分类号　S157.1
密　级　GK

专业代码　095107
学校代码　10298
学　　号　6314208

南京林業大學
NANJING FORESTRY UNIVERSITY

硕士专业学位论文

（全日制）

论文题目/Title ：Seasonal comparison of morphological characteristics of two different plant species Chinese fir and Oak under simulated acid rain in Southern Jiangsu Province of China

作　　者/Student Name:Muhammad Ramzan

学位类别/Degree：M.Sc

专业领域/Major：Forestry

研究方向/Research Area：Ecology

指导教师/Supervisor：Professor Jinchi Zhang

二〇一六年六月

分类号/Classification No.: 专业代码 Major Code: 095107

密级/Security Classification: 学校代码/University Code: 10298

学号/Student No.: 6314208

南京林業大學

NANJING FORESTRY UNIVERSITY

全日制专业学位研究生学位论文

Paper for Full-time Professional Master Degree

论文题目/Title：Seasonal comparison of morphological characteristics of two different plant species Chinese fir and Oak under simulated acid rain in Southern Jiangsu Province of China

作者/Name：Muhammad Ramzan

学位类别/Degree：M.Sc

专业领域/Major：Forestry

指导教师/Supervisor：Professor Jinchi Zhang

年/Year 月/Month

学位论文原创性声明

本人郑重声明：所呈交的学位论文，是本人在导师的指导下进行的研究工作所取得的成果。尽我所知，除文中已经特别注明引用的内容和致谢的地方外，本论文不包含任何其他个人或集体已经发表或撰写过的研究成果。对本文的研究做出重要贡献的个人和集体，均已在文中以明确方式注明并表示感谢。本人完全意识到本声明的法律结果由本人承担。

学位论文作者（本人签名）：　　　　　　　　201年 6月 14日

学位论文出版授权书

本人及导师完全同意《中国博士学位论文全文数据库出版章程》、《中国优秀硕士学位论文全文数据库出版章程》（以下简称“章程”，见www.cnki.net），愿意将本人的学位论文提交“中国学术期刊（光盘版）电子杂志社”在《中国博士学位论文全文数据库》、《中国优秀硕士学位论文全文数据库》中全文发表和以电子、网络形式公开出版，并同意编入CNKI《中国知识资源总库》，在《中国博硕士学位论文评价数据库》中使用和在互联网上传播，同意按“章程”规定享受相关权益。

论文密级：

☑公开 □保密（___ 年___月至___年___月）（**保密的学位论文在解密后应遵守此协议**）

作者签名：＿＿＿＿＿＿＿＿　　　　导师签名：＿＿＿＿＿＿＿＿

2016年6月15日　　　　　　　　　2016年6月15日

稿酬领取通知

学位论文出版后，杂志社向被录用论文作者支付稿酬，稿酬支付标准为博士论文作者一次性获得价值400元人民币的“CNKI网络数据库通用检索阅读卡”和100元人民币的现金稿酬；硕士论文作者一次性获得价值300元人民币的“CNKI网络数据库通用检索阅读卡”和60元人民币现金稿酬。

请作者直接与杂志社联系领取学位论文发表证书和稿酬。联系方式如下：

联系人： 吴老师　　**电话：** 010－62791817（兼传真）、62793176、62701179（兼传真）

通讯地址： 北京 清华大学邮局84-48信箱 采编中心　　**邮编：** 100084

Declaration

The author hereby declares that the work presented in this thesis paper has been carried out and composed by himself as part of a program of two-year studies at the College of Forestry of the Nanjing Forestry University, China. All views and opinions expressed to remain therein the sole responsibility to the author, and it has not been presented in my previous application for a degree.

Candidate...........................

Date..6/13/..2016.......

Copyright

Candidate supervisor Jinchi Zhang Date: 6/13/2016

Seasonal comparison of morphological characteristics of two different plant species Chinese fir and Oak under simulated acid rain in Southern Jiangsu Province of China

by

Muhammad Ramzan

M.Sc. Forestry

A Thesis Submitted in Partial Fulfillment of Requirements for the Degree of

MASTER OF SCIENCE

In

Forestry

Supervised by

Prof. Jinchi Zhang

College of Forestry

Nanjing Forestry University, P.R. China

2016

"It is the time you have spent on your rose that makes your rose so important".

ACKNOWLEDGMENTS

I consider as my foremost duty to acknowledge the omnipresent kindness and love of Almighty ALLAH who made it possible for me to complete the writing of the thesis. I am thankful to Almighty ALLAH the most Gracious, the Merciful who blessed me with good health, talented teachers, affectionate patents, brothers and sisters, sympathetic friends and bestowed me with the potential and ability to contribute a little to the existing scientific knowledge. I offer my thanks from the deepest core of my heart to Holy Prophet Muhammad (PBUM) who is forever a torch of guidance and knowledge for humanity as a whole.

The preparation of the present manuscript is a long term and extremely demanding effort that requires input and cooperation on the part of many different individuals. I am sincerely grateful to all of you who in one way or another have contributed to this work. I would like to extend my gratitude to Asia-Pacific Network for Sustainable Forest Management and Rehabilitation (APFnet) and Nanjing Forestry University for financial support and facilitating my studies in China.

It is quite delectable and to become to avail of this most propitious opportunity to articulate with utmost gratification, my profound and intense sense of indebtedness to my ever affectionate supervisor, Prof. Dr. Jinchi Zhang (Vice President College of Forestry), for his pleasant teaching and introducing me into the fascinating world of Forestry and precision management; for his proficient counseling, valuable suggestions, boundless forbearance, indefatigable help with anything, anywhere, anytime, consummate advice and thought-provoking instructions in piloting this research venture and to reach its present effective culmination. Special thanks for him would always be due.

