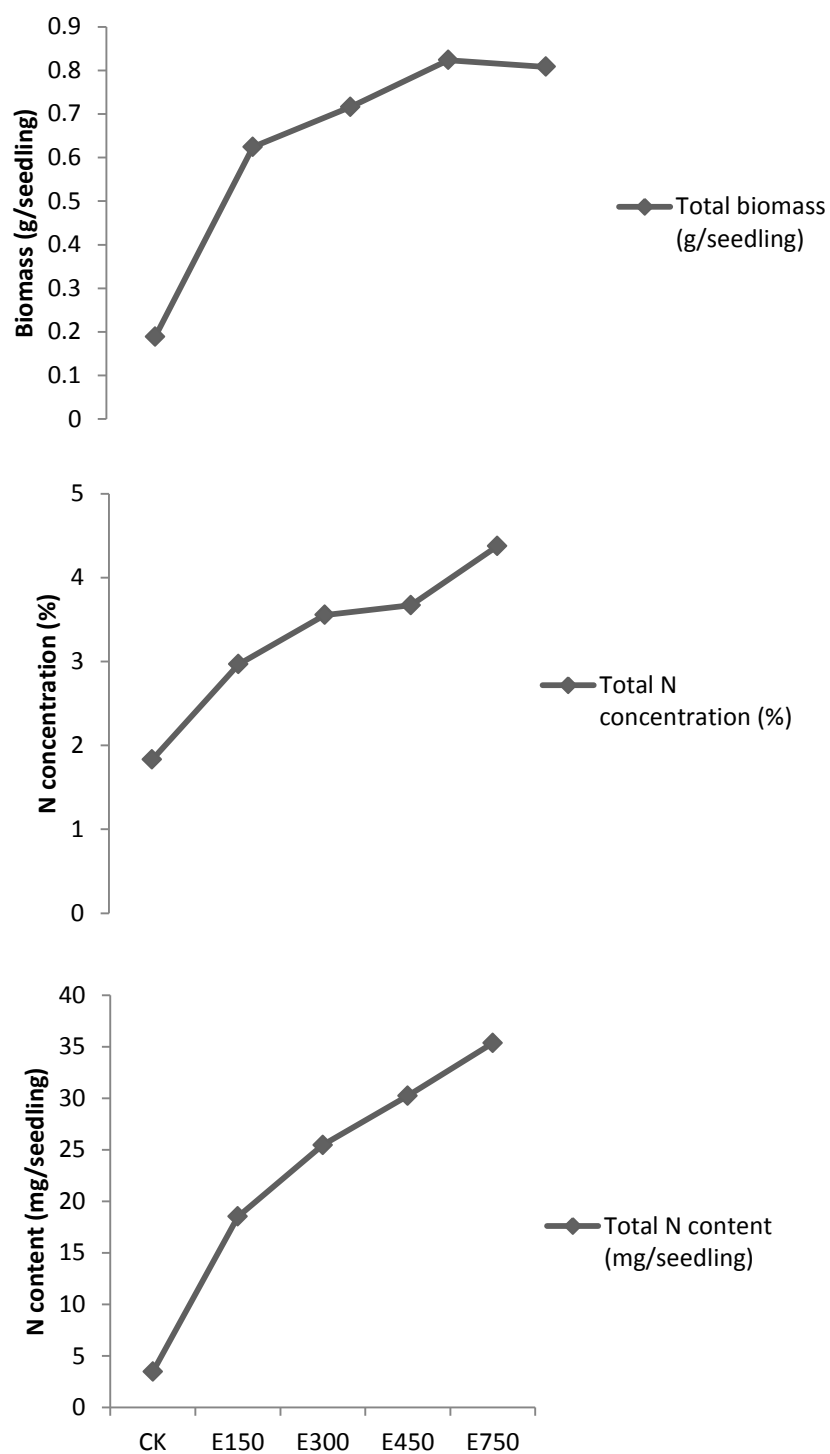
**Figure 2.10.** White spruce stem height under different treatments over the entire growth period in 2009. *\*Treatments with the same letter at the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*



**Figure 2.11.** White spruce component dry mass with different treatments after 18 weeks of growth in 2009.\* *Treatments followed by the same letter in the same sampling are not significantly different ( $p>0.05$ ). Mean separation was based on the total biomass per seedling. Error bars are standard deviations of the means.*



**Figure 2.12.** Nitrogen content data of white spruce under different treatments over the entire growth period in 2009. **a.** White spruce leaf nitrogen content under different treatments over the entire growth; and **b.** White spruce total nitrogen content under different treatments over the entire growth period. \*Treatments with the same letter at the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.



**Figure 2.13.** Relationships among nutrient supply, plant growth and tissue nutrient content and concentration for white spruce at 18<sup>th</sup> week in 2009.

## **Chapter 3. Nutrient Loading of Aspen, Jack Pine and White Spruce Seedlings Following a Modified Exponential Model**

### **1. Introduction**

Trembling aspen (*Populus tremuloides*), jack pine (*Pinus banksiana*) and white spruce (*Picea glauca*) are widely distributed commercial tree species that are native to the oil sands reclamation area in Alberta. Currently, LFH (a thin organic layer on the forest floor consisting of litter (L), fibric (F) and humic (H) material), peat-mineral mix, upland surface soils and transitional soils are the major reclamation materials available for use as cover soil (BGC Engineering Inc., 2010). As the majority of the landscape within the mineable oil sands region is comprised of organic soils, cover soils comprised of peat or peat-mineral mix have been, and will continue to be, the dominant reclamation material used on post-disturbance landscapes for most mines (Bardour et al., 2007). However, there is concern about adequate regeneration of young aspen, jack pine and white spruce trees because of the low nutrient availability in the peat-mineral mix. Although broadcast fertilization after transplanting is one way to increase seedlings' early outplanting performance, fertilizer applications in the field may strengthen competition from natural vegetation (Burdett et al., 1984; Munson and Timmer, 1981; Imo and Timmer, 1998). Using chemical herbicides is a way for competition control, however, strong public concerns are often raised against the



application of herbicides in forestry because of the potential harmful effects to wildlife and human health around the area (Wagner, 1993). Hence, a more efficient way to fertilize seedlings is to load them with nutrients in the greenhouse phase. This technique has been successfully tested in eastern Canada, where nutrient loaded black spruce seedlings exhibited better growth than conventionally fertilized seedlings, especially on weed-prone sites (Timmer, 1997).

Exponential fertilization in the nursery better synchronizes nutrient supply with crop demand, which maintains stable internal N concentration in plant tissues over time, referred to as steady-state nutrition (Ingestad, 1979; Imo and Timmer, 1992). Exponential fertilization was first examined using solution culture (Ingestad, 1979; Ingestad and Lund, 1986). This concept was further refined and validated for application with commercial soil culture (Timmer and Armstrong, 1987a; Timmer et al., 1991) and for bareroot production systems (McAlister and Timmer, 1998). A modified exponential function was adapted to increase initial nutrient addition to facilitate development of small root systems early in the season and to reduce application near the end of the season, thereby avoiding potential nutritional imbalances associated with over-fertilization (Imo and Timmer, 1992). Greater root:shoot ratio and improved stomatal control by nutrient-loaded plants can also contribute to better performance under drought conditions (Timmer and Miller, 1991; Imo and Timmer, 1992).

Originally, the exponential nutrient loading theory was applied to tree seedling nutrient studies in Sweden. Ingestad (1987) found that maximum seedling growth occurred when nutrients were applied at a high, exponentially increasing rate to match plant relative growth rate. Later, Timmer (1997) proposed a model for exponential nutrient loading (Figure 1.1), which suggests that plant growth and nutritional response to increased fertilization conforms to a curvilinear pattern describing phases of nutrient states in plants ranging from deficiency to toxicity, which help rationalize fertilizer prescriptions to improve nutrient diagnosis (Salifu et al., 2005). Additionally, this model can be used to quantify and determine target rates ( $n$ ,  $f$ ,  $l$  and  $e$  in Figure 1.1) for production of forest tree seedlings for field planting (Timmer, 1997).

In comparison to conventional fertilization, exponential nutrient loading may be superior as nutrients are supplied at an exponentially increasing rate exceeding seedling growth rate to minimize the danger of nutrient toxicity for young seedlings. In addition, exponential nutrient loading also increases nutrient addition rates to higher levels with growth time whereas the conventional one only applied constant fertilization rate (Timmer, 1997). Theoretically, nutrient loaded seedlings have higher internal nutrient contents when outplanted and will therefore have the potential for superior growth and an advantage over competing vegetation, relative to conventionally fertilized seedlings. This has been found in several earlier studies under field and controlled environmental conditions

(Timmer and Miller, 1991; Malik and Timmer, 1995; Malik and Timmer, 1996; Xu and Timmer, 1998).

Exponential fertilization has been extended to several evergreen forest tree species and resulted in specific fertilizer recommendations for given cultural regimes (McAlister and Timmer, 1998; VanderSchaaf and McNabb, 2004; Close et al., 2005). For example, it was recommended that a rate of 64 mg N plant<sup>-1</sup> season<sup>-1</sup> can be used to maximize growth of containerized black spruce seedlings (*Picea mariana* [Mill.] BSP) (Salifu and Timmer, 2003) and this is also used for commercial production of this species in Ontario, Canada. Although exponential nutrient loading has been examined for many different deciduous and coniferous species (Timmer, 1997; Qu et al., 2003; Close et al., 2005; Dumroese et al., 2005; Salifu et al., 2009), no published information is available on trembling aspen, jack pine and white spruce. There could be large gains in efficiency, survival rates and initial growth rates with effective nutrient loading practices for these species are outplanted. According to the study in 2009, the main problem was the lack of nutrient supply in the early growth period in nursery production might have limited the success of the nutrient loading process. Thus, the objectives of this study were 1) to determine target nursery fertilization rates and models for effective nutrient loading, so as to increase seedling nutrient storage at the time of harvest for aspen, jack pine and white spruce; 2) to understand relationships among nutrient supply, plant growth, tissue nutrient content, and concentration for

aspen, jack pine and white spruce with the model in Figure 1.1. Such understanding will guide the nutrient loading research for future improvement; 3) to address shortfalls in 2009 model and increase N storage in plugs for outplanting.

## **2. Materials and Methods**

### **2.1. Plant material and growing conditions**

Aspen, jack pine and white spruce seedlings were reared in styroblocks (615 A) (45 cavities per block, 340 mL per cavity) filled with a mixture of peat moss and perlite (9:1, v/v) at the Smoky Lake Forestry Nursery (54°05"N, 112°14"W) in Alberta. Lime (500 g for each m<sup>3</sup> of peat moss) was added to the growth media for aspen seedlings in order to adjust the pH level to between 5 and 6. The pH level for jack pine and white spruce was between 5 and 5.5. The growth periods for aspen, jack pine and white spruce were 14, 22 and 24 weeks, respectively, according to the nursery protocols.

Prior to seeding, styroblocks packed with the growth media described above were watered and allowed to drain freely for 24 h and that process repeated for 3 days to achieve full saturation. Five stratified seeds of each species were sown per cavity, germinating in about 3 weeks. Germinants were thinned during week 5 to leave one seedling per cavity. Seedlings were fertilized using a 20-20-20 commercial water-soluble fertilizer (20:20:20 plus micronutrients) from Plant

Products Co. Ltd. (Brampton, ON) once a week from this time forward until the end of each species' growth period (see the growth periods for each species and year described above). The application rate was calculated based on N as was described in Timmer (1997) and Birge et al. (2006).

Seven treatments (detailed treatment information is described in Section 2.2) were randomly assigned to 28 trays, arranged in a completely randomized design with four replicates (seven treatments  $\times$  four replications = 28). Styroblocks were placed on raised benches in a heated and ventilated greenhouse at the Northern Forestry Centre of the Canadian Forest Service where the temperature was maintained between 18 and 30 °C. Conditions were maintained at 65-85% humidity and 20-h photoperiod. The extended photoperiod was maintained with natural light supplemented with sodium vapor lamps at a light intensity of 250  $\mu\text{mol m}^{-2} \text{s}^{-1}$ . Styroblocks were rotated periodically (once every two weeks) to minimize edge effects. After completing the whole fertilization process, blackout curtains were used for reducing greenhouse photoperiod from 20 h to short-day length (8 h) artificially to induce bud set or hardening (Bigras and Daoust, 1992).

## **2.2. Fertilization model**

The fertility treatments were CK, C120, E120, E180, E300, E500, and E700 for aspen; CK, C200, E200, E300, E500, E700, and E900 for jack pine; and CK, C300, E300, E450, E750, E900, and E1050 for white spruce. Among those

treatments, CK represents the control treatment with addition of water only, C denotes conventional nursery practice and E denotes exponential fertilization, and the value represents the total amount of N (mg per plant) applied over the entire growth period. The reason for choosing these fertility treatments for each species is explained below with aspen as an example. For aspen, C120 was the standard fertilization rate at the Smoky Lake Forest Nursery, the exponential rates selected to match and surpass conventional rates. The greater fertilizer application rate in the exponential than in the conventional fertilization treatments was intended to find the rates that will 1) allow maximum nutrient storage without causing toxicity and 2) cause toxicity as described in Figure 1.1.

The modified nutrient loading model was used in which a compensation period was included. The compensation period overcomes two problems: 1) the growth medium itself has very low nutrient supply and 2) the seedlings have a poorly developed root system initially. Furthermore, Timmer et al. (1991), Imo and Timmer (1992) and Miller and Timmer (1994) all found that steady-state nutrient (explained below) culture in the seedlings could be ensured by using this modified exponential regime. At steady-state nutrient status, seedlings have stable internal nutrient concentrations over the growth period. In contrast, conventional methods usually result in decreased tissue nutrient concentrations in seedlings over the growth period (Timmer, 1997).

For this model, an amount of N compensation ( $N_c$ ) was initially subtracted

from the last two applications calculated from Eq. (2). Then  $N_c$  was delivered exponentially to correspond with exponential expansion of the root system based on Eq. (3):

$$N_c = N_0 (e^{-rt} - 1) \quad (3)$$

Where  $r$  was the relative addition rate required to increase  $N_0$  to  $N_c$ ;  $t$  was the compensation period,  $t=4, 8$  and  $8$  weeks for aspen, pine and spruce, respectively;  $N_0$  was the final amount of nutrient added during the compensation period (Imo and Timmer, 1992; Jacobs and Timmer, 2005).

The detailed fertilizer delivery schedules for the three species are shown in Figure 3.1.

### **2.3. Plant sampling and chemical analysis**

From seedling emergence and first leaf flush (representing the baseline or time 0) to the end of the growth period, three samplings for each species were conducted (the detailed sampling times are listed in Table 3.1). Another sampling at harvest was done to study nutrient translocation after leaf fall (for aspen) or after the on-set of dormancy (for spruce and pine). Five seedlings per styroblock (20 per treatment) were harvested and placed in coolers ( $2^\circ\text{C}$ ) for further processing at the University of Alberta. Samples were washed by using deionized water following lab procedure to clean the dirt and measured for stem height and root-collar diameter (RCD), then separated into stem, leaf and root. After the

samples were oven-dried for 72 h at 70 °C, dry mass of different parts were weighed. Plant samples were subsequently ground using a ball grinder and sent for chemical analysis. The combustion method was used to determine total N concentration (both organic and inorganic compounds) using a CE440 Elemental Analyzer (Exeter Analytical Inc, North Chelmsford, MA, USA). Plant samples were also digested in nitric acid plus perchloric acid (Kalra and Maynard, 1991) and concentrations of other elements (P and K) determined using inductively coupled plasma mass spectrometry (ICP-MS) (Perkin Elmer's Elan 6000, PerkinElmer Inc., Waltham, MA, USA).  $\delta^{15}\text{N}$  was determined with a continuous-flow stable isotope ratio mass spectrometer (IsoPrime-EA, Micromass, UK). N isotope composition ( $\delta$ ) was calculated as

$$\delta(\text{‰}) = [(R_{\text{sample}}/R_{\text{standard}}) - 1] * 1000$$

where R is the ratio of  $^{15}\text{N}/^{14}\text{N}$ , and the standard was Pee Dee Belemnite (PDB) standard for atmospheric  $\text{N}_2$ .

## **2.4. Vector diagnosis**

Vector diagnosis was also employed in this project to simultaneously compare plant dry mass and nutrient status of plants or plant components. This approach can provide comprehensive and accurate diagnostic information and corroborate the occurrence of growth dilution, deficiency, luxury uptake, toxicity and nutrient interactions (Timmer and Armstrong, 1987b; Haase and Rose, 1995).



Treatment values were normalized to a reference treatment set at 100 to facilitate relative comparisons. The diagonals or slanting lines (stemming from the origin) represent lines of equal biomass. Thus, points on the same diagonal represent changes in nutrient concentration and content without change in biomass. Shifts to the left and right of the reference treatment represent net loss and gain (respectively) of nutrients, or decreases and increases (respectively) in dry mass. Change in nutrient availability (usually enrichment) affecting the three parameters are integrated into a vector defined by its orientation and magnitude. Vector length represents the degree of change or response magnitude. Vector direction (shifts A to F) is used to identify diagnostic interpretations and possible nutrient disorders (see box beneath Figure 3.2) relating to the change in nutrient status (Haase and Rose, 1995; Salifu and Timmer, 2003). Increased plant dry mass ( $z$ ) and nutrient uptake ( $x$ ) but declining nutrient concentration ( $y$ ) would cause growth dilution (shift A) which indicates growth rate was higher than nutrient uptake rate (Figure 3.2). Nutrient deficiency was defined as increases in all parameters that implied improved growth and uptake of the nutrient involved. Luxury consumption was signified by increases in  $x$  and  $y$  but no change in  $z$  (shift D) which indicated that growth rate is lower than nutrient uptake. Nutrient toxicity was characterized by an increase in  $y$  but decreases in  $z$  and  $x$  (shift E), reflecting toxic accumulation. The other nutritional response (F) is not applied in this study.

## 2.5. Statistical analysis

Morphological and nutritional data were analyzed by repeated measures analysis of variance for each species based on the linear model of Steel et al. (1996) (Eq. 4) using SAS (SAS, 2001):

$$Y = \mu + B + D + T + B(D + T + D*T) + e \quad (4)$$

where Y is the measured seedling response,  $\mu$  is the overall mean, B is the blocking effect that blocks out the environmental gradient and D is the fixed factor of sampling date, T is the fixed effect of the fertility treatment and e is the error (random effect) associated with measured response from replicates.

Significant treatment means were compared according to the Waller-Duncan's multiple range test at  $\alpha=0.05$ . The types of covariance matrix used were CS and AR(1) (CS is compound symmetry with 2 parameters and AR(1) is autoregressive(1) with 2 parameters).