In China, language was a big problem that was enough able to restrict learning process, but I have been fortune in having help and advice from a number of my friends and seniors. I offer my thanks to all of them and particularly acknowledge my debt to Dr .Qiang Zhang and my friends and Lab mates, Mr. Liu Xin, Mr. Zhou and Ms. Yue Jin. I also express my appreciative feelings for all my close friends in China and Pakistan Dr .Muhammad Faheem, Dr. Khalid and Dr. Sadaf Shabbir for their benevolent encouragement.

At last but not the least, words are lacking to express humble obligations to my family members particularly affectionate father, loving mother and beloved brothers and sisters for their patience, sacrifices, strong determination and offering countless prayer for success during my entire study and enabling me to achieve this destination.

Finally, I apologize for the names left out of my memory; I am not inconsiderate. We should accept the reality that absence of names on paper will not undermine the assistance rendered directly or indirectly. Their support enabled to me complete my research work and this manuscript, as well. I will remain indebted to all of them and they will always have a place in my heart.

Muhammad Ramzan

Table of Contents

List of Figures

摘要

酸雨是指 pH 值小于 5.6 的降雨。由于其所造成的全球性环境问题，这种形式的空气污染已成为当前广泛争议的课题。本研究采用人工模拟酸雨在温室中处理杉木（*Cunninghamia lanceolata*）和欧洲栎（*Quercus robur*）幼苗 11 个月。人工模拟酸雨包括 5 种 H_2SO_4 和 HNO_3 配比，即 S:N 1:0、S:N 5:1、S:N 1:1、S:N 1:5 和 S:N 0:1，以及 3 种 pH 梯度即中性 pH 值（对照）、pH4.5、3.5 和 2.5。试验结果显示，pH 3.5 以下的所有酸雨处理对两种树苗的苗高、胸径、冠径和新枝长度均有显著影响。与杉木相比欧洲栎的生长率在对照组（Ck）最大，而杉木在 S:N 1:1 处理时生长较好。在 pH <3.5 时，欧洲栎可见明显的叶片损伤。总之，就试验的各项指标而言杉木比欧洲栎对酸雨损伤更具耐受性。数据显示人工模拟酸雨对两个树种的新生枝条长度具有促进作用。枝条长度的增加可能是由于 HNO_3 的施用为其提供了氮元素。在另一组试验中，我们利用根系扫描仪（WinRHIZO）分析了杉木的根系特征，包括根的体积、直径、表面积和长度。S:N 5:1 pH 4.5 的酸雨处理具有较好效果。在所有季节中，pH 4.5 的酸雨对杉木根系损伤最严重。即 pH 2.5 在各季节中对根系参数都有抑制作用。

关键词：人工模拟酸雨，生长，季节，处理

Abstract

"Acid rain," or more precisely acid precipitation, is the word use to describe the rainfall that has a pH level less than 5.6. This form of air pollution is currently a subject of great controversy because of its worldwide environmental damages. Chinese fir (*Cunninghamia lanceolata*) and Oak (*Quercus robur)* seedlings were exposed to simulated acid rain in green house for a period of 11 months. Simulated acid rain contained five different ratios of H_2SO_4 and HNO_3, S:N 1:0; S:N 5:1; S:N 1:1; S:N 1:5 and S:N 0:1 with three different pH levels i.e. neutral (control), 4.5, 3.5 and 2.5. The data revealed that all the parameters of two species including seedlings height, DBH, crown diameter and new branches length were significantly affected at pH 3.5 or less than this, for all ratios of acids. Compared with Chinese fir, the rate of growth of Oak was highest under controlled conditions (Ck), while seedlings of Chinese fir performed better after exposure to S:N 1:1. Visual symptoms of leaf injuries were also observed in Oak after exposure to pH lower than 3.5; for all the ratios as compared with Chinese fir. Overall Chinese fir proved to be slightly more tolerant during whole experiment with regard to all experimental parameters than Oak. Data related to new branches length for both species, showed slightly better effects of simulated acid rain. This enhancement in the length of branches depicts the slight effect of nitrogen fertilizer in case of HNO_3 application. In a second set of experiments, root characteristic of Chinese fir including root volume, root diameter, surface area and length were evaluated using root scanner, WinRHIZO; S:N 5:1 at pH 4.5, among all the others ratios of acid showed better results. Overall effect of pH throughout all the seasons on all the parameters showed; 4.5 was better than others; i.e. pH 2.5 had inhibiting effect on all the root parameters during all the seasons.

Key words: Simulated Acid Rain, Growth, Seasons, Treatments,

Introduction

Acid rain is a universal phenomenon that affects plants, marine life and the environment. It is caused due to the emission of sulfuric dioxide and nitrogen oxide which react with light and water molecules in the environment to produce acid (sulfuric and nitric acid) (CBEF, 2016).

Main causing agents of acid precipitation is emission from natural resources (volcanoes, industrial smoke, decaying vegetation and light) or anthropogenic sources (fossils fuels combustions and wildfire (CBEF, 2016).Industrial revolution has resulted in environmental destruction including air pollution that is the major cause of acid rains and posing a threat to the healthy existence of natural and artificial ecosystems (Tripathi and Gautam, 2007).Large quantities of sulfuric oxides and nitrogen oxides are emitted into atmosphere from chimneys of industrial plant and other industrial sources causing profound deterioration of urban air quality resulting from urbanization and economic growth associated with an increase in energy demands (Kabir et al., 2012).

1.1 Mechanism of Acid Rain

Sulfuric dioxide (SO_2) reacts with oxygen in the atmosphere and converts it into sulphur trioxide (SO_3) then this sulphur trioxide reacts with water molecules in the environment and are converted to sulfuric acid (H_2SO_4).On the other hand, similar mechanism is involved in the conversion of nitrogen oxides, when they react with water molecules in the environment and finally converts the oxide into nitric acid (HNO_3) Fig. 1.1(QLD, 2013).

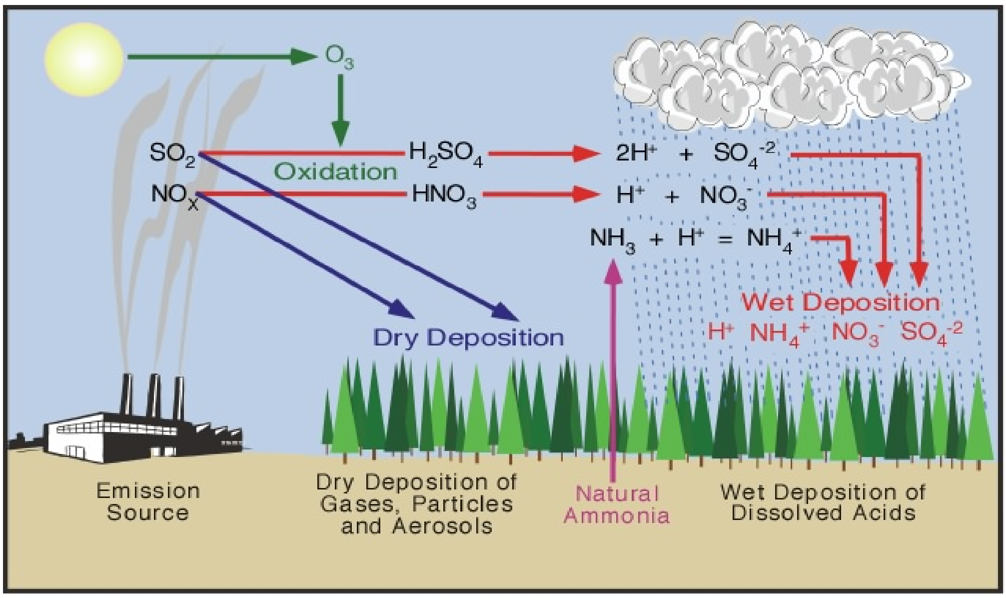

Fig. 1.1 Mechanism and formation of acid rain in the environment

pH of non-polluted precipitation generally in forested regions ranges from 5.0 to 5.6 (due to presence of carbonic acid formed by dissolution of CO_2 into water molecules (Troiano et al., 1982). Acidity of a substance is determined by the value of pH of that substance. Concentration of hydrogen ions (H^+) in a substance is called its pH and it is measured on a scale of 0.0 to 14.0; values lower than 7.0 are acidic, and more than 7.0 are alkaline and 7.0 is neutral (Chesapeake Bay Ecological Foundation, 2016). The United States' national average pH of rain is between 5.6 and 6.2, and any rain that has a pH lower than 5.6 is considered acid rain (NADP, 2012).

The foremost components of acid rain are sulfuric acid and nitric acid derivative mainly produced by incineration of fossil fuels (Troiano et al., 1982; Wagh, et al., 2006). Fossil fuels account for about 80% of energy consumption in Asia (Balasubramanian et al., 2007). The main reason is its sufficient and easy recoverability, coal is becoming the main fuel of choice to fulfill the ever increasing energy demands in developing parts of the world such as to manage the electricity deficit in Pakistan as well as in India and China. Day by day increase in coal incineration will also boost up the present increase of greenhouse gases and oxides of sulphur and nitrogen, driven by the quick growth of Asian economies, inadequacy of energy use, the dependence on coal as major energy supply and the hasty increase in the number of vehicles (Bhattacharya et al., 2004).

1.2 Factors affecting acid rain

Atmospheric acidic pollution, meteorology, topographic structure, and geographic position are considerable factors influencing the happening of acid rains (Patrinos et al., 1989; Anatolaki and Tsitouridou, 2009). Meteorological factors play an important role in acid rain as well (Singh et al., 2007; Lin et al., 2009). The amount of precipitation is a key factor for acidity due to its skulk processes that influence at rainfall composition. This relationship between precipitation, acidity and amount has also been reported by Prado-Fiedler (1990) and Arti, et al. (2010). However, it does not mean all the kinds of precipitation showed a negative influence on pH, since precipitation does not only comprised of washout of the acid causing ions of $SO_4^{-2}$and NO_3^{-1}, but also of the alkaline compounds which act as neutralization factors. In fact, the acidity of rainfall was higher in the dry season than that of rainy season in Beijing, Chongqing, and Vietnam (Viet et al., 2001; Tang et al., 2005). While an opposite affect was observed in Central Mexico and Guangzhou (Baez et al., 2006; Huang et al., 2009).

Direction and speed of wind before and during precipitation are also imperative parameters, briefly describing rainfall composition as a function of natural (aquatic, erogenous, biological, and volcanic) and anthropogenic (industrialized, traffic, heating, agriculture) factors (Vautz et al., 2003; André et al., 2007). Furthermore, temperature, relative humidity, and atmospheric pressure also have an effect on the diffusion of the precursors of SO_2 and NO_2. Normally, atmospheric pressure had positive effects, whereas temperature and relative humidity had a negative influence on the concentrations of $SO_2$and NO_2 in the environment reported by Romero et al. (1999) and Çelik and İbrahim (2007). Geography can also influence wind direction, wind speed, and the quantity of rainfall at a particular point and the circulation of rainfall across a given site due to the forced uplift of moist air (Grimm and Lynch, 2004).