## 3. Results and discussion

### 3.1. Aspen

#### 3.1.1. Seedling growth and nutrition

Considering the minimum outplanting height (25 cm) and RCD (2.5 mm) requirements from the nursery protocols, E240 has the lowest H:D ratio among all the exponential treatments that produced seedlings meeting the minimum size

requirement (Figure 3.3).

Compared with the unfertilized treatment (control) at the last collection, fertilization significantly increased plant dry mass (Figure 3.4). This signified nutrient deficiency in the control and the importance of nutrient supply to seedling growth (Imo and Timmer, 1992). In addition, there was greater proportional allocation of dry mass to roots than to stem within each treatment. The N concentration in aspen seedlings in the last collection was shown in Figure 3.5. Aspen N content of different parts over the growth period is summarized in Figure 3.6 to help understand the nutrient status in the seedlings. Trends in plant nutrient concentration (Figure 3.5) were similar to those shown in Figure 1.1, increasing gradually with N supply in the deficiency range due to growth dilution and rapidly at higher addition rates due to accumulation effects. Plant P and K content (Figures. 3.6e and 3.6f) increased with substrate fertility by 582-818% for P and by 600-899% for K up to E240, and then declined thereafter, likely due to the onset of toxicity at E300. The P content decreased with increased fertilizer application rate after E300 (Figure 3.6e) but K content (Figure 3.6f) increased again at E500 which had the same trend discussed above. The decreased P may indicate P limitation is another factor limiting seedling growth at higher N fertility (Birge et al., 2006). Severe competition for a limiting nutrient may cause this condition. As N, P and K dynamics are closely related, more N uptake at the higher fertility treatment levels can be stimulated by increasing P and K

availability (Nambiar and Fife, 1991; Malik and Timmer, 1996). The importance of using balanced fertilizers containing N and P can also be demonstrated by this study. However, the observed P limitations in this study can be corrected by increasing P supplementation (Boivin et al., 2004).

When the experimental data in Figs. 3.4, 3.5 and 3.6 were summarized into Figure 3.9 to describe plant growth and nutritional (N, P and K) responses for aspen seedlings in week 12 under different fertilization rates, the relationships conform partially to the conceptual model (Figure 1.1). For instance, plant growth (Figures 3.3, 3.4 and 3.7) significantly increased with N supply at the deficiency range, remained relatively stable during the luxury consumption of N, but eventually declined at higher N application rates (E300) because of toxicity. However, the highest N loading rates (E500) still caused increases in plant growth. By examining data from Figs. 3.3 and 3.7, the increased fertilization (increased about 108% in fertilization usages by comparing with E240) of E500 mostly contributed to height growth and only increased N content by 34% comparing with E240. Moreover, the high cost of transporting the very tall aspen seedlings in the nursery is always a big concern nowadays which indicated the appropriate size of seedlings are very crucial. Thus, E240 was better than E500 with respect to fertilizer use efficiency and economic aspect.

It has been found that greater plant growth in nutrient loaded seedlings would enable greater absorption of aboveground resources (such as light and CO<sub>2</sub>) to

increase photosynthesis and promote early establishment (Grossnickle and Folk, 1993). In addition, the higher RCD also indicates that the seedlings will have better strength, root system size and associated protection from drought and heat damage (Bayley and Kietzka, 1997). Besides stem height and RCD, height:diameter ratio is another important stock quality indicator that will always be considered by nursery producers. It has been suggested that seedlings with a low ratio would be more resistant to flattening by snow or dead herbaceous vegetation and promote vigorous early growth (Burdett, 1983). Apparently, acute toxicity induced stunting in seedlings raised at the E300 regime (Figure 3.9). The consistent pattern in Figure 3.9 with trends in the conceptual model (Figure 1.1) confirms suitability of the modified exponential model for aspen seedling production in the nursery. Greater nutrient allocation in roots for all the nutrient loading treatments between the last sampling and after hardening (Figure 3.6c) suggested that roots act as primary sinks for nutrient storage that could serve as a critical nutrient source to support new growth (Salifu et al., 2009).

### **3.1.2. Identifying the optimum nutrient loading rate**

Assuming that N accumulated in the control seedlings reflected N availability from the peat substrate, the  $n$  of N supply in the conceptual model (Figure 1.1) was small (about  $2.63 \text{ mg N plant}^{-1} \text{ season}^{-1}$ , Figure 3.7). Supplemental fertilizer (f) countered this deficiency and increased growth to the sufficiency level at E180

(Figs. 3.9a and 3.9b). The response was associated with a 36, 41, and 7% increase in dry mass, N content and concentration, respectively (Figure 3.7). The sufficiency level found for aspen was greater than the C120 rate commonly used for the production of containerized aspen seedlings.

The loading rate ( $I$ ) that induced luxury nutrient uptake occurred between E180 and E240, in which seedling N content increased without changing dry mass or seedling size (Figure 3.7) compared with the sufficiency level (i.e., E180, as previously discussed). Compared with the C120 treatment, the E240 treatment increased N, P and K uptake by 117, 65 and 53%, respectively. This target threshold is higher than the  $100 \text{ mg N plant}^{-1} \text{ season}^{-1}$  loading estimated for oak seedlings (Salifu and Jacobs, 2006). Due to the process of resorption, most of nutrients taken up through induced luxury uptake in aspen seedlings would not be lost through leaf fall. It has been found that 50-90% of nutrients from senescing leaves can be remobilized and stored as reserves in stem and root tissues and available for new growth after outplanting (Tagliavini et al., 1998). Therefore, increased internal nutrient reserves resulting from the nutrient loading method may be used for facilitating new growth at outplanting (Tagliavini et al., 1998). N supply ( $e$ ) exceeding optimum levels may result in N toxicity that inhibits plant growth (Timmer, 1991). Excessive fertilization ( $e$ ) could be found at E300 which decreased dry mass and nutrient content as compared with the E240 treatment (Figure 3.7). This result can also be shown by using vector diagnosis (shift E)

(Figures 3.2 and 3.9d). The results exemplify the need to determine optimum fertilizer rates before nutrient loading. This will help avert over-fertilization and toxicity effect.

## **3.2. Jack pine**

### **3.2.1. Seedling growth and nutrition**

Generally, stem height and RCD (Figure 3.11) of jack pine conform well to the model trend (Figure 1.1). Moreover, Figures. 3.11 also suggested that luxury uptake did not significantly stimulate growth. The recommended height for pine seedlings for outplanting was between 18 and 25 cm or less than 30 cm tall and foresters are advised not to purchase or plant pine seedlings taller than this (Bayley and Kietzka, 1997). The E500 treatment produced seedlings that met the outplanting height and RCD (2.5 mm) requirements and had the lowest height:diameter ratio among all the exponential treatments (Figure 3.11c).

Trends in plant nutrient concentration (Figure 3.13) were similar to those shown in Figure 1.1, increasing gradually with N supply at the deficiency range and rapidly in toxic addition rates due to accumulation effects (Haynes, 1986; Timmer, 1997). Plant P and K uptake (Figures 3.14e and 3.14f) increased with substrate fertility by 384-632% for P and 489-590% for K up to the E500 treatment, and then declined thereafter, likely due to toxicity. The P and K content decreased with increasing fertilizer application rate after E500.

In order to examine plant growth and nutritional responses of pine seedlings under different fertilization rates, experimental data in Figures 3.12, 3.13 and 3.14 were summarized into Figure 3.15; the relationships conformed well to the conceptual model (Figure 1.1). Seedling growth increased with increasing fertilization rate in the deficiency range, remained relatively stable during luxury uptake, but declined at very high N rates ( $700 \text{ mg N plant}^{-1} \text{ season}^{-1}$ ) associated with toxicity (Figure 3.15). Toxicity induced stunted growth in seedlings produced in the E900 treatment. The consistent patterns in Figure 3.15 with trends in the conceptual mode (Figure 1.1) confirm the suitability of the modified exponential model as a useful framework for quantifying and characterizing fertility targets for jack pine seedling culture as previously validated for black spruce (Salifu and Timmer, 2003).

### **3.2.2. Identifying the optimum nutrient loading rate**

Assuming that N accumulated in non-fertilized trees reflected availability from the growing substrate, the  $n$  supply of N in the conceptual model (Figure 1.1) was calculated as total N in the control minus  $N_s$ , which equaled  $8.12 \text{ mg N plant}^{-1} \text{ season}^{-1}$ . This index is within the range of  $1\text{-}8 \text{ mg N plant}^{-1} \text{ season}^{-1}$  estimating for black spruce (Salifu and Timmer, 2003). Supplemental fertilizer ( $f$ ) countered deficiency and increased growth to the sufficiency level at E200 (Figure 3.17a). Growth dilution occurred with the E300 treatment (Figure 3.17b).



The deficiency response was characterized by 551, 973, 478 and 411% increases in dry mass, and N, P and K contents, respectively, as compared with the control treatment (Figure 3.15). The sufficiency level found here for pine was equal to the conventional level of 200 mg N plant<sup>-1</sup> season<sup>-1</sup> commonly used for the production of containerized planting pine seedlings.

The loading rate (*l*) induced luxury nutrient uptake along a broad fertility range (300-500 mg N plant<sup>-1</sup> season<sup>-1</sup>), which increased seedling N content without significantly changing dry mass compared with the sufficiency index (Figures 3.15 and 3.17c). Compared with the C200 treatment, the maximum target rate (500 mg N plant<sup>-1</sup> season<sup>-1</sup>) induced 60, 62 and 12% increases in N, P and K uptake, respectively. The target threshold is higher than the 64 mg N plant<sup>-1</sup> season<sup>-1</sup> loading estimated for spruce seedlings (Salifu and Timmer, 2003). N supply in excess (*e*) of target levels (E700 and E900) induced toxicity associated with diminished plant growth (Figures 3.15, 3.17d and 3.17e).

### **3.3. White spruce**

#### **3.3.1. Seedling growth and nutrition**

By looking at the morphological data (Figure 3.19), the E750 treatment produced seedlings that met the outplanting height (25 cm) and RCD (2.5 mm) and had the lowest height:diameter ratio among all the exponential treatments.

Trends in plant nutrient concentration (Figure 3.21) were similar to those

shown in Figure 1.1, which increased gradually with N supply in the deficiency range due to growth dilution and rapidly in toxic range due to rapid nutrient accumulation.

Seedling P and K uptake (Figures 3.22e and 3.22f) increased with increasing fertilizer application rate by 461-565% for P and by 751-950% for K up to the E450 treatment, and then declined thereafter likely due to toxicity. The P and K content decreased with increasing N fertilizer application rate after E450.

Figure 3.23 demonstrated that seedling growth and nutritional responses to increased fertilization conformed closely to trends depicted in the conceptual model (Figure 1.1). For example, seedling growth significantly increased ( $P < 0.05$ ) with N supply in the deficiency range, remained fairly stable in the luxury consumption range and began to decline at higher N addition, suggesting toxicity (details in Section 3.3.2). Toxicity induced stunting in seedlings in the 1050 mg N per seedling or E1050 treatment (Figure 3.23). The consistent pattern in Figure 3.23 with trends in the conceptual model (Figure 1.1) confirms the suitability of the dose response model as a useful framework for identifying the optimum nutrient loading rate for spruce.

### **3.3.2. Identifying the optimum nutrient loading rate**

Assuming that N accumulated in the control seedlings reflected availability from the peat substrate, the  $n$  supply of N in the conceptual model (Figure 1.1)

was small (about  $1.79 \text{ mg N plant}^{-1} \text{ season}^{-1}$ , Figure 3.23). Supplemental fertilizer (f) countered deficiency and increased growth to the sufficiency level at E300 (Figures 3.23 and 3.25a). The sufficiency level found here for spruce was the same as the C300 commonly used for the production of containerized planting spruce.

The loading rate (*l*) that induces luxury nutrient uptake occurred between E300 and E450, in which seedling N content increased without significantly changing dry mass or seedling size (Figures 3.23 and 3.25b) when compared with the sufficiency level (E300 as previously discussed). Compared with the C300 treatment, the E450 treatment increased N, P and, K uptake by 10, 8 and 13%, respectively. Excessive fertilization (*e*) started at E750 which decreased dry mass and nutrient content as compared with the E450 treatment (Figures 3.23, 3.25c, 3.25d and 3.25e).

The process of identifying the optimum nutrient loading rates would not only help avoid over-fertilization and potential nutritional imbalances in seedlings, but also result in production of high quality seedlings with high internal tissue nutrient concentration which should help to optimize seedling field performance (Birge et al., 2006; Salifu and Jacobs, 2006; Salifu et al., 2009).

### **3.4. Similar patterns of the total nitrogen and available nitrogen in growth media and $\delta^{15}\text{N}$ data of seedlings with the three species**

The lower  $\delta^{15}\text{N}$  of fertilized seedlings than that in CK (Figure 3.8 for aspen; Figure 3.16 for jack pine; Figure 3.24 for white spruce) implied increasing N uptake by fertilization;  $\delta^{15}\text{N}$  of fertilizer is generally lower than that of soil (Robinson, 2001; Kwak, et al., 2009). N isotope can be fractionated through assimilation and translocation, inducing relatively low  $\delta^{15}\text{N}$  in root and stem tissue compared to that in leaves. The  $\delta^{15}\text{N}$  of seedlings in CK decreased over time (Figures 3.8, 3.16 and 3.24). The possible scenario is that, during early stage, seedlings used N with relatively high  $\delta^{15}\text{N}$  produced from previous N isotope fractionation processes, e.g.,  $\text{NH}_3$  volatilization and denitrification, increasing  $\delta^{15}\text{N}$  while seedlings in CK, in later periods, took up N newly-mineralized from peat with relatively low  $\delta^{15}\text{N}$ .

The nutrient loading method significantly increased both the total N and available N concentrations for each species (Figure 3.10 for aspen; Figure 3.18 for jack pine; Figure 3.26 for white spruce) in the growth media by comparing conventional and unfertilized treatments. The improved nutrient availability in the growth media would be another aspect that the nutrient loading technique could be beneficial for the success of oil sands reclamation. The exponentially fertilized seedlings always exhibited stable internal nutrient levels during the active growing seasons than those of conventionally fertilized trees, which experienced

declining nutrient contents with time because of growth dilution (Salifu and Timmer, 2003).

### **3.5. Discussion of model suitability**

The proposed conceptual model of nutrient loading suggested plant growth and nutrient status follow a curvilinear pattern ranging from nutrient deficiency to toxicity with increased fertilization (Figure 1.1). Close correspondence of experiment data (Figures 3.7, 3.15 and 3.23) with trends in Figure 1.1 demonstrates model suitability for application in both hardwood and softwood seedling culture. Similar results have been noted for containerized production of black spruce (Salifu and Timmer, 2003) and red oak seedlings (Salifu and Jacobs, 2006). Thus, exponential nutrient loading can be effectively translated from a controlled greenhouse setting to practical nursery production systems as demonstrated here and elsewhere (McAlister and Timmer, 1998). Exponential nutrient loading induced luxury nutrient uptake and storage in plant tissues in agreement with results of other studies (Imo and Timmer, 1992; McAlister and Timmer, 1998; Qu et al., 2003; Salifu and Timmer, 2003). In addition, the exponential fertilizer delivery schedule was more effective in promoting nutrient acquisition and storage in seedling tissues than the current constant fertilization approach used in practice, which corroborates with results of other studies (Timmer and Aidelbaum, 1996; Timmer, 1997). For example, N, P and K

increased by 60, 62 and 12%, respectively, in E500 compared with C200 for jack pine. Fertilization and nutrition monitoring of seedlings crops are often based on plant tissue analysis expressed in nutrient concentration alone (Timmer et al., 1991). From this study, it can be found that a more integrated approach is to utilize plant dry mass, plant nutrient concentration, and content (Figure 1.1) which can significantly improve diagnostic precision (Figures 3.7, 3.15 and 3.23).

#### **4. Conclusions**

In conclusion, the optimal exponential fertility treatment for aspen, pine and spruce was E240, E500 and E450, respectively, according to results from the modified exponential model in the 2010 study. Nutrient loading promoted seedling stem and root growth, suggesting that the approach may have the potential to increase competitive success and drought avoidance of these seedlings when transplanted under field conditions (Timmer, 1997). Even though conventional methods could produce seedlings that meet the outplanting size requirement, the regime would decrease N content in tissues over time, whereas all the exponential treatments would maintain relatively stable nutrient content and increase the nutrient reserves over time. The greater nutrient reserves in the nutrient loaded seedlings could help seedlings overcome competitive stress for nutrients from the competing vegetation in the field as these nutrient reserves can promote internal nutrient redistribution to support new growth of seedlings soon

after transplanting (Salifu et al., 2009). Furthermore, broadcast field fertilization may stimulate growth of competing vegetation and cause nutrient leaching, while building up nutrient storage in seedlings at the nursery stage provides a better means to promote seedling performance after outplanting in the field. Moreover, Timmer (1997) also reported that even though the exponentially fertilized trees had lower biomass, P and K contents in shoots before planting, the first year growth and nutrition after outplanting of these seedlings were better than conventionally fertilized seedlings. The validated model provides a comprehensive framework for rationalizing and quantifying optimum fertilizer prescriptions, which may improve plant nutrient diagnosis and enhance the quality of nursery stock for field plantings.