1.3 Effects of acid rain

Acid rain is capable to modify the pH of water masses, such as lakes, ponds, streams, and soil (NADP, 2012; Chesapeake Bay Ecological Foundation, 2016). Lakes and soil regularly have a basic pH value, behaving as a buffer, or neutralizer, for acidic depositions, but if the quantity of acid rain is very high, the pH of the soil and water may lower to a point negatively affecting the flora and fauna, and it could also lead to higher fish mortality (NADP, 2012; CBEF, 2016). Acid rain also raises the ordinary rate of wear and tear of rocks and some metals, which can escort to devastation of stone buildings and hard structures (USGS, 2016). In particular, acidic fog, affects humans by adjoining to acidic water vapors, which humans can inhale, and can result in respiratory disorders (NADP, 2012). Moreover, acid precipitation can devastate the leaves of plants, such as the needles on the evergreen pine trees in Black Forest, Germany, where all of the trees are barren of needles and the trunks are black due to extreme acid precipitation (USGS, 2016). Acid precipitation also affects crops by altering the chemical properties of the soil, leaching down the soil nutrients, and slowing the rates of processes within the crops. Earlier methodological studies that looked at simulated acid rain's effects on crops revealed that the acid rain may inhibit, slow down, or even pick up root development and overall escalation of the crops (NADP, 2012).

Simulated acid rain can inhibit the growth of pollen tube (Wertheim and Craker, 1987). Acid precipitation can strip the defensive wax from leaves, allowing leaves to scorch and die (Percy and Baker, 1990). Acid precipitation also induces changes in the cellular biochemistry and functioning of the whole plant. The effects of acid rain on plants are various and complex, and include discernible symptoms of damage (chlorosis and necrosis) and indiscernible effects such as abridged photosynthesis, nutrient loss from leaves, tainted water balance, variation of enzyme actions, changes in pollen composition and ultra-structure (Van Huylenbroeck et al., 2000). Acid precipitation can also decrease the pH of soil (Balasubramanian et al., 2007).

1.4 Current status of acid rain in China

Currently China has got 3rd position in ranking of largest acid rain area in the world, only after Europe and North America. At the same time as China's speedy economic development, industrialization, urbanization, and its energy consumption has risen fleetly in the last few decades. Under this situation, massive sulfur dioxide emits into the environment, which has become the major source to a great deal of acid precipitation and caused considerable damage on flora and fauna (Chang, 2012). Acid precipitation emerged as significant environmental problem in China in the late 1970s (Percy and Baker, 1990). It was mentioned in previous studies that the loss of forest ecological remuneration due to acid precipitation exceeded 110 billion Yuan per year only in China (Feng, 2000; Xinmin, et al., 2010). At present, acid precipitation has been reported to cover at least 1/3rd of Chinese territory (Ping et al., 2011).

1.5 Objectives

The objectives of present study were as followed:

- To determine the effects of simulated acid rain on early growth patterns of two different plants species i.e. Chinese fir and Oak by using different acid concentrations and pH.
- To evaluate the potentiality and behavior of both species in simulated acid rain with reference to morphological characteristics.

2 Review of literature

Currently, acid precipitation has become one of the top ten worldwide environmental issues. Acid precipitation is becoming the key cause of slower growth rate, injury, or decline of forests. It causes dramatic effects on forests in south China since the late 1970s and the situation is deteriorating (Xiaoqin and Wangand Fu, 2013).

Effects of SAR (simulated acid rain) at two different varieties of mash crop observed by Muhammad Asif Imran and Meo, (2014) in which they observed minute visual symptoms of foliar injury, poor chlorosis and wilting of some old basal leaves due to the low pH treatment i.e. 3.5. Reduction in plant height is reported by Balasubramanian et al. (2007) and Imran and Meo, (2014). They used different pH values implying H_2SO_4, HNO_3, and their different combinations. Data showed that low pH (3.5) of either sulfuric acid or the mixture of H_2SO_4 and HNO_3 more severely affected all the parameters including number of leaves, shoot: root ratio, water contents of shoot and root. On the other hands data showed comparatively better outcomes for a few parameters like plant height and number of branches; the simulated acid rain of solution of pH 4.5 and 3.5 by using HNO_3 plant growth, the root length was increased in case of SAR of solution of pH 3.5 by using H_2SO_4+HNO_3. Balasubramanian et al. (2007) conducted a study on the response and measurable effects of simulated acid rain at different pH levels (3.5, 4.5, 5.5, 6.5 and 7.0) on *Acacia nilotica* seedlings grown on black and red soils for six months. Their study showed that for every one-unit increase in pH, there was a reciprocal increase in plant height. For the black soil, the addition was more at pH 5.5 (32.0 cm) compared with pH 3.5 (20.8cm). Increased plant height with simulated acid rain at pH 7.0 showed improved potential in producing more number of leaves per plant, i.e. with 1069 leaves compared with pH 5.5 and 4.5 with 577 leaves and pH 3.5, 155.Lower pH has stressful effects of acid rain on vegetative or foliar growth and biomass of plants simulated acid rain, affects number of leaves by causing foliar injury (Mandre and Klyshejko, 1995).