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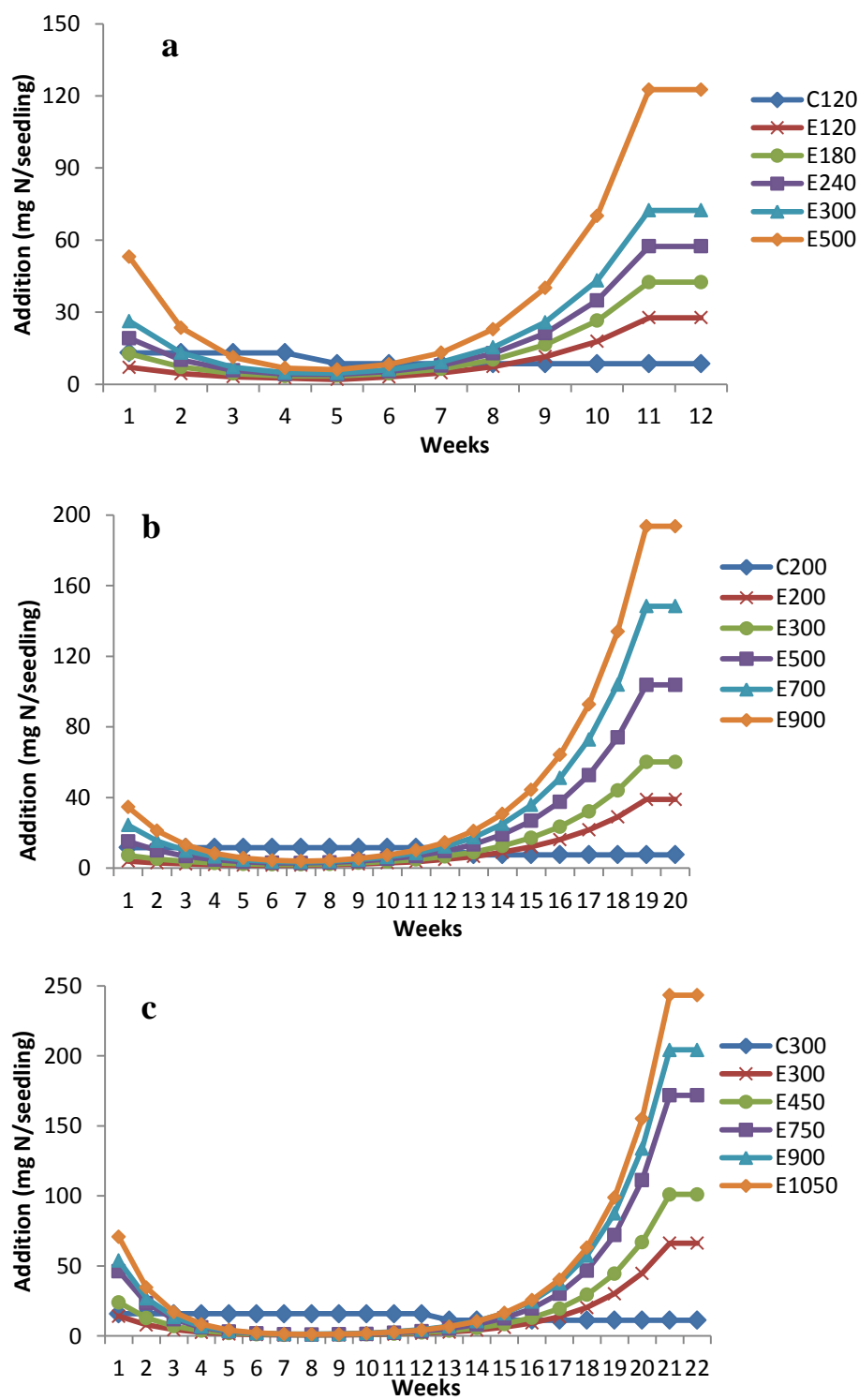
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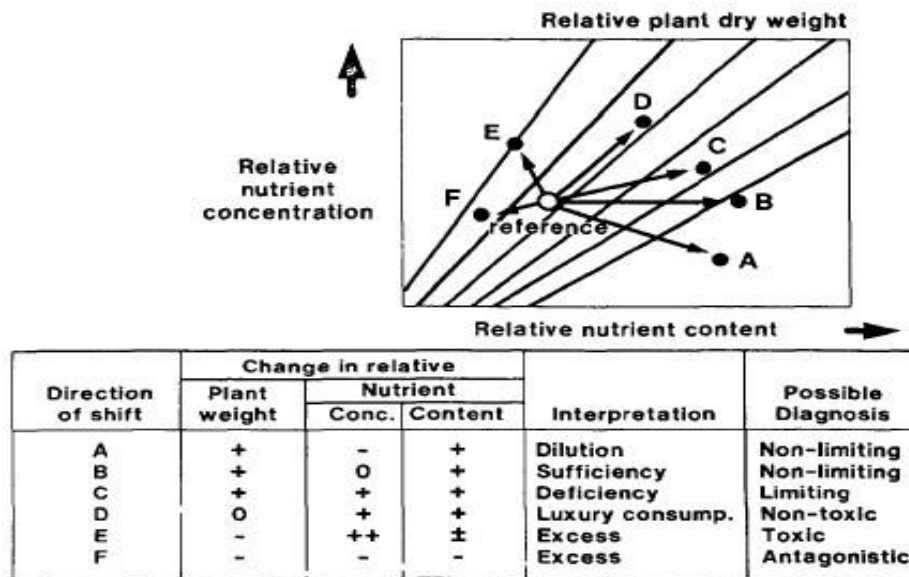
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**Table 3.1.** Sampling schedule during seedling's growth period

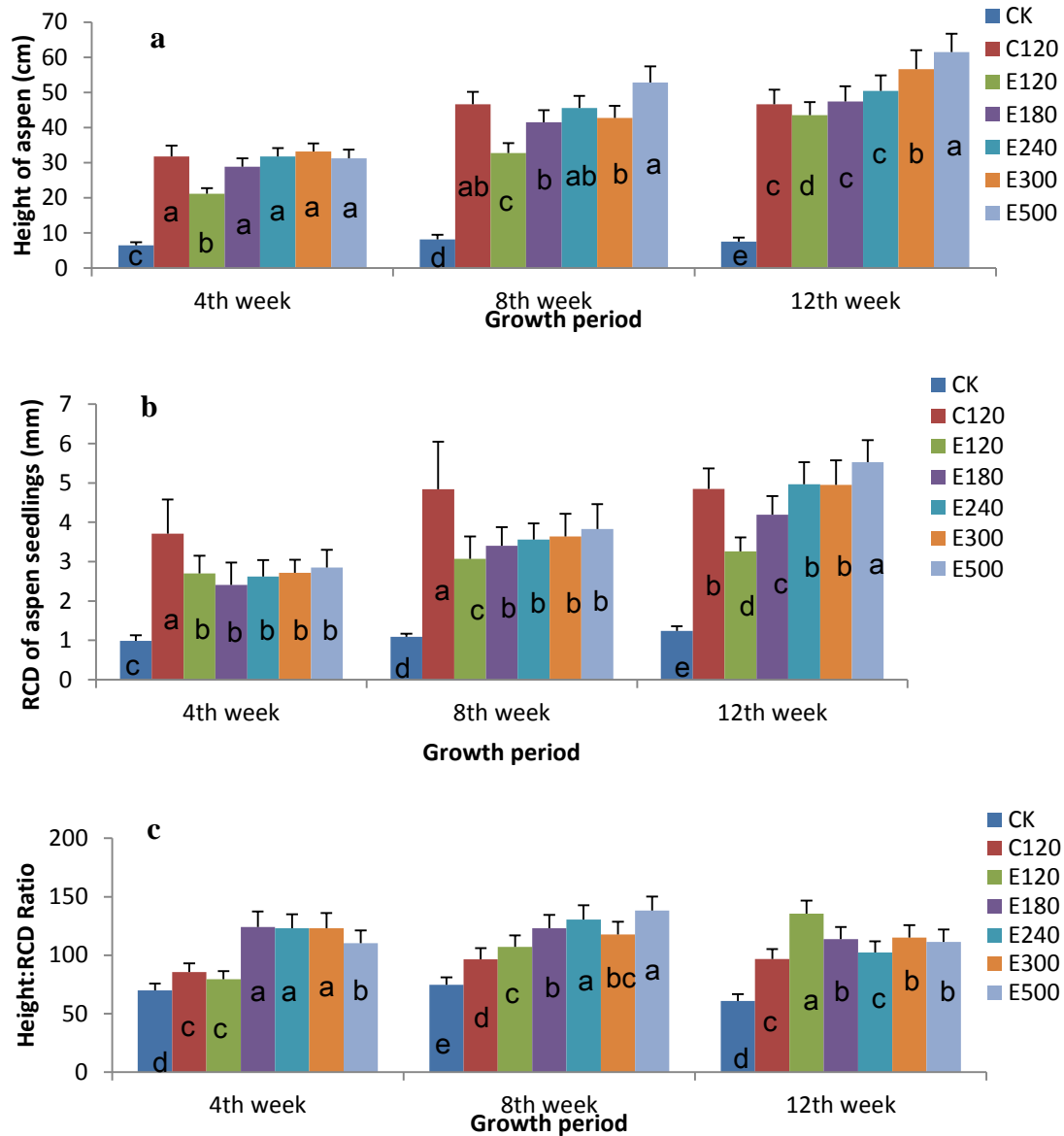
Species		Sampling time		
Aspen	week 4	week 8	week 12	
Jack Pine	week 6	week 15	week 21	
White Spruce	week 8	week 16	week 23	



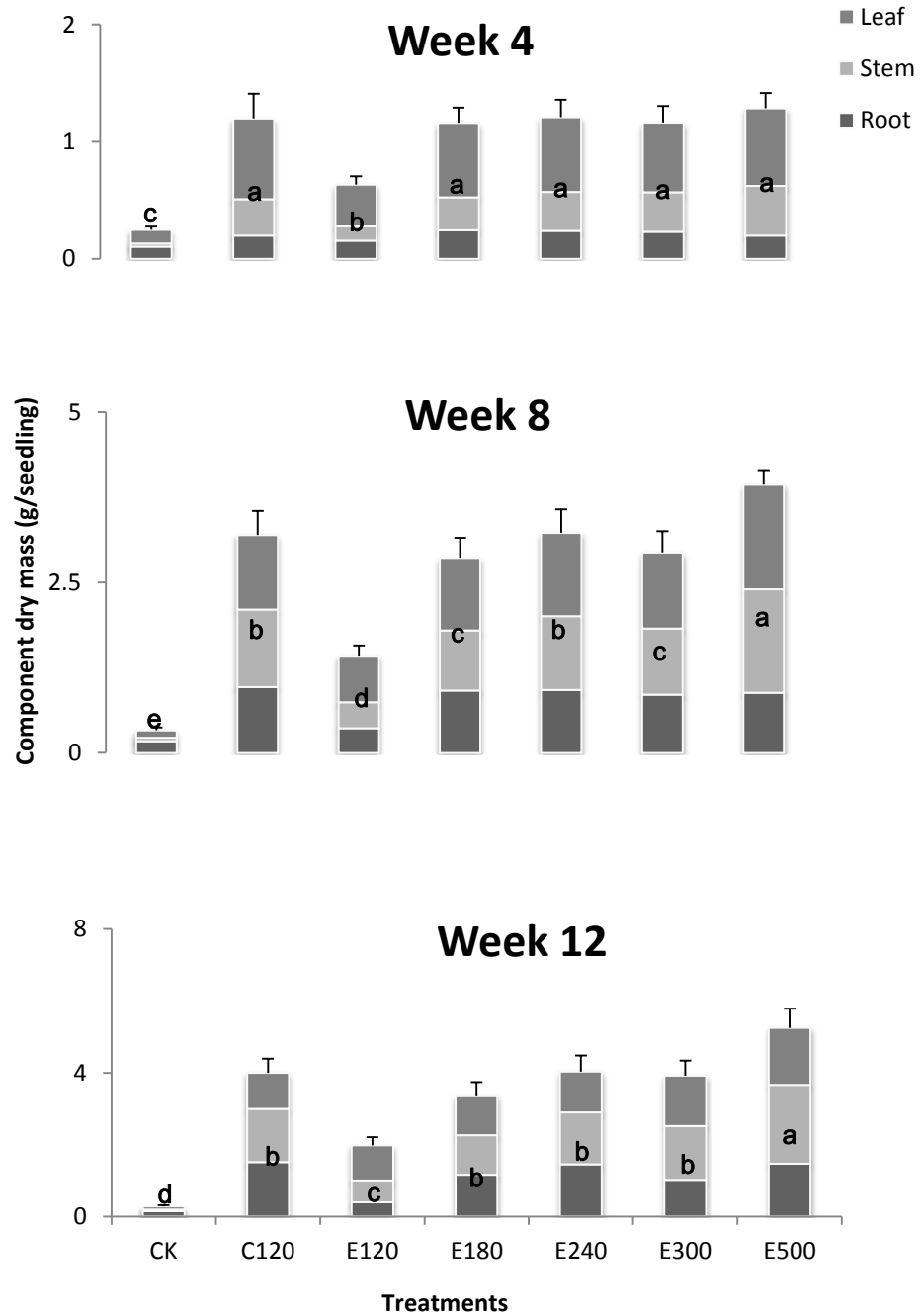
**Figure 3.1.** The detailed fertilizer delivery schedule for three species. **a.** Aspen; **b.** Jack pine; and **c.** White spruce.



**Figure 3.2.** Interpretation of directional differences (vectors) in nutrient concentration, nutrient content, and dry weight between plants of contrasting treatments (Timmer and Armstrong, 1987b). A vector (arrow) for each treatment represents the relative difference in the concentration, content and biomass between plants, and originates from one point when the status of the reference seedling is normalized to 100. Diagnosis is based on vector analysis or on the direction and magnitude of the vector. The directional shift, observed as increase (+), decrease (-) or no change (o) in each of the three parameters, is matched with a sequence shown in the box of Figure 3.2 suggesting a specific nutrient disorder or condition. The degree of deficiency, excess or dilution of individual treatment is reflected by vector magnitude or length. Simultaneous comparisons of several elements on a relative basis facilitate assessment of nutrient balance (Salifu and Timmer, 2003).

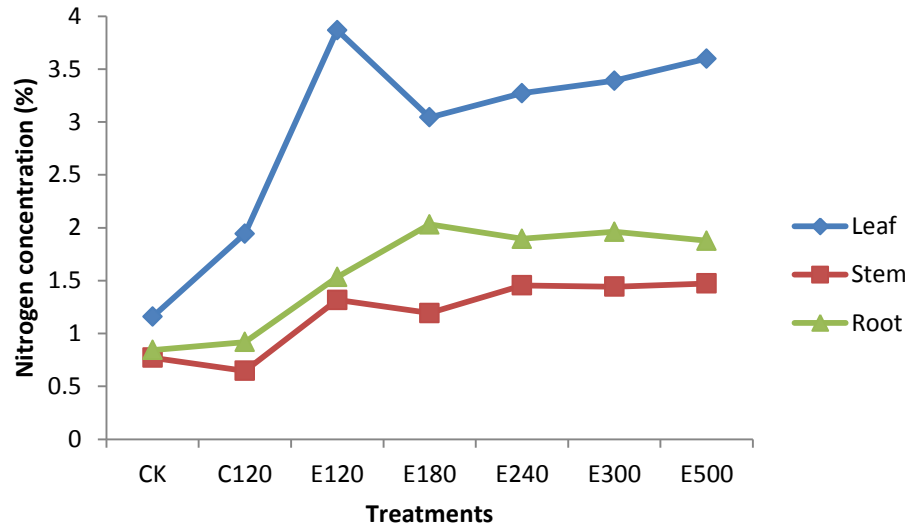


**Figure 3.3.** Aspen morphological data in 2010. **a.** Stem height under different treatments over the entire growth period; **b.** Root collar diameter (RCD) under different treatments over the entire growth period; and **c.** Height:RCD ratio under different treatments over the entire growth period. \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*

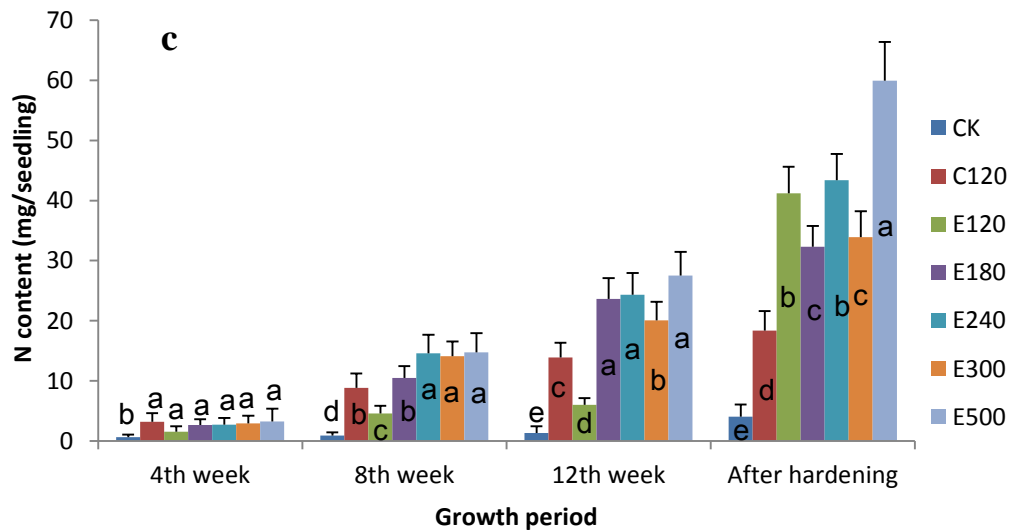
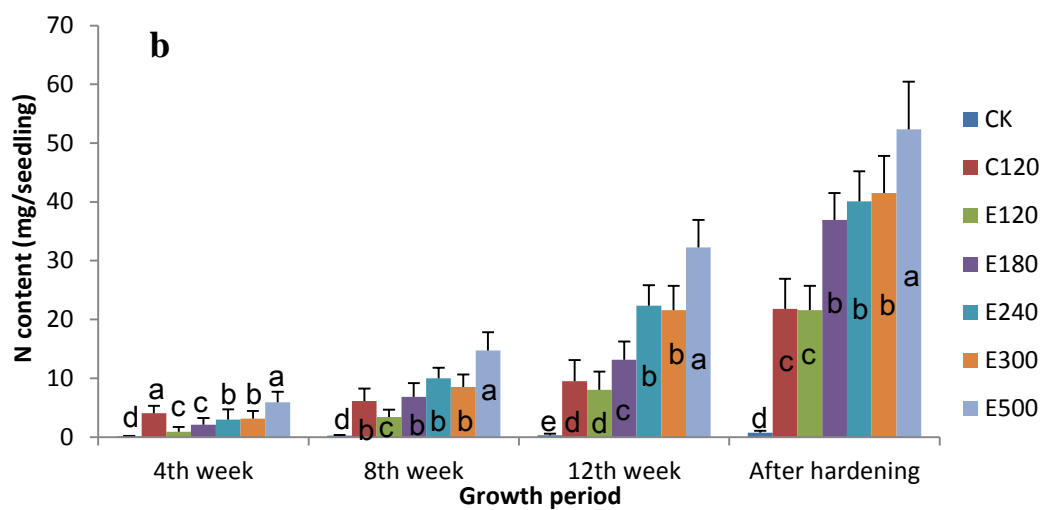
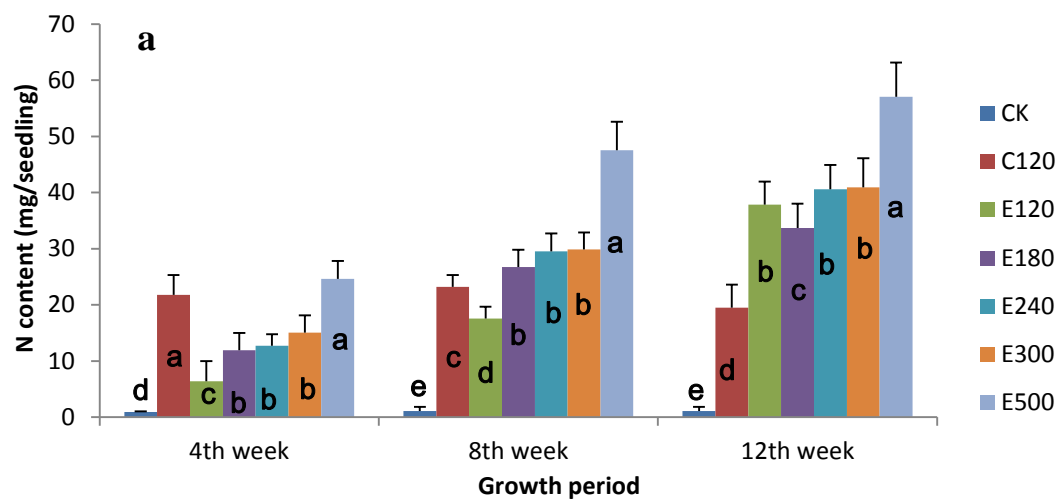