Francisco et al. (2006) reported the cause of lower number of leaves in different plant species including *Spondiasdulcis* Forst. F., *Mimosa R. temisiana* Heringer, Paula and *Gallesia integrifolia* (Spreng).Following plants were exposed to the acid precipitation for 20 min for on a daily basis 10 days. As result of this experiment, necrotic spots on the leaf blade and most of the injuries onset on the epidermis were observed. They identified necrosis as punctual regions, characterized by the wearing down of the epicuticular wax, cell shape alterations, and burst of some epidermis regions. Insignificant and inter veinal necrosis was identified in reaction to the acidic precipitation. Mainly the necrosis began on the adaxial epidermis that showed a blackened look because of the phenolic compounds accumulation. Ramlall et al. (2015) described leaf necrotic spots and chlorosis, while plants were exposed to simulated acid rain under pH of 3.0. Silva et al. (2005) also reported erosion and morphological modification of the epicuticular wax and alterations in the epidermis were detected on the upper and lower leaf surfaces. While Ashenden and Bell (1987) performed an experiment on three different seedlings of winter barley, perennial ryegrass and white clover they grown all these on a range of British soils for 21-24 weeks and exposed to simulated acid precipitation treatments of different pH(5.6, 4.5, 3.5 and 2.5). The leaves of white clover showed leaf lesions after 18 weeks of exposure to pH 2.5 treatments, they did not find any signs of visible injury to other two species. Singh and Agrawal (2004) reported the results of a field experiment conducted to evaluate the acid precipitation of different pH i.e. 5.6 (control), 5.0, 4.5, 4.0 and 3.0 on two wheat species (*Triticum aestivum)*, Malviya 213 and Sonalika. Leaf epidermal cracking was evident in both varieties of *T. aestivum* at pH 3.0 after reach age 75 days. Leaf area decreased significantly at pH 4.0 and 3.0 at both ages of growth in M213 and 75 days' age in Sonalika.

Significant reductions in the number of leaves were observed in a pH range of 4.5, 3.0 in M213 and pH 4.0 and 3.0 in Sonalika to 75 days. Further they observed the variation in the total biomass; reductions in total biomass were significant at pH 3.0 and 45 days and at pH range 4.5–3.0. Kohno (1992) reported total dry weight of plants faced to acid rain at pH 2.0 for 5 and 7 weeks was lower than that of plants exposed to acid rain at pH 3.0 or higher. Leith et al. (1989) observed leaves injuries during severe pH i.e. 2.5 and2.7. Sonia and Khan (1996) described the effect of different pH levels of simulated acid precipitation water in spur leaf senescence. The number of leaves decreased with high acidity due to stress mechanism. Leaf growth was affected by simulated acid rain because it had inhibited transpiring area with little uptake of essential nutrients.

Acid precipitation can also decrease the pH of soil and as acid precipitation of pH 3.5 decreased from 8.5 to 7.8 and from 7.8 to 7.0.Electric Conductivity of the soil were dropped from 2.60-0.77 dS^{m-1} in black soil compared to 1.80-1.48 dS^{m-1} in red soil at pH 7.0. Simulated acid precipitation brought changes in the content of organic matter in both soils at lower pH indicated that mineralization of organic matter had occurred. Existing nitrogen, phosphorus and potassium in black soil dropped in pots that received acid rain of pH 3.5, i.e. from the level of 200, 11.0 and 418 kg ha^{-1} to 131, 6.3 and 257 kg ha^{-1}. In red soil, the values were from 178, 9.0 and 386 kg ha^{-1} to 141, 6.8 and 252 kg ha^{-1}, respectively (Balasubramanian et al., 2007). Furthermore, Kohno (1992) also observed initial pH of the cultivated soil prior to fertilization was about 6.0 and dropped to 5.4 after fertilizer application. After continuous 3 weeks of acid precipitation treatments, they did not observe any difference in soil pH among any of the four acid rain treatments. Electric conductivity (EC) of unfertilized soil was 80 $\mu S\ cm^{-1}$ and increased to about 480 $\mu S\ cm^{-1}$. Conductivity of the soil at pH 2.0 after seven weeks' application of simulated acid rain was significantly greater than that at any other pH. Additionally, Ramlall et al. (2015) had also mentioned and observed significant difference in soil pH.

While Tamm and Hallbäcken (1986) reported an acidification of the C horizon in Swedish soils after 57year that might be endorsed to acid deposition. However, they had also mentioned a decrease in the pH of A soil horizon with age of the spruce stands sampled, which they confirmed that it was stronger than the difference between old and new sampling. This indicated that the long term effects of acid rain on the acidification of surface soil horizons may be insignificant in spruce stands.

Simulated acid precipitation decreased the root length within lower pH described by Balasubramanian et al. (2007).They observed root length, was lower at lower pH (11.3 cm at pH 3.5, 17.7 cm at pH 5.5 and 29.0 cm at pH 7.0); while Imran and Meo (2014) reported maximum value 18.47 cm AL- 9 (SAR of pH 3.5 by using combination of $H_2SO_4 + HNO_3$), this addition in root length showed effect of acidity in the rooting medium and subsequent increase in length of root cells further illustrate the resistance/avoidance as adaptive mechanism of this plant to deal with the lofty acidic levels by polluting roots deeper in the soil where acidity level remains reasonably low and established, whereas minimum value 14.78 cm was observed inAL-3 (SAR of pH 3.5 by using H_2SO_4). On the whole it was observed that acids in the collective form are much persuasive as compared to individual application of sulfuric and nitric acids in the form of simulated acid precipitation.

Harcourt and Farrar (1980) reported, root growth was constantly reduced when acid precipitation was enlarged from pH 3.5 to 2.5. According to Singh and Agrawal (2004), the result on roots was being accentuated by contact with sulphite. While according to Balasubramanian et al. (2007), this might be due to the high exchangeable aluminum present in the root zone of red soil which affected the growth and development of roots and, thus, causing root damage.

Balasubramanian et al. (2007) reported that the foliar application of simulated acid precipitation at pH 3.5 notably reduced morphology and growth characteristics, including, plant height, root length, leaf number, total dry matter accumulation, leaf area, single leaf size, specific leaf area, leaf area index, leaf area ratio and crop growth rate. Caporn and Hutchinson (1986) described that simulated acid rain treatments of pH 3.2 and 2.8, exposed at the cotyledons stage, caused lower plant growth by 17 and 15% correspondingly over a time period of 20 d. On the other hand, the similar treatments were exposed at later stages in development when the 'true' leaves were predominant, had no important effect on growth.

According to Lee et al. (1980), even if there will be no visible injuries develop under simulated acid precipitation conditions; reduction in crop growth could be detected. Sonia and Khan (1996) and silmilar results were reported by Gadallah (2000), they mentioned this variation might be due to differences in acidity level. Simulated acid rain of pH 3.5 affecting the function of cell expansion seemed to be more sensitive than the function of cell division and this has caused the reduction in plants total height. While Caporn and Hutchinson (1986) reported visible injuries in plants leaf surface area when pH of simulated acid rain was below then 3.2. Dixon and Kuja (1995) reported more plant height in sugar maple seedlings subjected to restrained levels of simulated acid rain than seedlings receiving normal rain (pH 5.6) and associated this to possible enhancement of photosynthesis after divulgence to acid rain.

Kohno (1992) reported total dry weight of plants supplied with the fertilizer was about two fold of those without fertilizer. Significant growth reduction at pH 2.0 of simulated acid rain but not at pH 3.0 or higher, Cr3, *Ptomeria japonica* without fertilizer treatment and bared to SAR at pH 2.0 did not show any significant growth reduction.

Walna et al. (2000) reported that availability of major nutrients, such as nitrogen, phosphorus and potassium is the main indicator of growth of any crops on soil medium. Available nitrogen was highest at pH 7.0 because of the effects of acidity on soil enzymatic activities that in turn influenced the growth of soil microorganism and their ability in soil mineralization. High quantity of phosphorus available in soil might be due to the influence of pH on the soil micro flora. Magnesium deficiency has been recognized as an imperative cause of forest decline (Haiyan and Stuanes, 2003). While acidic rain can be a major reason of soil acidification and magnesium loss in high altitude sites, it cannot be unspecified that acidic deposition will essentially lead to magnesium deficiencies. Research carried out in a mature maple stand at Turkey Lake, Ontario signaled that losses of calcium and magnesium due to acid rain were not cause for apprehension, since these losses were surplus to requirements (Bardswick et al., 1986) while at lower pH soil micro flora converts unavailable form of phosphorus into its available form. They found in treatments of high acidity, less available phosphorus was observed due to lower level of micro flora and in so emanated in abridged level of conversion of unavailable phosphorus into its available form. Kohno (1992) mentioned sulfur content in *Cr*^{3,}*Ptomeria japonica* increased significantly at pH 2.0 of SAR. This could be due to uptake of sulphur from simulated acid rain containing sulfate as one of most important ion components. Needle K contents notably increased at pH 2.0, but that of roots was found to be decreased. Tissue calcium contents significantly were lowered at pH 2.0. Magnesium content in current-year needles and roots also lowered. Foliar P contents were increased at pH 2.0. While A1 contents in roots notably increased at pH 2.0, which of current-year needles decreased significantly at pH 2.0. Evans et al. (1986) described small incentive in growth of plants at pH 4.0; in comparison to pH 5.6 controls may have been caused by an increase in absorption of Nitrate-N through the leaf surface and soil during acid precipitation treatment. According to Kohno (1992), it is not due to the absorption of N such stimulation in growth might be associated with higher number of root nodules in plants exposed to acid precipitation treatment at pH 4.0.

3. Material and Methods

3.1 Site description

The study was conducted in green house in Xia shu (Jurong) forestry research station of Nanjing forestry University, Jiangsu province of southern China. Geographically research site was located (31° 59’ N, 119°14’ E) in Nanjing, China (Fig. 3.1).

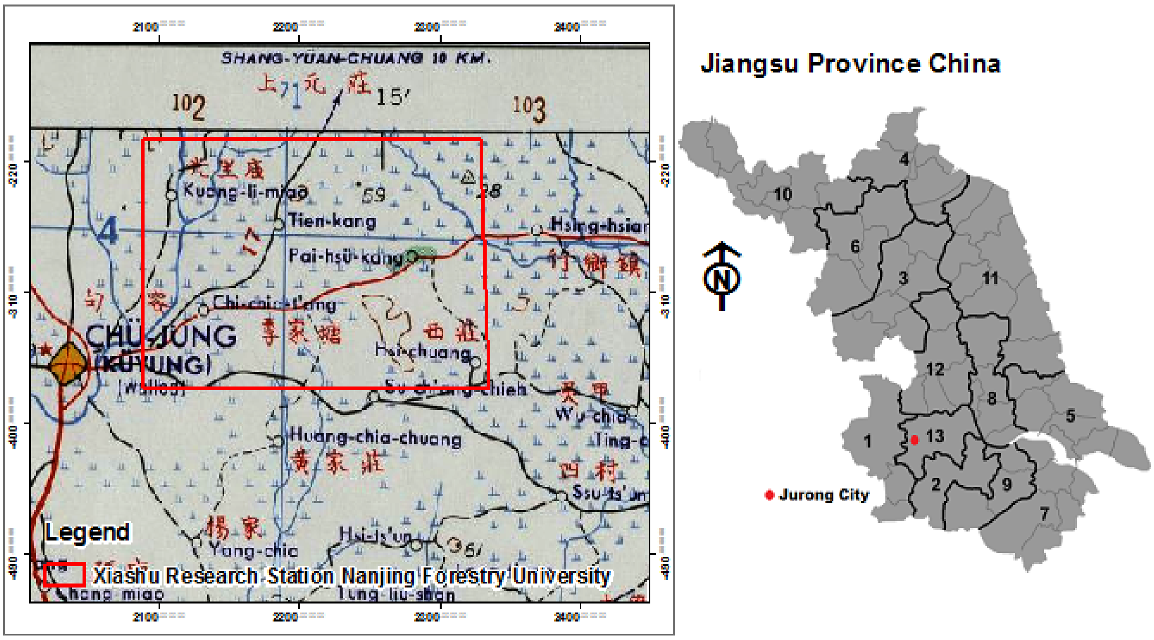

Fig. 3.1: Geographically location of Experimental site

This area belongs to sub-tropical monsoon climate zone, an altitude of447.1 m. The annual mean temperature is 15.3°C, with a monthly mean temperature reaching maximum of 28.2°C in July and a minimum of 1.9°C in January. The rainy season is from June to September, and the average annual precipitation is 1117.29mm, which includes 60% acid rain (Wang et al., 2007; JSSB, 2014). The average frost free period of 223 days, annual average relative humidity near 73% and annual evaporation amount 1309.1 mm. Soil type is yellow brown.