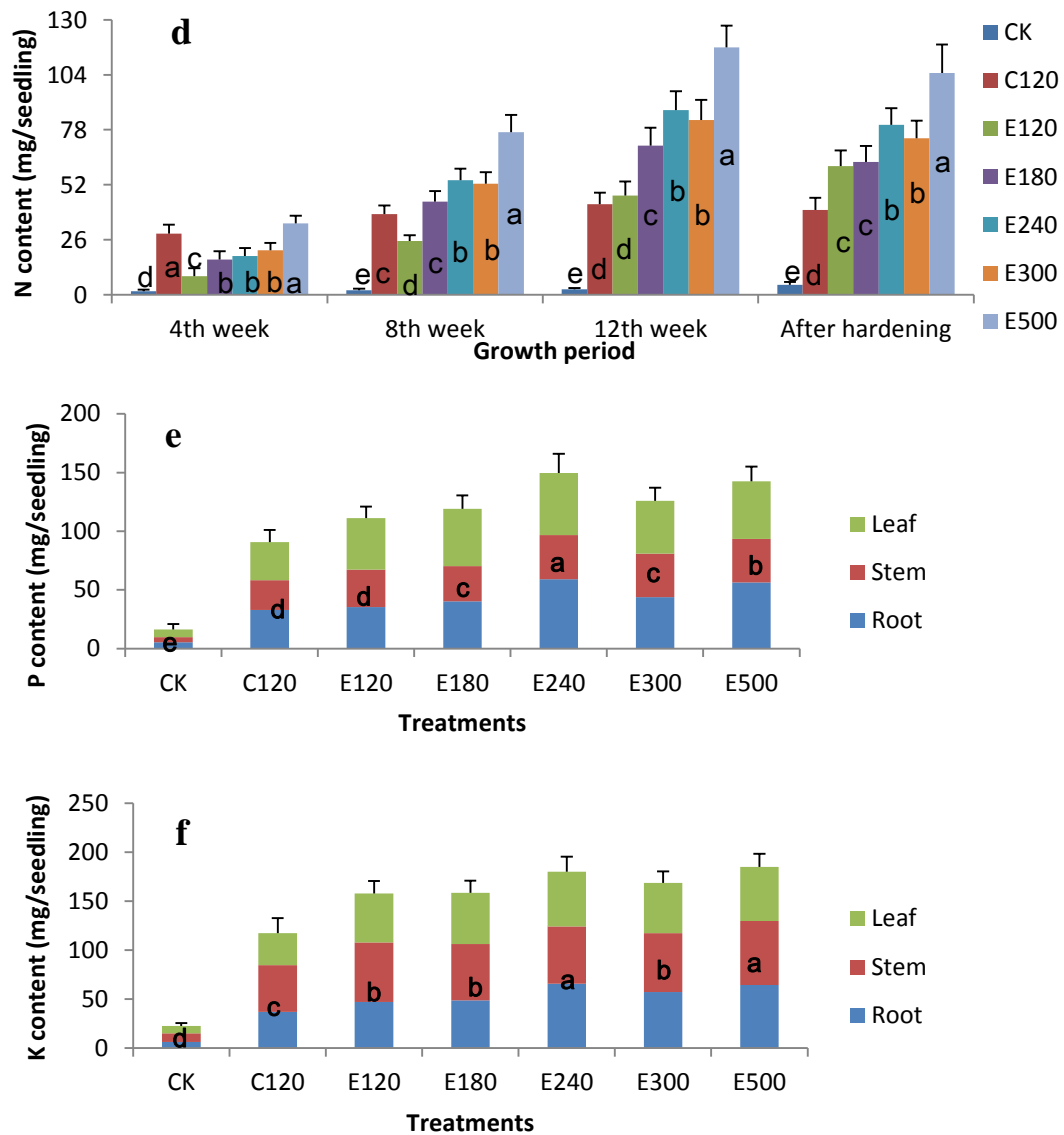
**Figure 3.4.** Aspen component dry mass under different treatments over the entire growth period in 2010. \* *Treatments followed by the same letter in the same sampling are not significantly different ( $p>0.05$ ). Mean separation was based on the total biomass per seedling. Error bars are standard deviations of the means.*



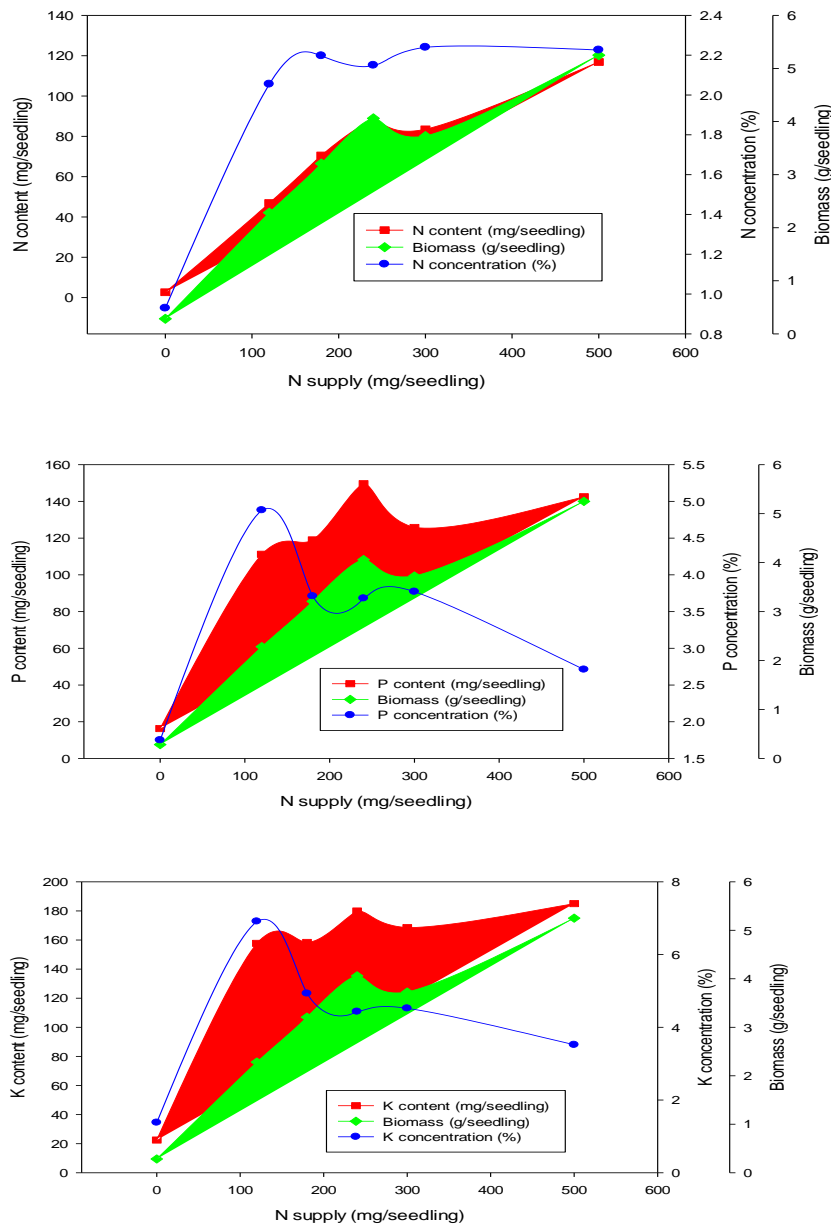


**Figure 3.5.** Aspen N concentration under different treatments in the last collection (12<sup>th</sup> week) in 2010.

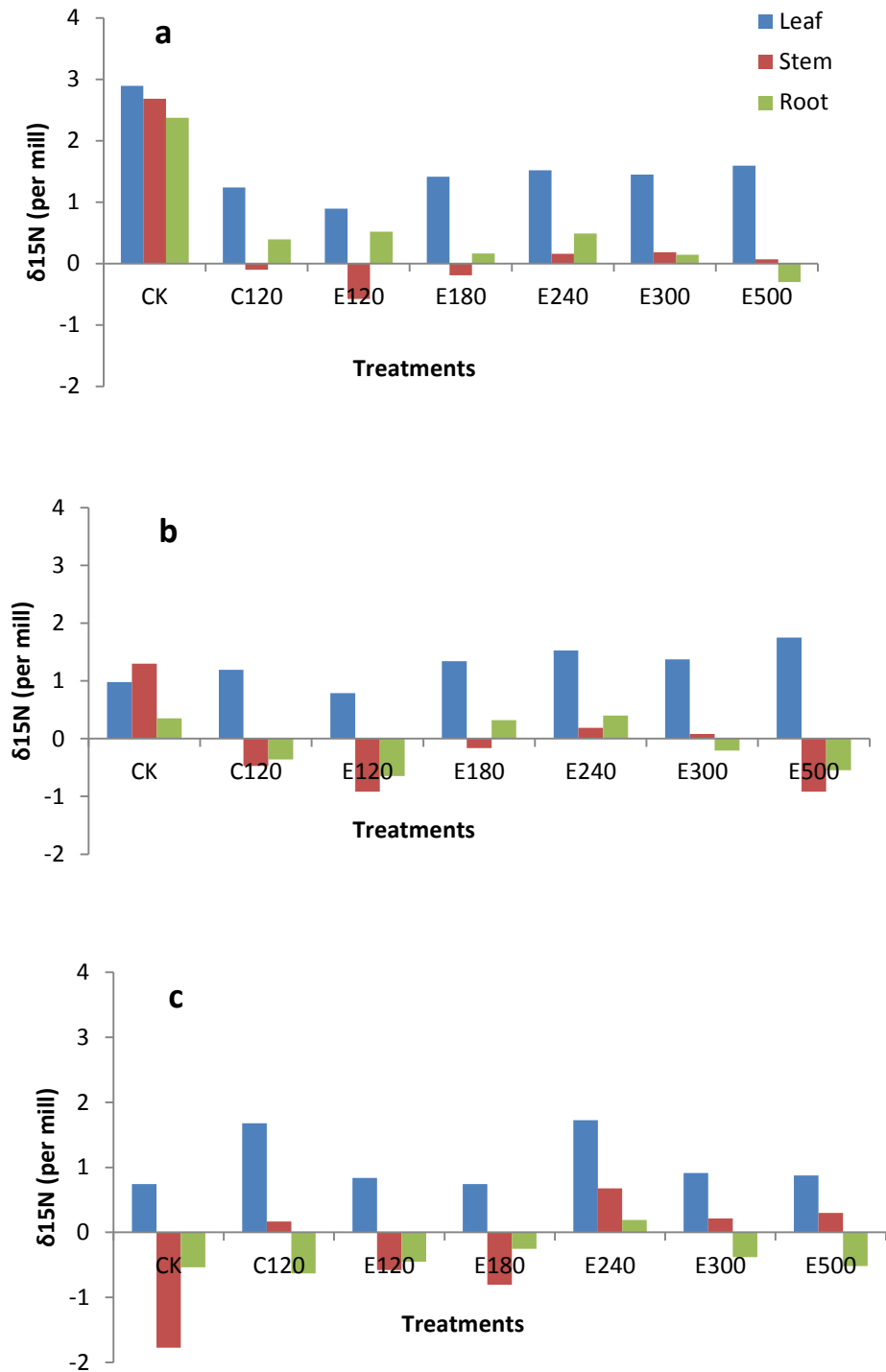




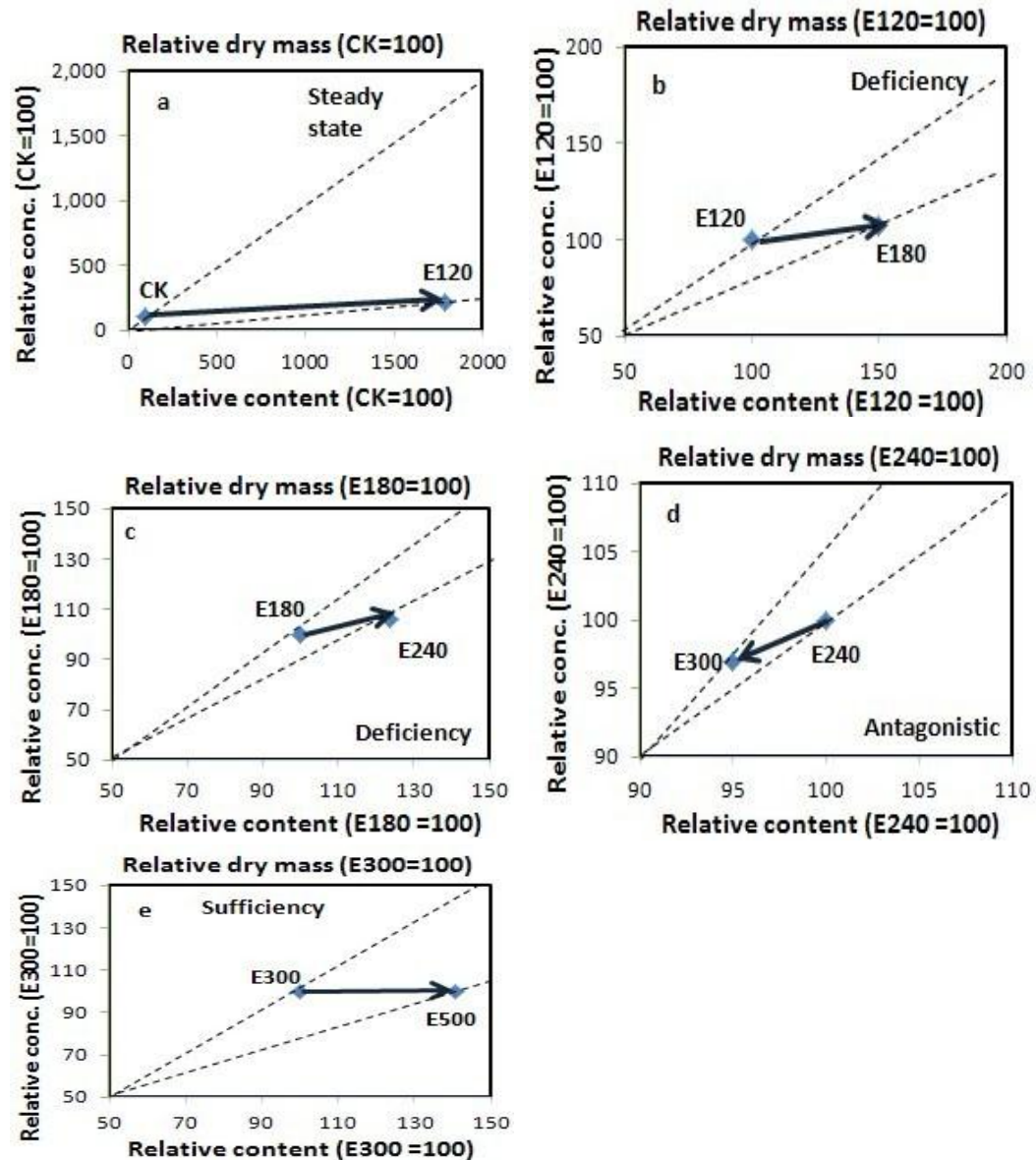
**Figure 3.6.** Component nitrogen, phosphorus and potassium content of aspen under different treatments over the entire growth period in 2010. **a.** Leaf N content; **b.** Stem N content; **c.** Root N content; **d.** Total N content; **e.** Total P content in different parts of the seedling; and **f.** Total K content in different parts of the seedling. \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*



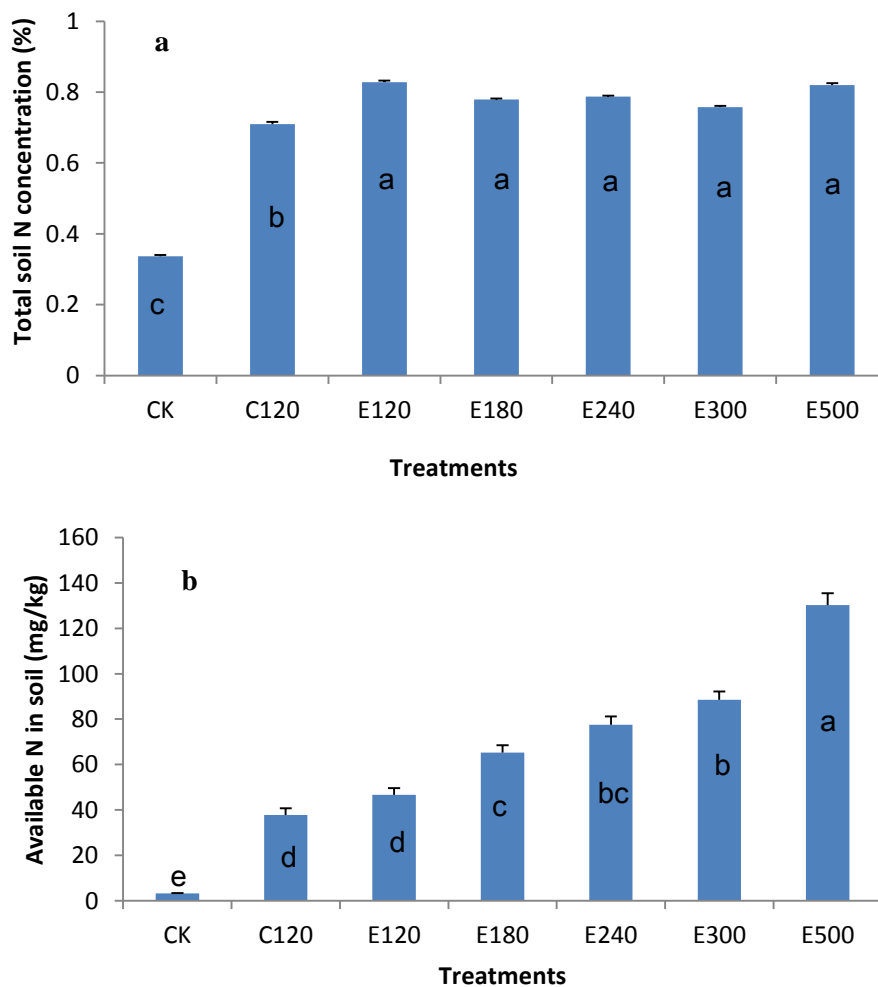
**Figure 3.7.** Relationships among nutrient supply, plant growth and tissue nutrient content and concentration of aspen at 12<sup>th</sup> week in 2010. Treatments used were CK, E120, E180, E240, E300 and E500 representing N supply rates of 0, 120, 180, 240, 300 and 500 mg N plant<sup>-1</sup> season<sup>-1</sup>, respectively, under the modified exponential fertilization model.



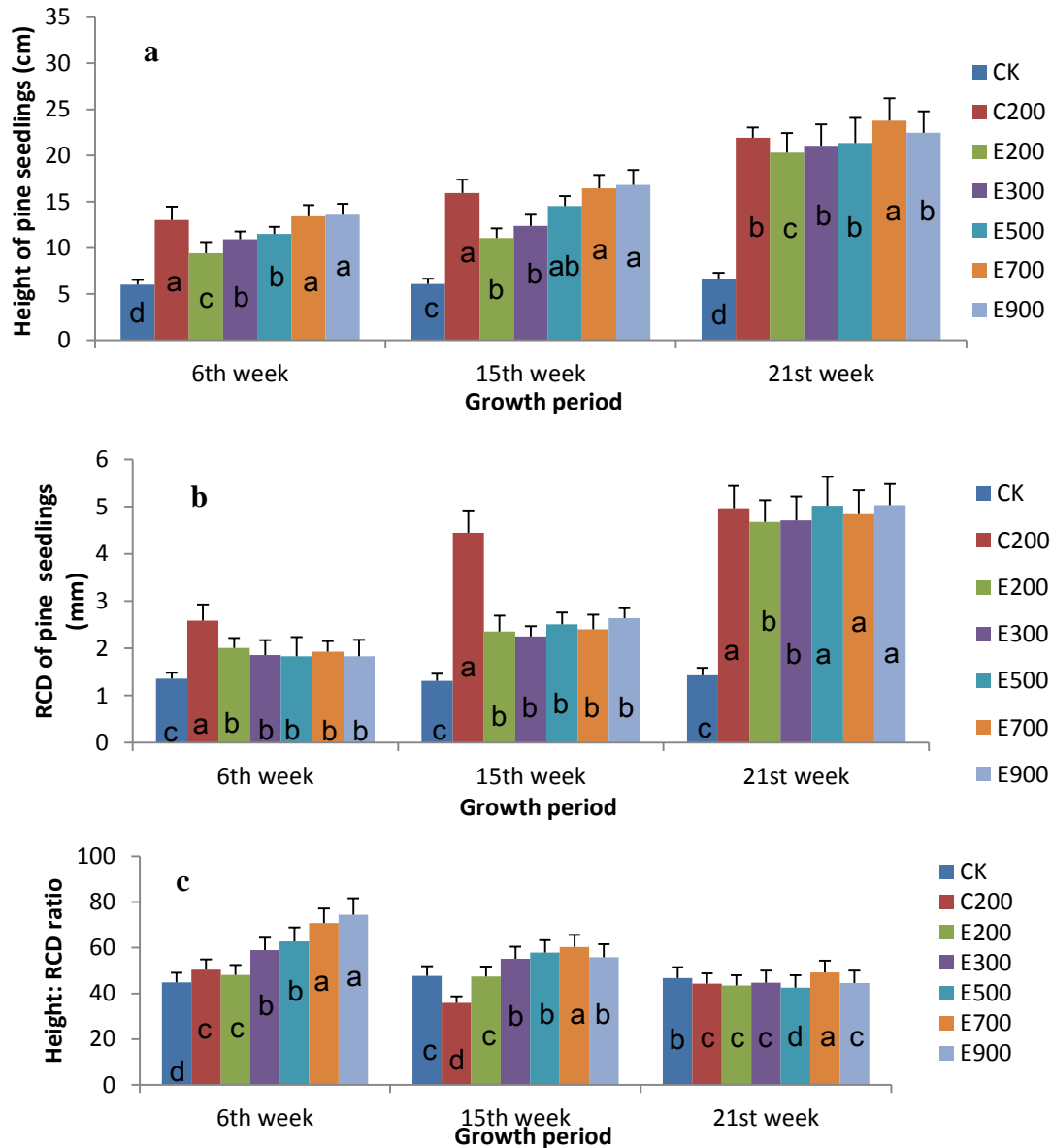
**Figure 3.8.** Component  $\delta^{15}\text{N}$  of aspen seedlings under different treatments over the entire growth period. **a.** at 4<sup>th</sup> week; **b.** at 8<sup>th</sup> week; and **c.** at 12<sup>th</sup> week.



**Figure 3.9.** Vector monogram of relative change in plant dry mass, N content, and concentration in aspen fertilized at different rates. Corresponding value at each point indicates seasonal dose applied (mg N/seedling; CK represents unfertilized or the control treatment). The type of nutritional response induced by enrichment is characterized by shift (or vector) direction and magnitude, described in Figure 3.2.

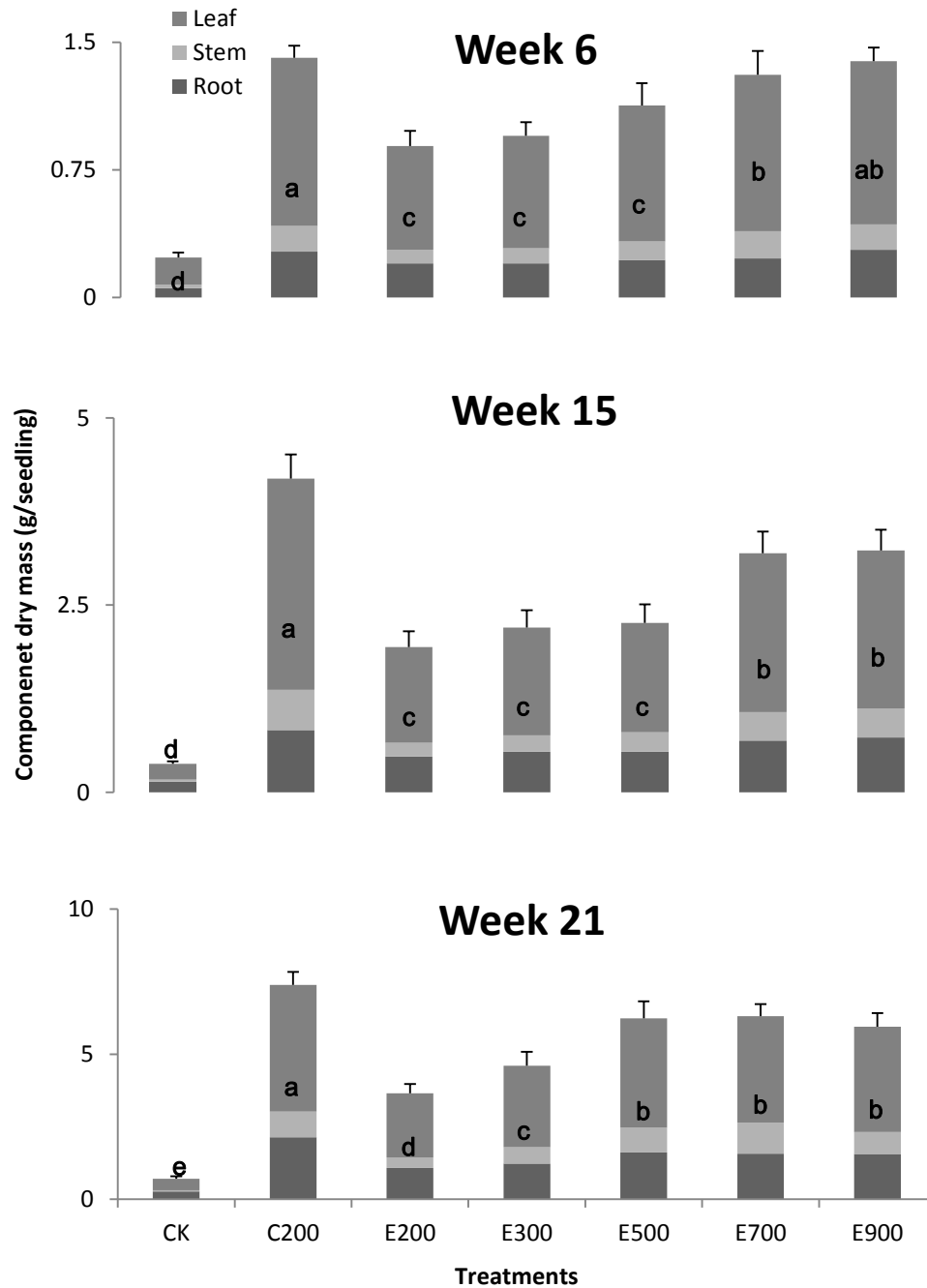


**Figure 3.10.** Nitrogen information in styroblocks in 2010. **a.** Total N concentration in the growth media in styroblocks planted to aspen in the last collection; and **b.** Available N in the growth media in styroblocks planted to aspen in the last collection.\* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*

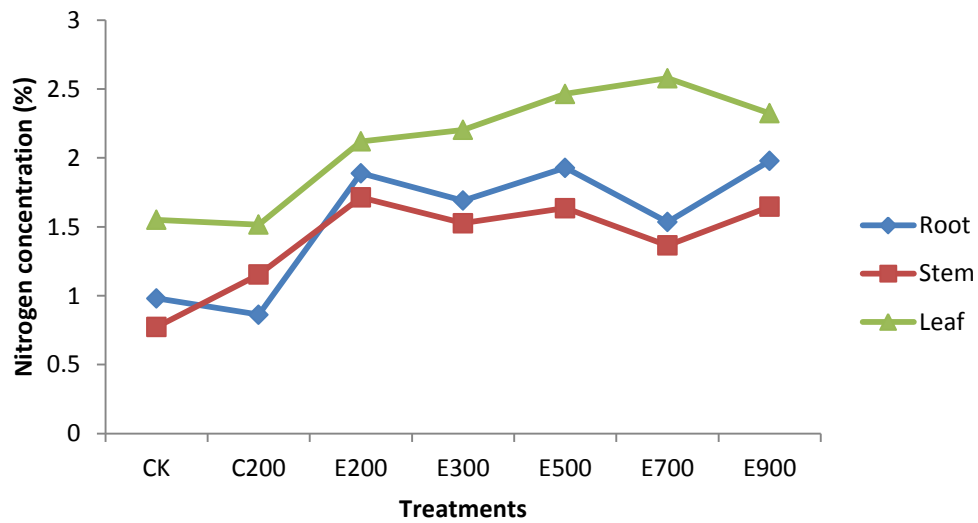


**Figure 3.11.** Pine morphological data in 2010. **a.** Stem height under different treatments over the entire growth period; **b.** Root collar diameter (RCD) under different treatments over the entire growth period; and **c.** Height:RCD ratio under different treatments over the entire growth period. \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*

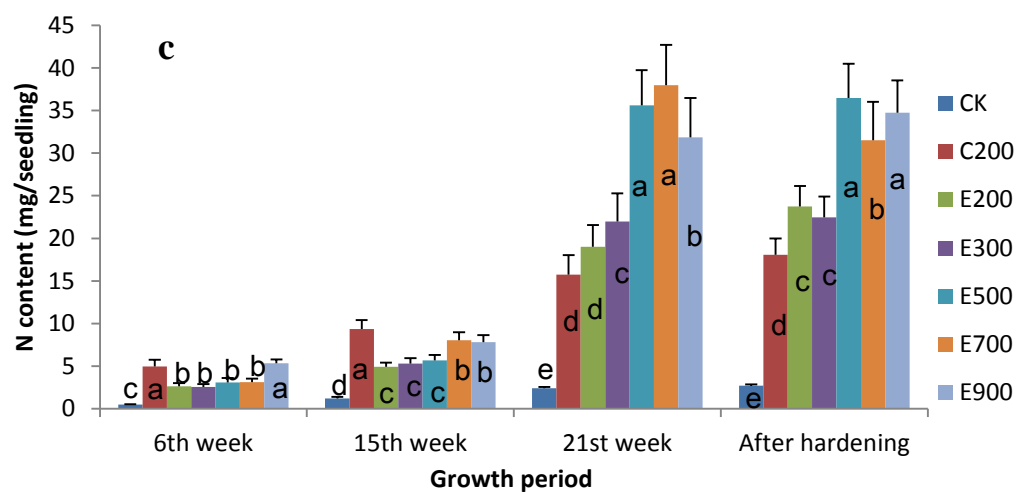
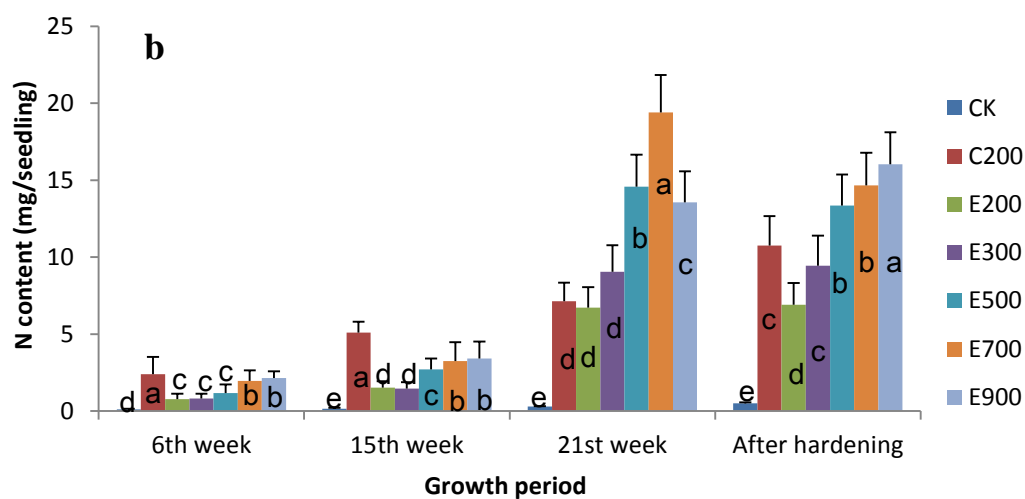
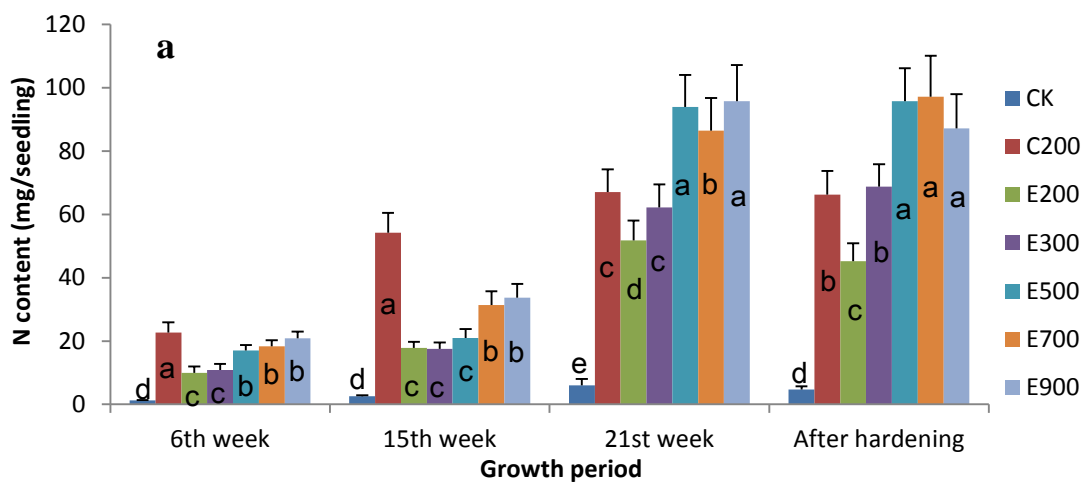


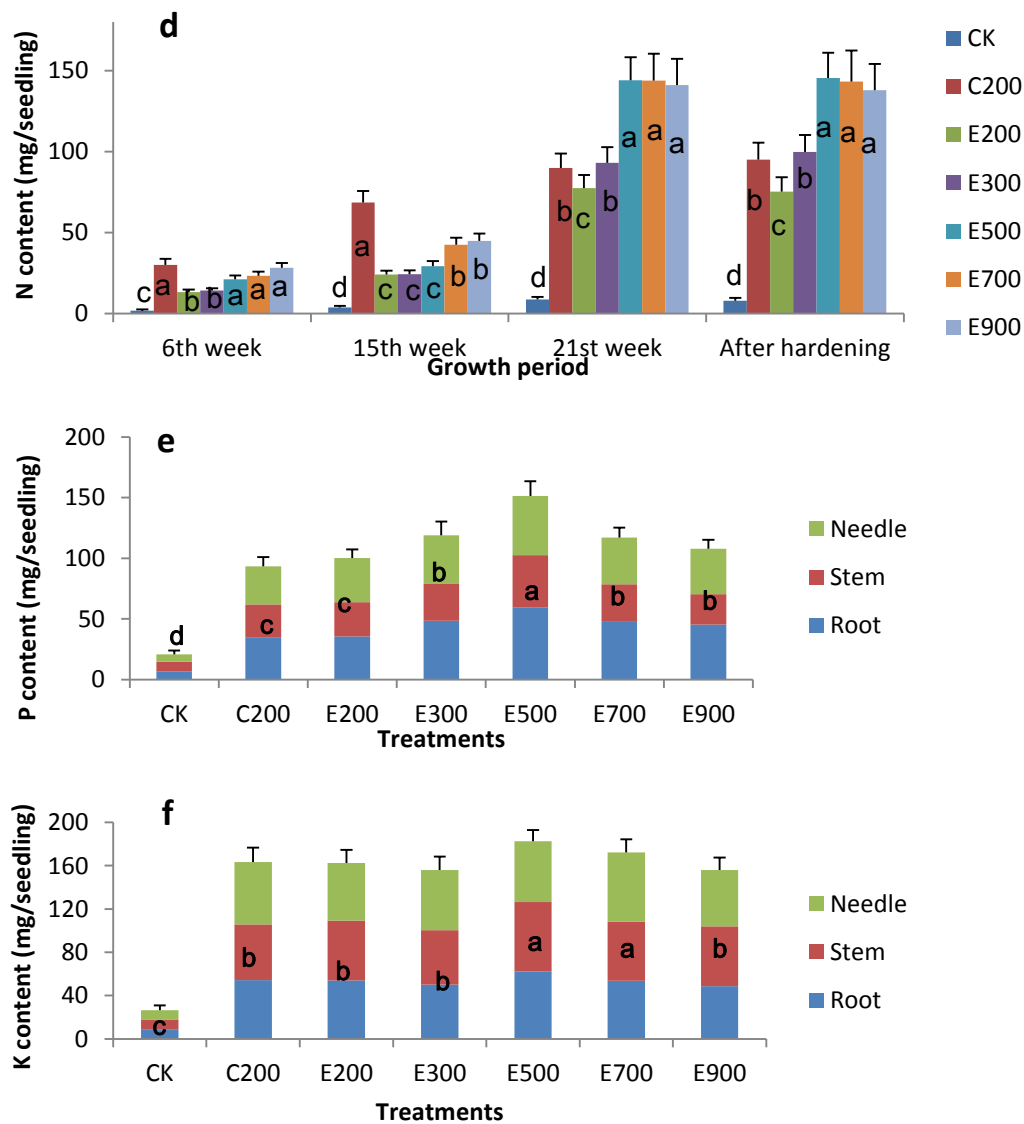


**Figure 3.12.** Pine component dry mass under different treatments over the entire growth period in 2010. \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Mean separation was based on the total biomass per seedling. Error bars are standard deviations of the means.*