3.2 Plants material

More than 260 seedlings (one year old) of each species Chinese fir and Oak were purchased from chuzhou forest nursery near to Nanjing, Jiangsu China. Both species are much valuable in china and have wide adoptability to grow in acidic conditions.

Chinese fir is a major tree in southern China. Its ability of timber production plays vital role in the national forestry economy (Jiang et al., 2002).

3.3 Experiment Design

All the seedlings were planted in plastic pots in green house. The size of each plastic pot was 25 cm × 20 cm (height × diameter). All seedlings were planted, one seedling per pot, and were grown for almost 11 months from Apr 2015 to Feb 2016 (Fu, 2013). The experiment was set up as a completely randomized block design with six replications and six treatments including one control, each single treatment was assigned with three different levels of pH except control. Every treatment had combination of sulfuric acid and nitric acid ratio S:N 1:0; S:N 5:1; S:N 1:1; S:N 1:5 and S:N 0:1 with pH 4.5, 3.5 and 2.5, respectively (Table 3.2).

Table 3.2: Experiment layout with different concentrations of S:N and pH levels.

pH / SO_4^{2-}/NO_3^-	4.5	3.5	2.5
CK	7.0		
1:0	SAR1	SAR2	SAR3
5:1	SAR4	SAR5	SAR6
1:1	SAR7	SAR8	SAR9
1:5	SAR10	SAR11	SAR12
0:1	SAR13	SAR14	SAR15

Five stock solutions of simulated acid precipitation were made by mixing 0.5 mol L^{-1} H_2SO_4 and 0.5 mol L^{-1} HNO_3 at ratios of 1:0, 5:1, 1:1, 1:5, and 0:1. Basic solutions of control and acid rain treatments were then prepared according to (Wang et al., 2010).

3.4 Application of Simulated Acid Rain

Sulfuric acid and nitric acid were the main ingredients of simulated acid rain because most of the environmental pollution contained sulfur dioxide and nitrogen oxide; these oxides react with water molecules in the atmosphere to produce acid rain. To fulfill the required acid precipitation, plants were exposed to SAR twice a month. First application of SAR was after one month of plantation. Simulated acid rain was prepared according to Wang Guo et al. (2010) from sulfuric acid (H_2SO_4) and nitric acid (HNO_3) added to distilled water to give different pH. The pH of each SAR solution was maintained by using latest digital pH meter. Required pH solutions were made immediately before application.

Application of simulated acid rain (SAR) was applied like procedure described by Evans et al. (1982) that allowed SAR to fall on the foliage as well as on the soil. Size of every plastic pot was 25 cm in height and 20 cm by diameter, by converting it into area total area of single plastic pot was 0.0314m^2 (0.1*0.1*3.14=0.0314m^2). According to the previous data annual rain fall is 1117.29 mm, which contain 60% of acid rain (JSSB, 2014).The rainfall at experimental site was observed to be 93.10 mm month^{-1} (1117.29/12=93.1075). The rate of simulated acid rain applied to every pot was 1775 ml month^{-1} (0.6*93.1075*0.0314*1000=1754).

3.5 Collection of data

Collection of data for morphological characteristic was performed manually. Measurements of both seedlings Chinese fir and Oak plant height and stem diameter were made after every two months during each of the four growing seasons. These measurements were made from a reference line drawn, DBH (cm) of both species took by latest digital DBH meter to reduce the error, and measurements were taken by twice for a single tree. Rest all of the parameters like tree height, new branches length and canopy diameter was measured by measuring tape (cm).

Three seedlings of Chinese fir were harvested in each season to study the morphological characteristic of roots by scanning them using WinRHIZO 2004b (Regent Instrument Inc.), it was installed on a pentium PC (Compaq Deskpro 4000, with 64 Mb RAM, Compaq Computer Corporation, USA) attached to a flatbed scanner (HP ScanJet 4c, Hewlett-Packard Co., USA) with a transparency adapter (HP Transparency Adapter, Hewlett-Packard). Before scanning, roots were washed properly with distilled water to avoid contamination and then placed at room temperature for few minutes to soak. All the fine roots done separated from tape root and placed in a clean plastic tray.

Measurements of data for every season were taken after every two months and divided it into four different seasons; Single data was taken fifteen days after application of simulated acid rain to study the morphological aspects under various levels of simulated acid rain.

Table 3.3 Concentration of different acid rain with pH (Control pH 7.0)

S:N	1:0			5:1			1:1			1:5			0:1		
SAR Type	SAR1	SAR2	SAR3	SAR4	SAR5	SAR6	SAR7	SAR8	SAR9	SAR10	SAR11	SAR12	SAR13	SAR14	SAR15
pH	4.5	3.5	2.5	4.5	3.5	2.5	4.5	3.5	2.5	4.5	3.5	2.5	4.5	3.5	2.5

3.6 Statistical Analysis

Statistical analysis was performed using SPSS 16.0 and $p<0.05$ indicated statistical significance. All figures were derived using Microsoft excel 2007.