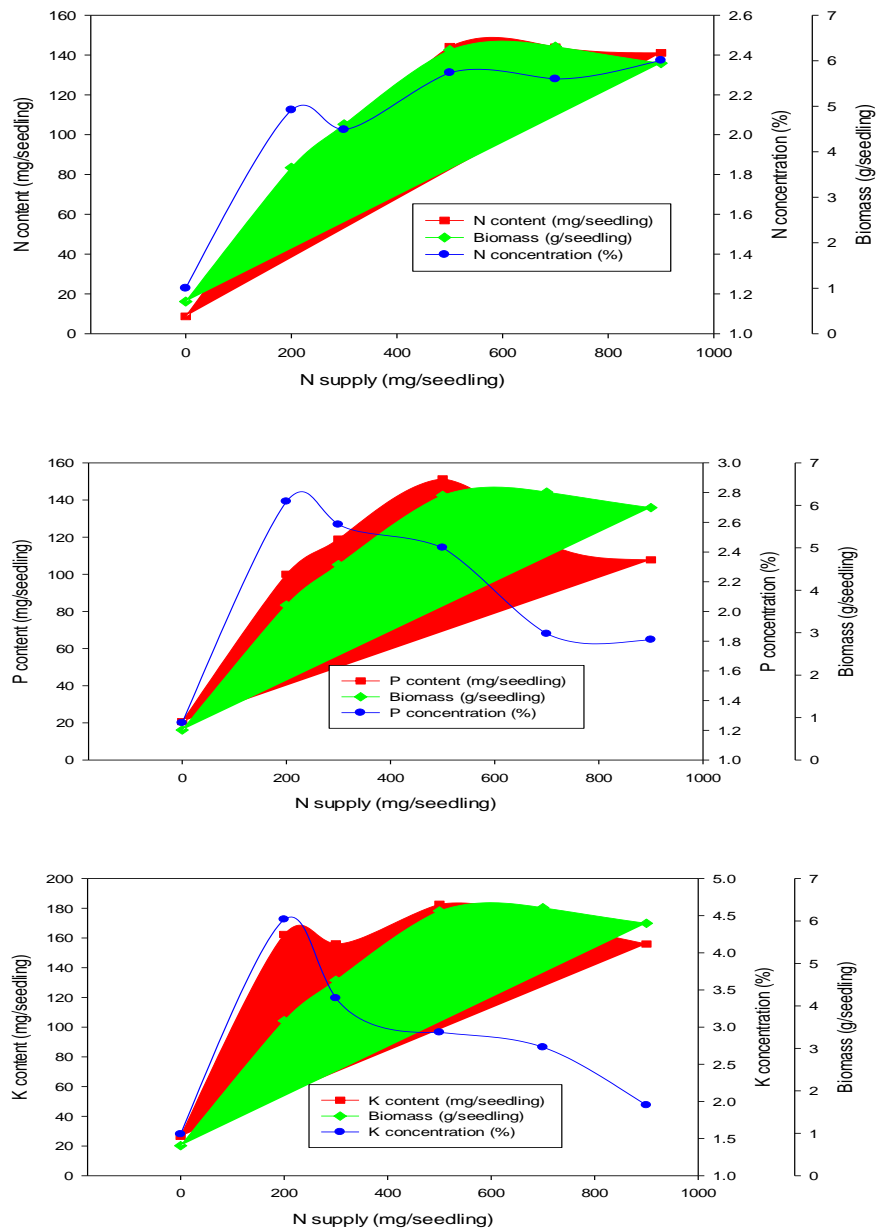


**Figure 3.13.** Pine N concentration under different treatments in the last collection (in the 21<sup>st</sup> week) in 2010.

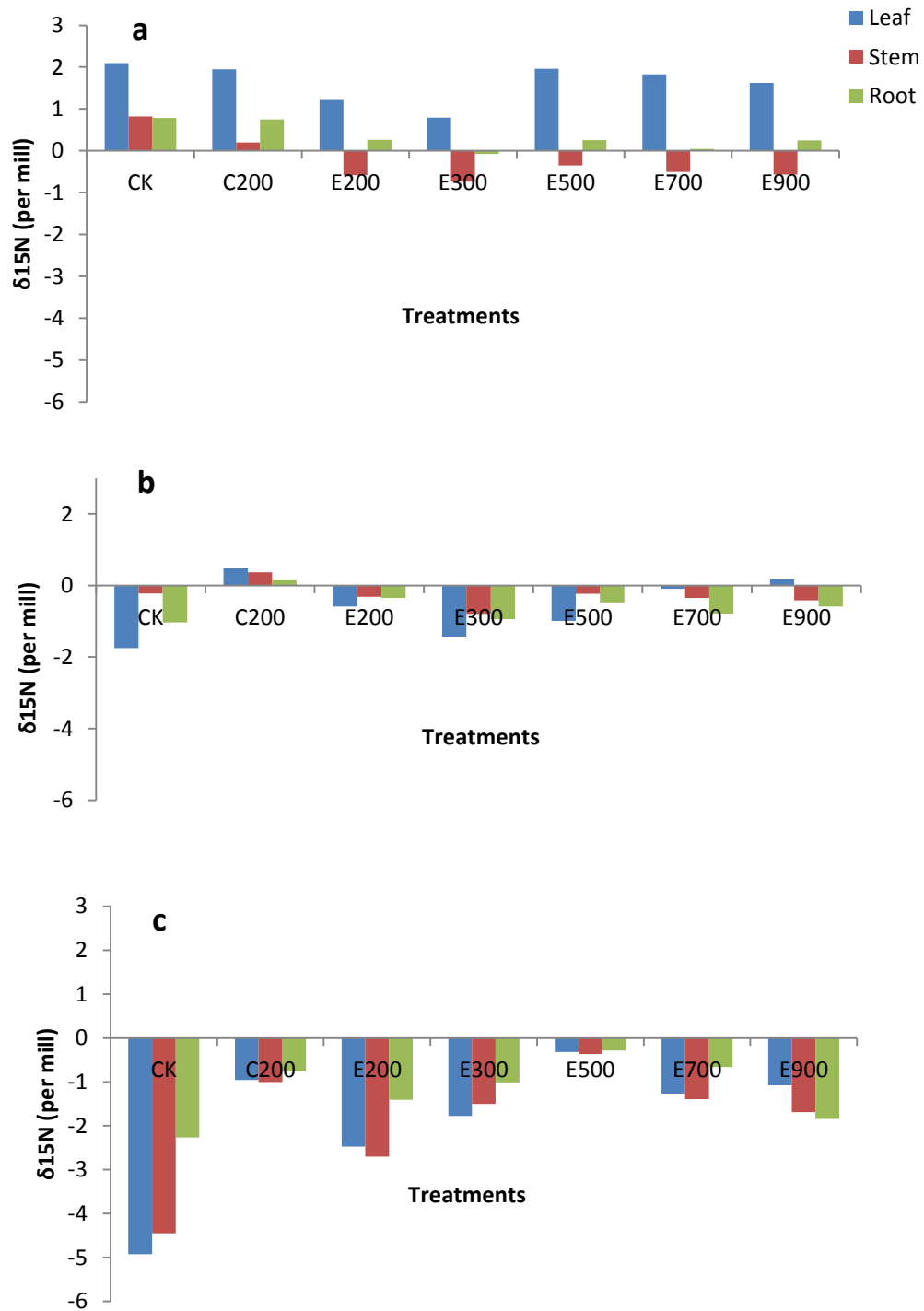




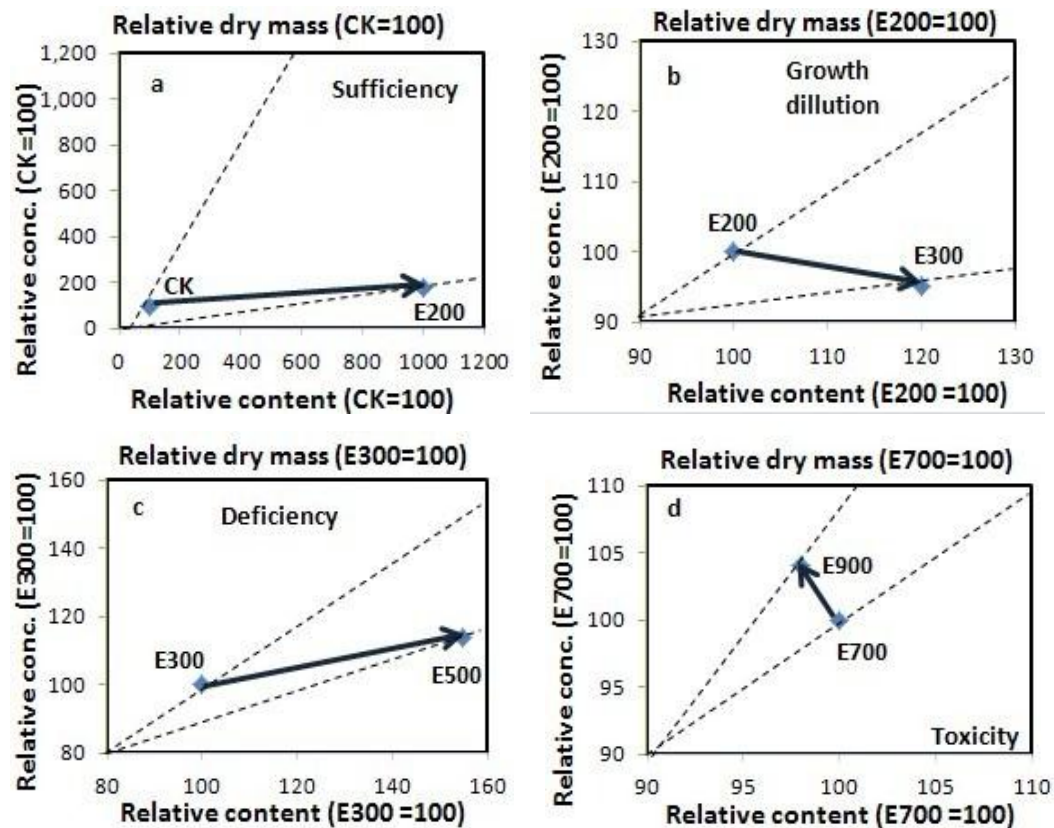
**Figure 3.14.** Component nitrogen, phosphorus and potassium content of pine seedlings under different treatments over the entire growth period in 2010. **a.** Needle N content; **b.** Stem N content; **c.** Root N content; **d.** Total N content; **e.** Total P content in different parts of the seedling; and **f.** Total K content in different parts of the seedling \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of means.*



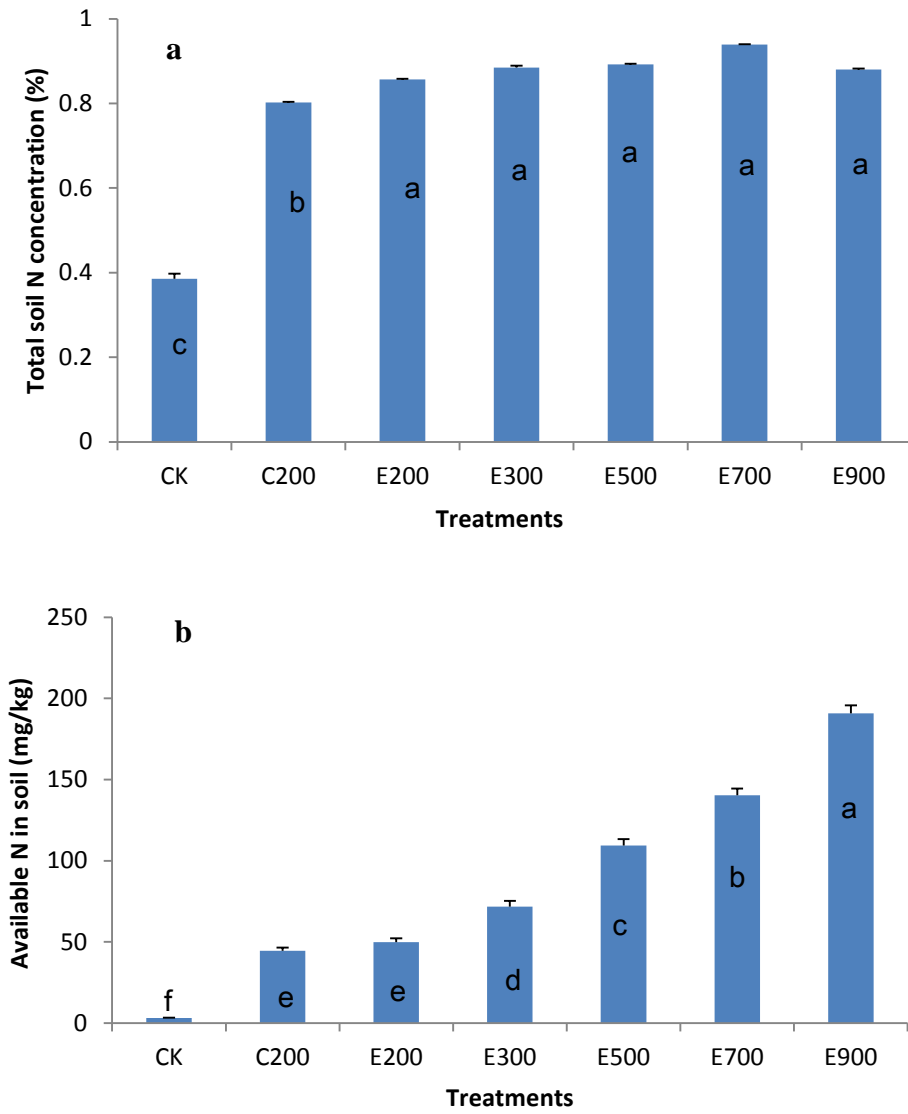
**Figure 3.15.** Relationships among nutrient supply, plant growth and tissue nutrient content and concentration of jack pine at week 21 in 2010. Treatments used were CK, E200, E300, E500, E700 and E900 representing N supply rates of 0, 200, 300, 500, 700 and 900 mg N plant<sup>-1</sup> season<sup>-1</sup>, respectively, under the modified exponential fertilization model.



**Figure 3.16.** Component  $\delta^{15}\text{N}$  of pine seedlings under different treatments over entire growth period. **a.** at 6<sup>th</sup> week; **b.** at 15<sup>th</sup> week; and **c.** at 21<sup>st</sup> week.

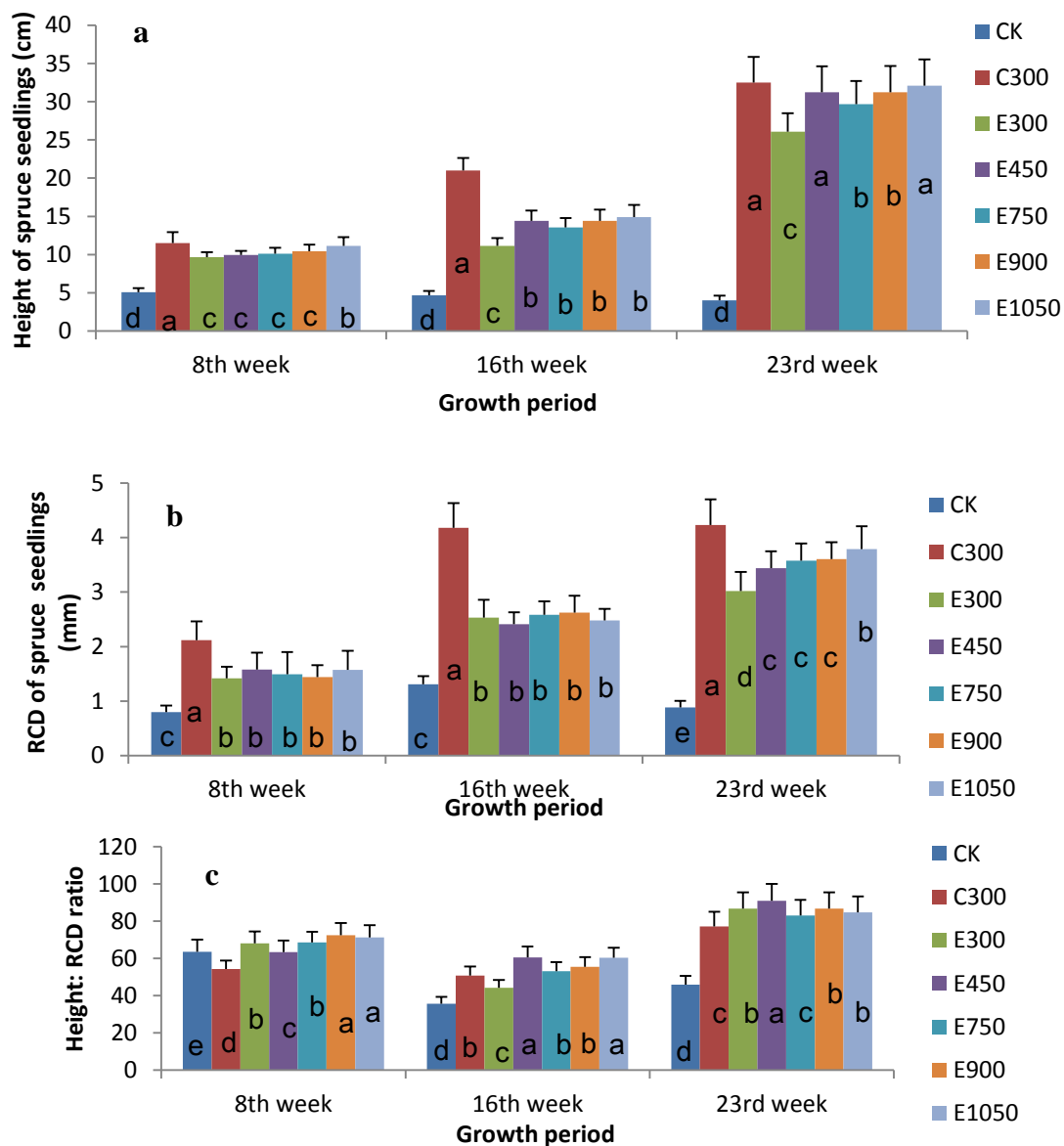


**Figure 3.17.** Vector monogram of relative change in plant dry mass, N content, and concentration in jack pine fertilized at different rates. Corresponding value at each point indicates seasonal dose applied (mg N/seedling; CK represents unfertilized or the control treatment). The type of nutritional response induced by enrichment is characterized by shift (or vector) direction and magnitude, described in Figure 3.2.

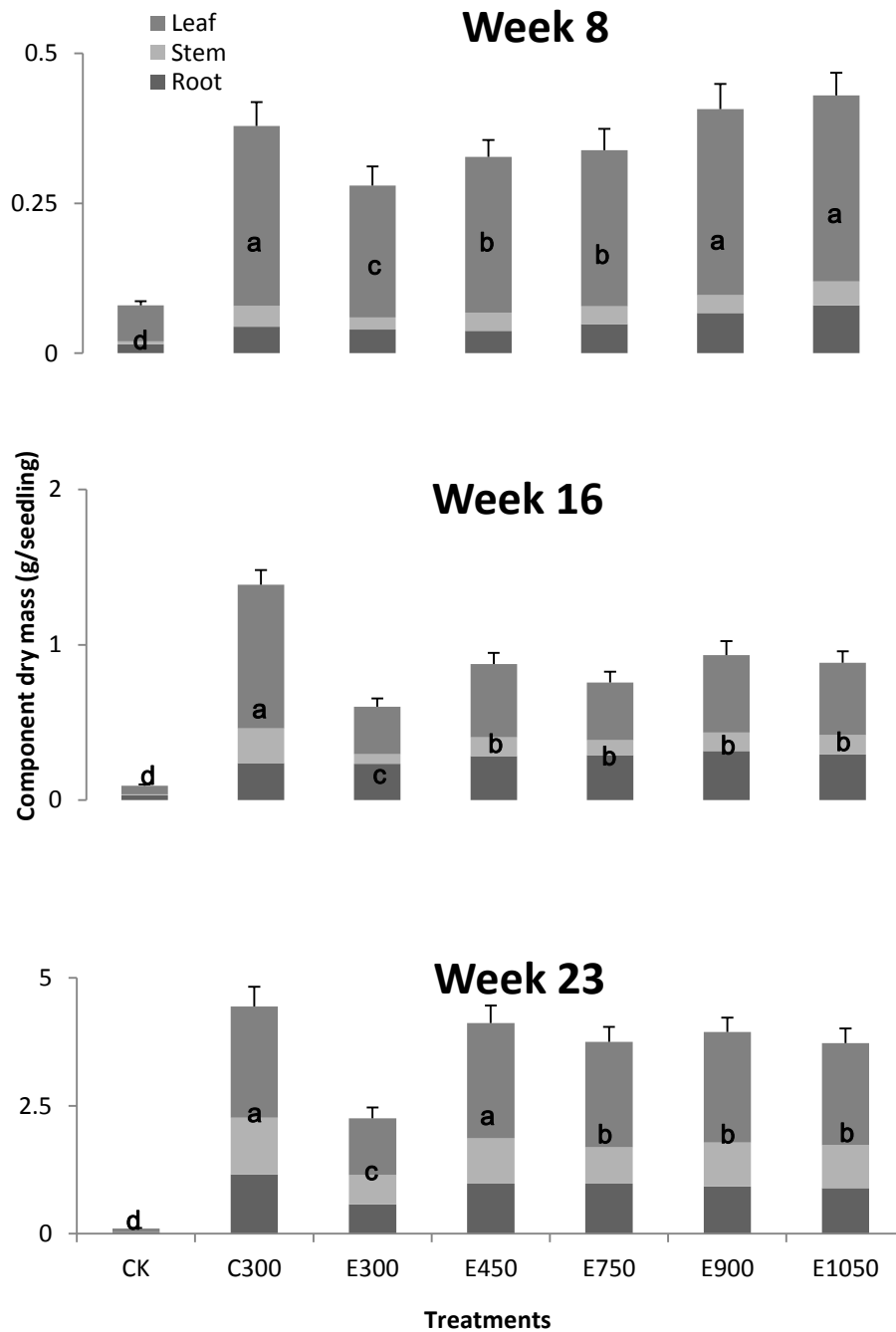


**Figure 3.18.** Nitrogen information in styroblocks in 2010. **a.** Total N concentration in the growth media in styroblocks planted to pine in the last collection; and **b.** Available N in the growth media in styroblocks planted to pine in the last collection in 2010 \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*

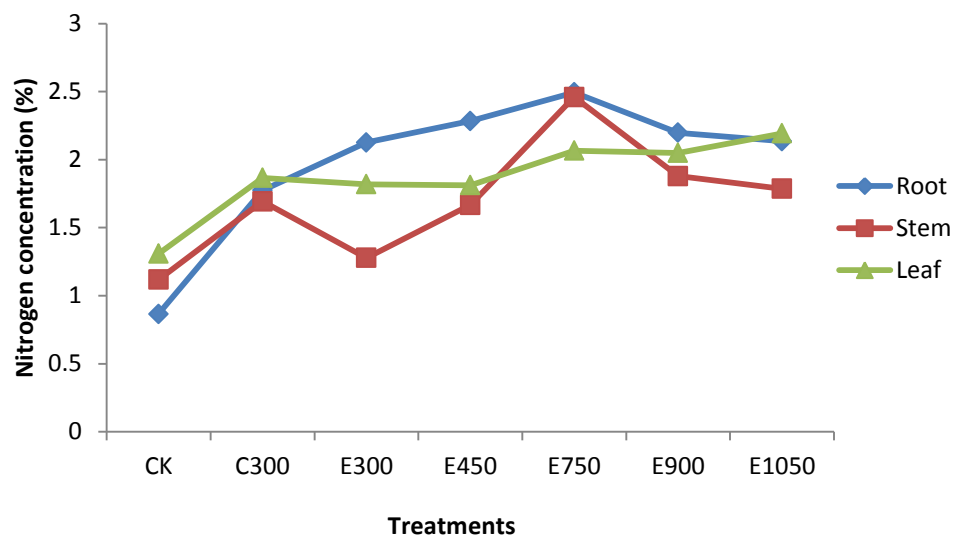




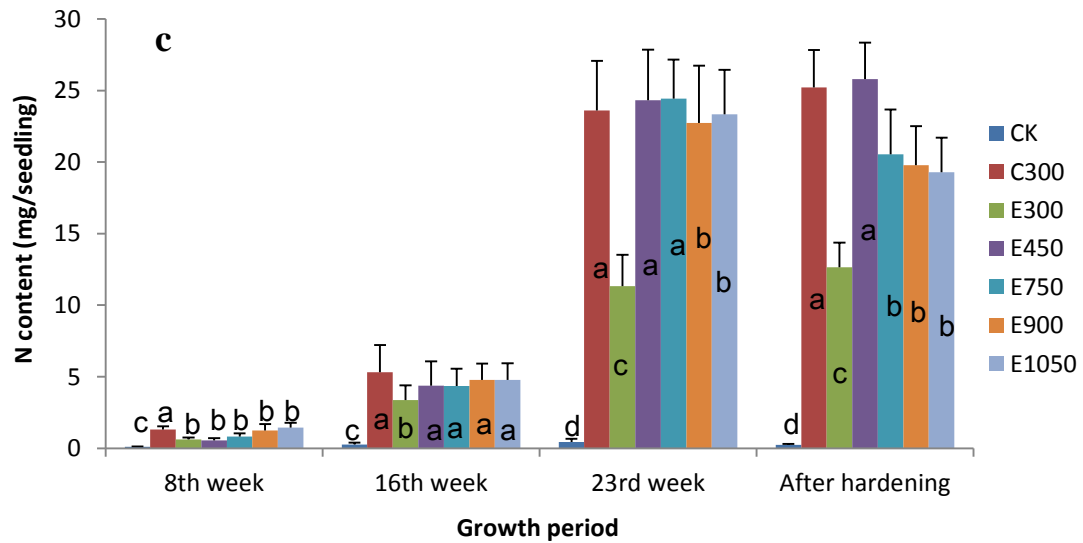
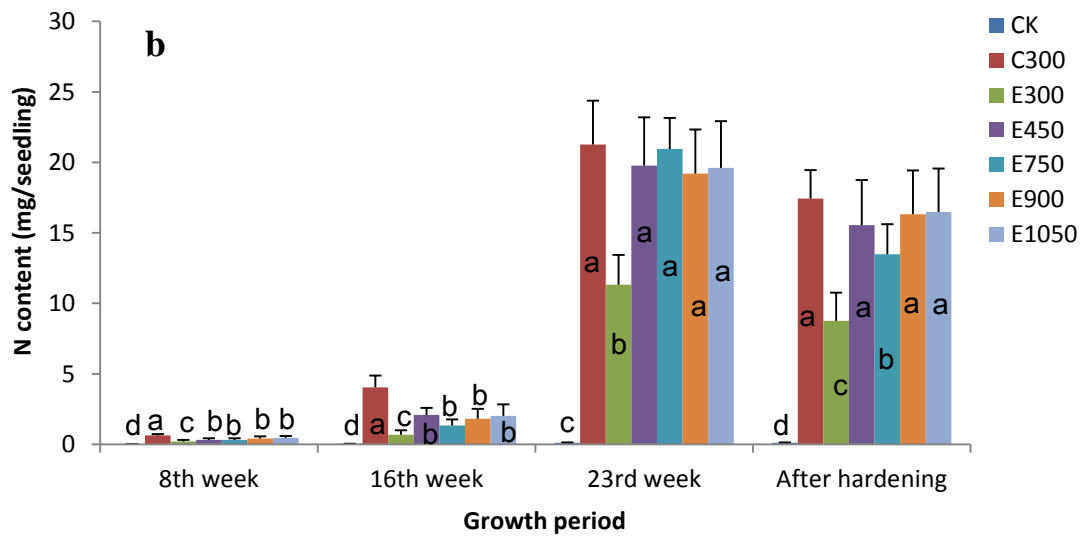
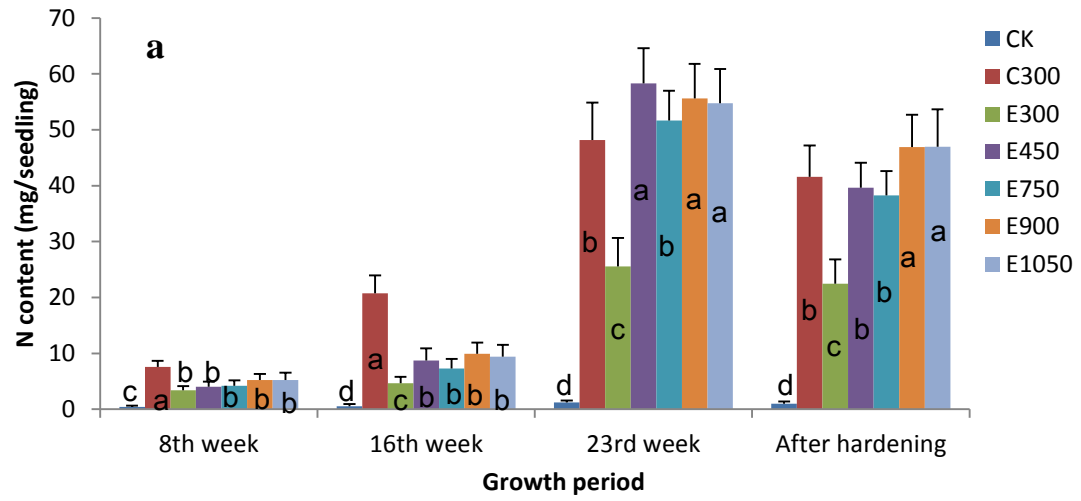
**Figure 3.19.** Spruce morphological data in 2010. **a.** Stem height under different treatments over the entire growth period; **b.** Root collar diameter (RCD) under different treatments over the entire growth period; and **c.** Height:RCD ratio under different treatments over the entire growth period. \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*

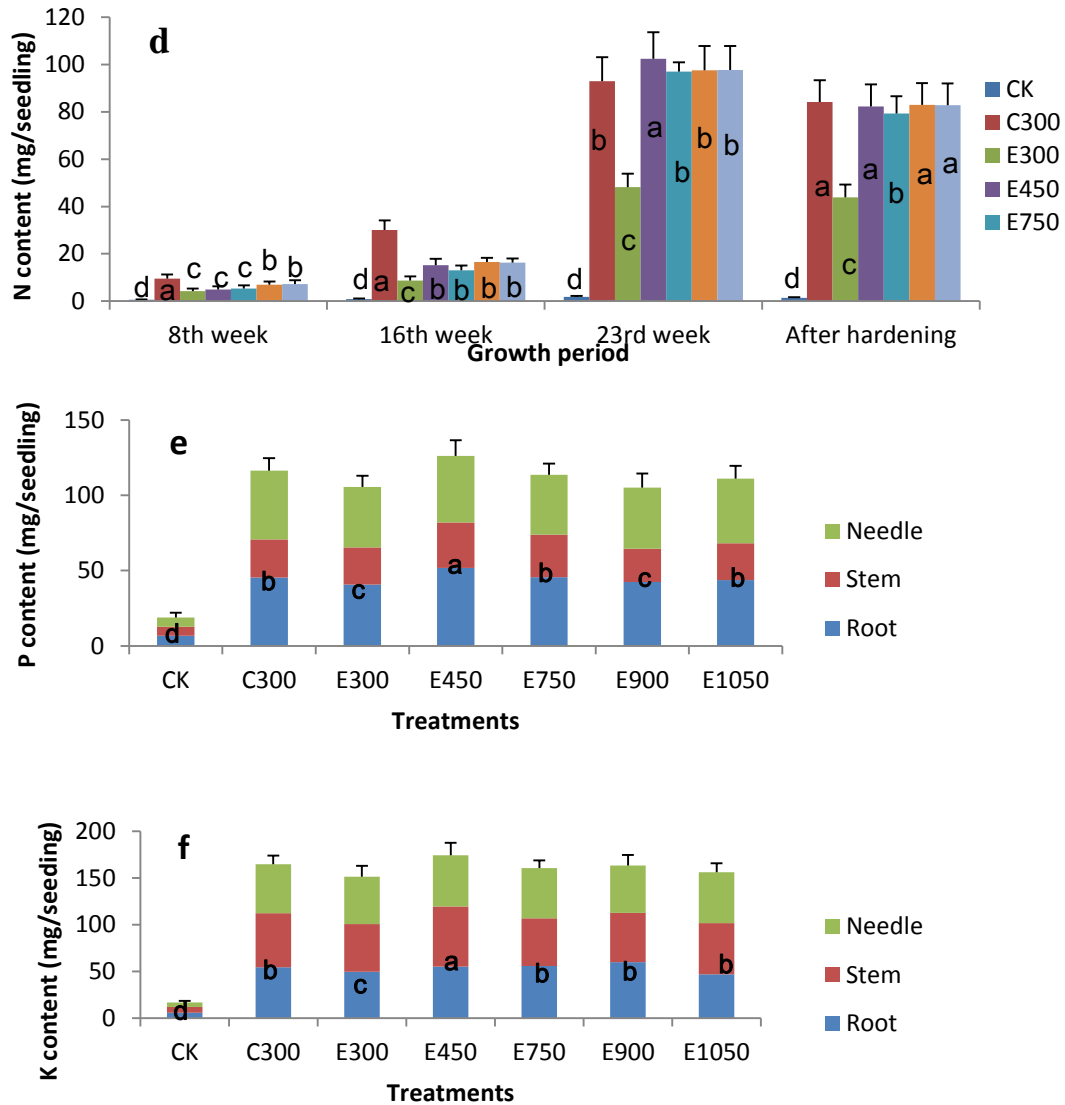


**Figure 3.20.** Spruce component dry mass under different treatments over the entire growth period in 2010. \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Mean separation was based on the total biomass per seedling. Error bars are standard deviations of the means.*

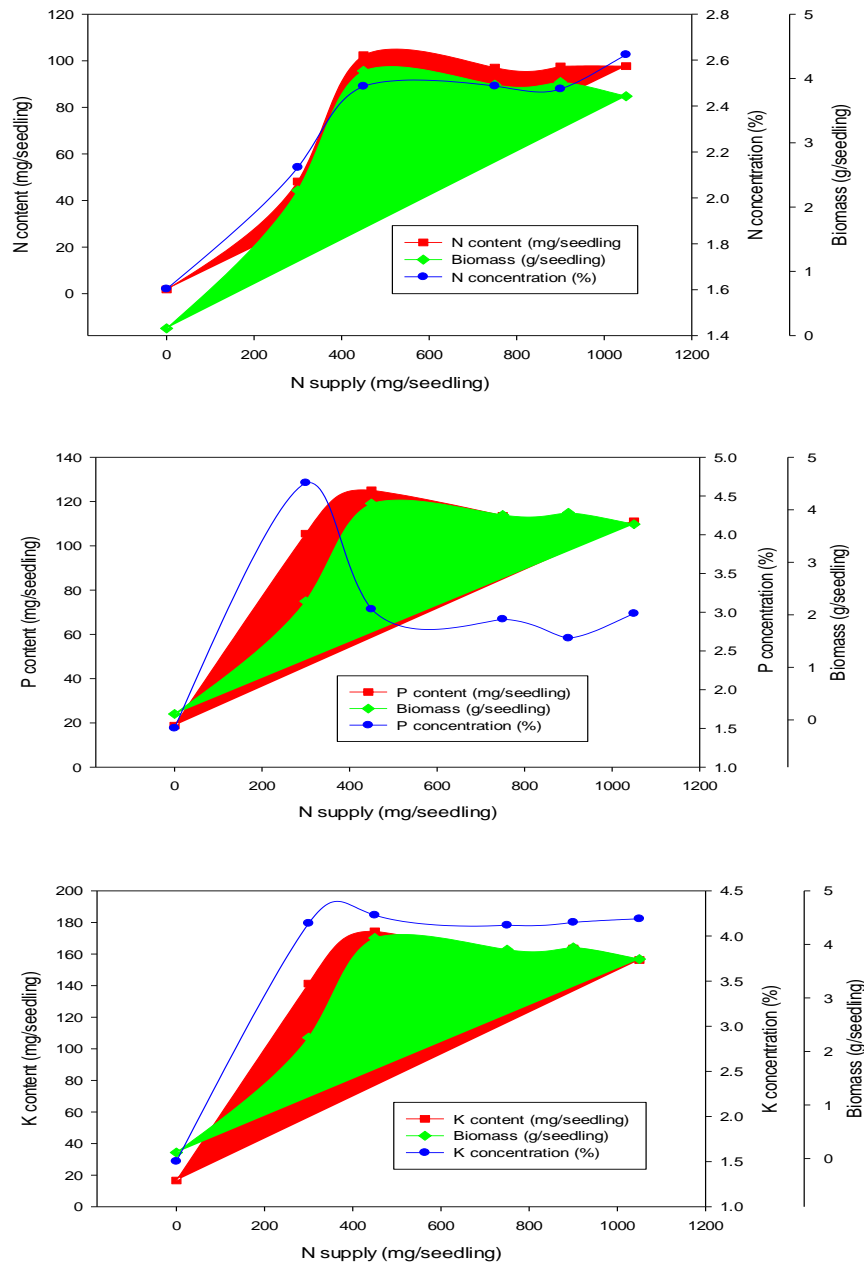


**Figure 3.21.** Spruce N concentration under different treatments in the last collection (23<sup>rd</sup> week) in 2010.

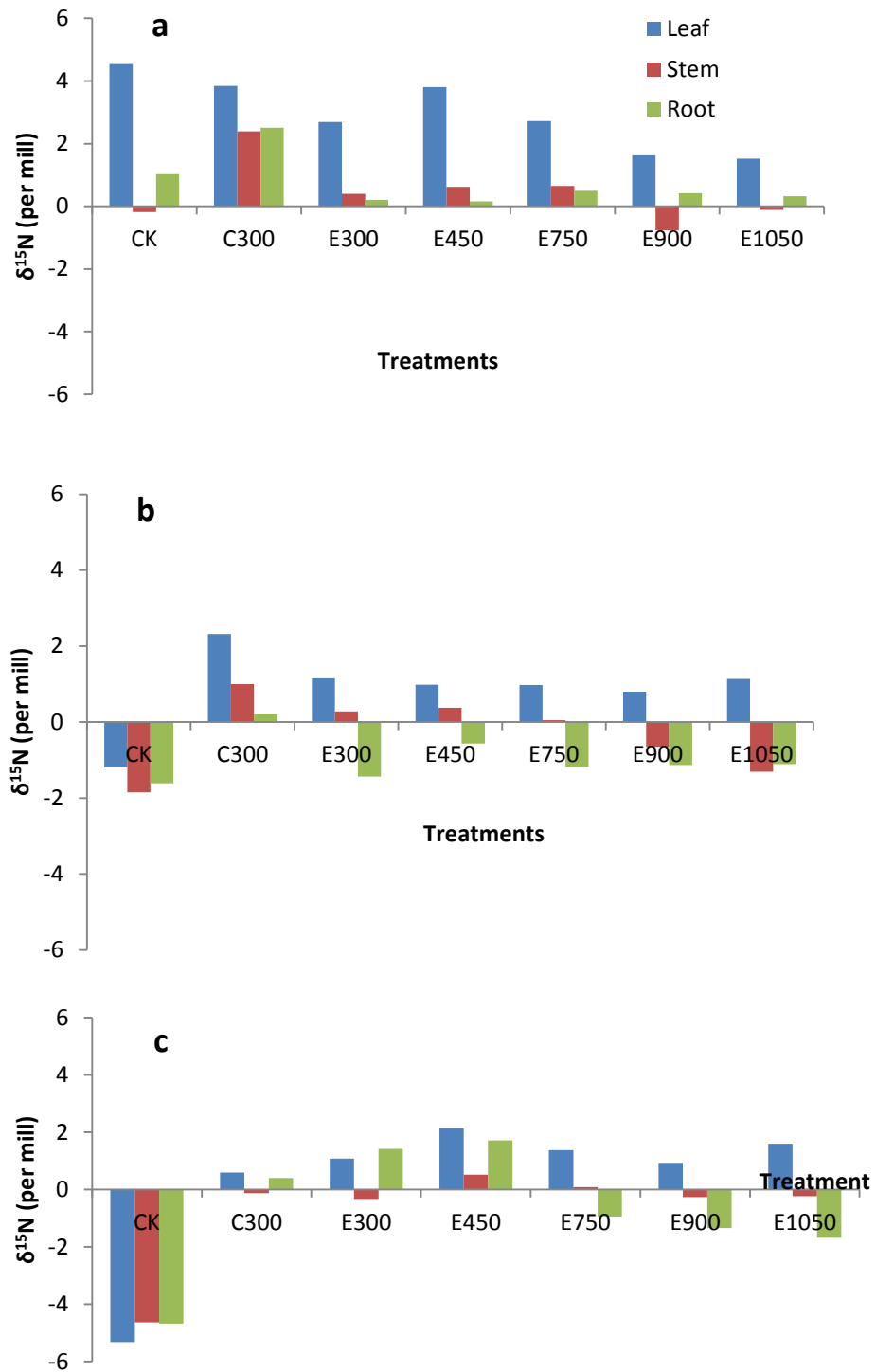




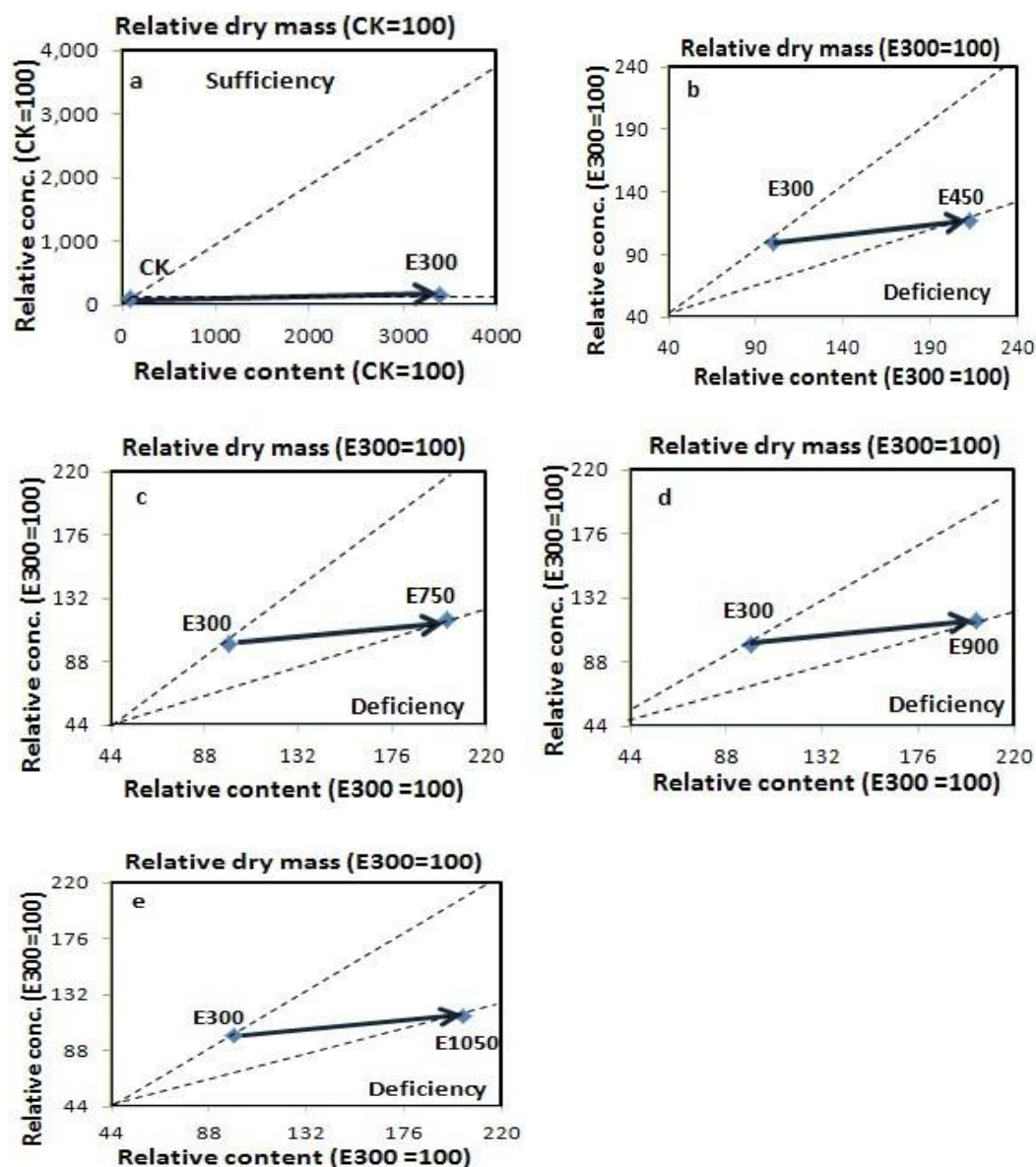
**Figure 3.22.** Components nitrogen, phosphorus and potassium content of spruce seedlings under different treatments over the entire growth period in 2010. **a.** Needle N content; **b.** Stem N content; **c.** Root N content; **d.** Total N content; **e.** Total P content in different parts of the seedling; and **f.** Total K content in different parts of the seedling. \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of means.*



**Figure 3.23.** Relationships among nutrient supply, plant growth and tissue nutrient content and concentration of spruce at week23 in 2010. Treatments used were CK, E300, E450, E750, E900 and E1050 representing N supply rates of 0, 300, 450, 750, 900 and 1050 mg plant<sup>-1</sup> season<sup>-1</sup>, respectively, under the modified exponential fertilization model.

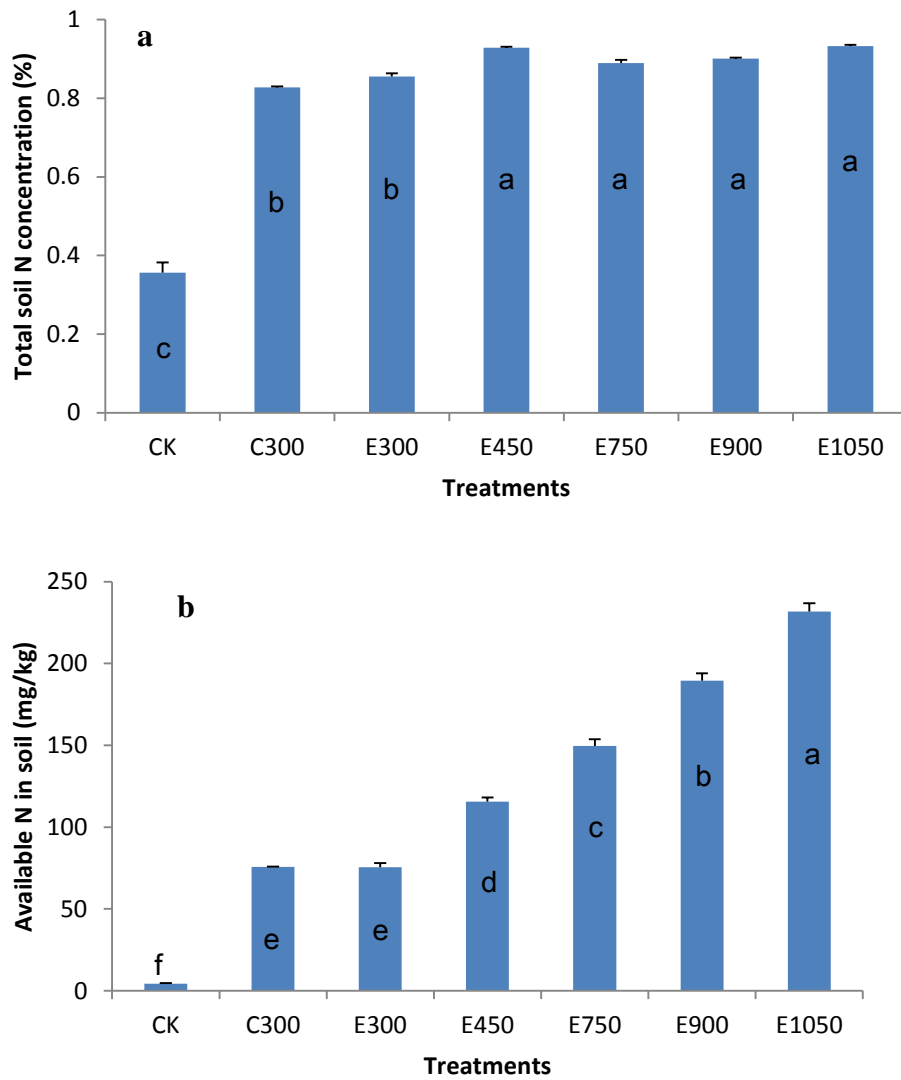


**Figure 3.24.** Component  $\delta^{15}\text{N}$  of spruce seedling under different treatments over entire growth period. **a.** At 8<sup>th</sup> week; **b.** At 16<sup>th</sup> week; and **c.** At 23<sup>rd</sup> week.



**Figure 3.25.** Vector monogram of relative change in plant dry mass, N content, and concentration in white spruce fertilized at different rates. Corresponding value at each point indicates seasonal dose applied (mg N/seedling; CK represents unfertilized or the control treatment). The type of nutritional response induced by enrichment is characterized by shift (or vector) direction and magnitude, described in Figure 3.2.





**Figure 3.26.** Nitrogen information in styroblocks in 2010. **a.** Total soil N concentration in the growth media in styroblocks planted to spruce in the last collection; and **b.** Available N in the growth media in styroblocks planted to spruce in the last collection. \* *Treatments with the same letter in the same sampling are not significantly different ( $p>0.05$ ). Error bars are standard deviations of the means.*

## **Chapter 4. Synthesis of Results and Recommendations for Future Research**

### **1. Synthesis**

Low nutrient availability in peat-mineral mix material used as a cover soil in oil sands reclamation is a major concern. Broadcast fertilization is a common way to overcome the problem; however, this method will also benefit competing vegetation and thus affect early tree establishment. Aspen, jack pine and white spruce are most commonly planted species for oil sands reclamation. The success of planting such species for oil sands reclamation can be affected by intense weed competition (Crow, 1988) and poor soil conditions (Andersen et al., 1989). Variable performance of planted seedlings in oil sands reclamation suggests that additional research is needed to improve the quality of nursery-grown seedlings for field planting. One possibility is to produce high quality seedlings that are well conditioned nutritionally for field planting, but information is lacking for seedling nutritional targets for these species.

This thesis research evaluated the suitability of a proposed modified exponential nutrient loading model (Figure 1.1) as a means to improve post-planting performance for aspen, jack pine and white spruce seedlings.

Results presented in Chapters 2 and 3 show that the modified exponential model was more effective for loading nutrients into aspen, jack pine and white

spruce seedlings than the simple exponential model. The modified model increases the initial fertilizer addition to facilitate nutrient supply in the early growth stage (that period is called a compensation period) and reduces the rate at the last application to avoid over-fertilization that may induce potential nutritional imbalance near the end of the production period (Timmer et al., 1991). The compensation period overcomes the problems of low nutrient supply in the growth medium itself and the initially poorly developed root system of seedlings.

Similar to results found in other conifer production systems (Imo and Timmer, 1992; Malik and Timmer, 1996; Birge et al., 2006), exponential fertilization increased nutrient storage in aspen, jack pine and white spruce seedlings in this study. Close correspondence of experimental data (Figures 3.7, 3.15 and 3.23) with trends in the dose response model (Figure 1.1) demonstrated suitability of the exponential nutrient loading model for application in nursery hardwood and softwood seedling production. Nutrient loading maximized growth and nutrient storage at 2 or 3 times that of the conventional rate used by industry for producing aspen, jack pine and white spruce seedlings (Figures 3.7, 3.15 and 3.23). The optimal loading rate was determined to be 240, 500 and 450 mg N plant<sup>-1</sup>season<sup>-1</sup> for aspen, jack pine and white spruce seedlings, respectively (Figures 3.7, 3.15 and 3.23). The highest fertilization rate induced toxicity associated with increased nutrient concentration in plant tissues but reduced growth and nutrient uptake.

This response highlighted the problems associated with over fertilization and the

need to quantify target rates for nursery production of planting stocks.

Results suggest that fertilizer application using the modified exponential model could increase nutrient loading into seedlings. Moreover, applying fertilizer using the modified exponential model could provide N to seedlings in a way that better matches the demand for nutrients and could reduce possible leaching and increase nutrient uptake efficiency. The significantly increased nutrient availability in the growth medium in the nutrient loading treatment also indicated that this method can not only benefit the seedling itself but also increase the nutrient storage in the plug (Figures 3.10, 3.18 and 3.26). Increased nutrient availability in the growth medium could further benefit the seedlings after out-planting. Currently, most container grown seedlings use fertilizers containing N only, which may raise some problems of inadequate P and K to affect plant growth. This study demonstrated that the use of balanced fertilizers can benefit seedlings by loading P and K along with N during the growth period. Overall, this study demonstrated the suitability of the dose response model (Figure 1.1) for quantifying and rationalizing fertility targets for the production of high quality forest tree seedlings for field planting.

## **2. Recommendations for future research**

The modified exponential model can significantly improve seedling growth and nutrient contents, however, the compensation period for these species was not

long enough which affected the early seedling growth and lowered the nutrient content of seedlings. The compensation period used for this experiment was chosen based on a study conducted by Birge et al. (2006). As that study was focused on red oak and white oak, the compensation period may not be suitable for the species studied in this thesis. In addition, we found that jack pine and white spruce seedlings required more time and higher initial nutrient supply for early root growth. Thus, there is a need to extend the compensation periods for these three species (aspen, jack pine and white spruce) to 6, 11 and 12 weeks, respectively, to obtain better nutrient loading results. During the experiment, jack pine seedlings developed long needles that affected their height growth and nutrient uptake. This may have been caused by the long day length in the early part of the growth period in the nursery/greenhouse. Thus, it would be better to germinate the seedlings early and begin the production of jack pine seedlings in early April to be consistent with nursery practices to reduce the potential for jack pine to develop excessively long needles. Many studies have been conducted in the past to show that seedlings relied heavily on internal partitioning to meet new growth demands when outplanting (Malik and Timmer, 1996; McAlister and Timmer, 1998; Salifu and Timmer, 2003). For example, retranslocation explained between 67-80% of the N demand for new growth in red oak and white oak seedlings. Thus, it would be interesting to know if aspen, jack pine and white spruce respond similarly. The labeled stable  $^{15}\text{N}$  isotope can be used to help

explain if the nutrient loading response persists in time. By using the results from Chapter 3 to compare the conventional treatment and exponential treatments, the advantages of using nutrient loaded seedlings was demonstrated. Finally, field testing is needed to evaluate the seedlings' real performance after outplanting in oil sands reclamation areas.

The limitations of using this technology should also be concerned in this project. First, different species will have different features, thus, the modified exponential model should be adjusted to fit each species instead of using only one mathematical model for all the species; Second, the optimal nutrient loading rates are always higher than the conventional rates which could cause an economic concern when we applied this technology in real practices; Last but not least, the process of applying fertilization (currently by using a syringe to inject solution into each cavity) should also be adjusted to a more suitable way for large scale production to save labour and time. Otherwise, it will also cause an economic concern for using this technology in a large industrial scale.

### 3. References

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