4. Results

4.1 Comparison of height and DBH of two species in spring

4.1.1 Comparison of height of two species in spring

The survival percentage of Chinese fir and in all treatment was about 99% during the whole experiment. Readings of summer, spring, autumn and winter were noted on5-08-2015, 8-12-2015, 10-13-2015 and 01-18-2016, respectively. During spring season no significant difference was observed between both species in relation to height with reference to all acid concentrations. While different level of pH behaved slightly different. In case of Chinese fir with reference to height, the maximum value,28.61 cm greater than CK, was observed in treatment 4, had S:N 1:5 and pH 4.5. Minimum was 23.21 cm in treatment 1, had S:N 0:1 and pH 2.5. Compared to with Oak, the maximum value was found within same treatment as it was for Chinese fir 33.65 cm but minimum 22.17 cm, was in treatment 2, had S:N 5:1, and pH 2.5 (Fig. 4.1.1).

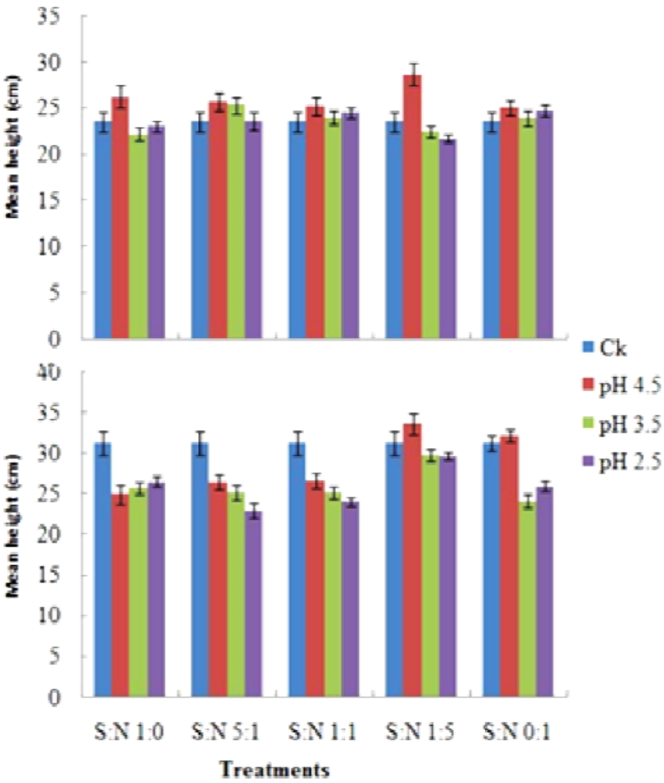

Fig. 4.1.1: Height variations of two species in spring under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.1.2 Comparison of DBH of two species in spring

For DBH, the maximum values of Chinese fir and Oak were recorded as 0.48 and 0.29 cm, respectively, in treatment 4 (S:N 1:5) at pH 4.5,. S:N 1:5 and S:N 1:0 more severely affected both species at pH 2.5 (Fig. 4.1.2).

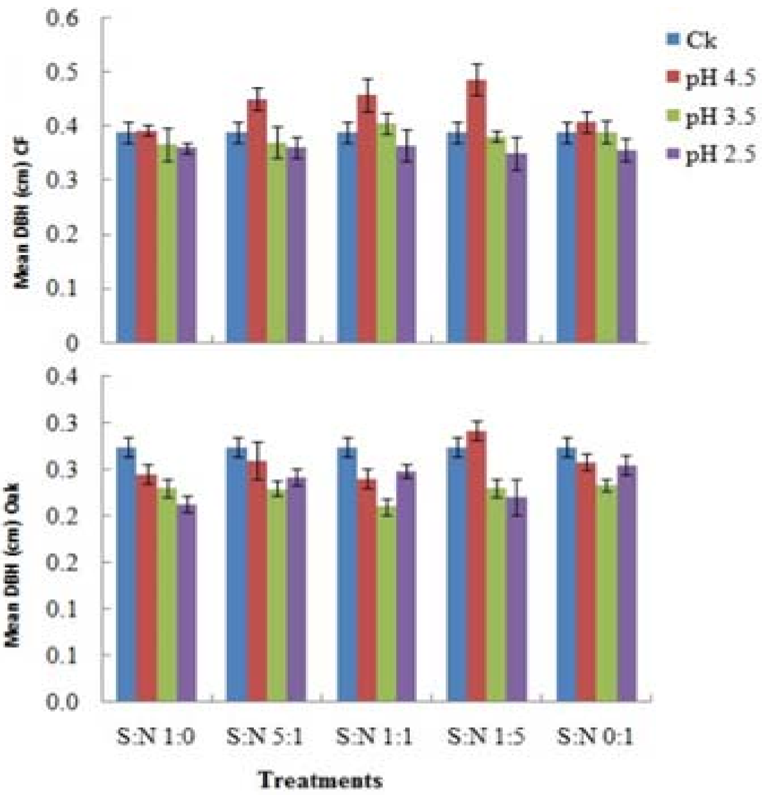

Fig. 4.1.2: DBH variations of two species in spring under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.2 Comparison of Crown diameter and new branches of two species in spring

4.2.1 Comparison of crown diameter of two species in spring

Total growth of crown diameter for Chinese fir and Oak and variations between both species is presented in Fig. 4.2.1

With reference to crown diameter, the Chinese fir performed better in Ck treatment as compared to other acid concentrations and attained maximum growth i.e. 24.34 cm. Although, seedlings were grown in treatment 4 and 3 also showed a bit better performance only at pH 4.5 not lower than this, while minimum value were recorded in treatment 1, S:N 0:1 at2.5 pH. For Oak, highest value for crown diameter was found in treatment 4 (S:N 1:5 at pH 4.5) i.e. 20.12 cm, . Whereas minimum value was recorded in treatment 5 (S:N 5:1 at pH 2.5) i.e.14.21 cm,,.

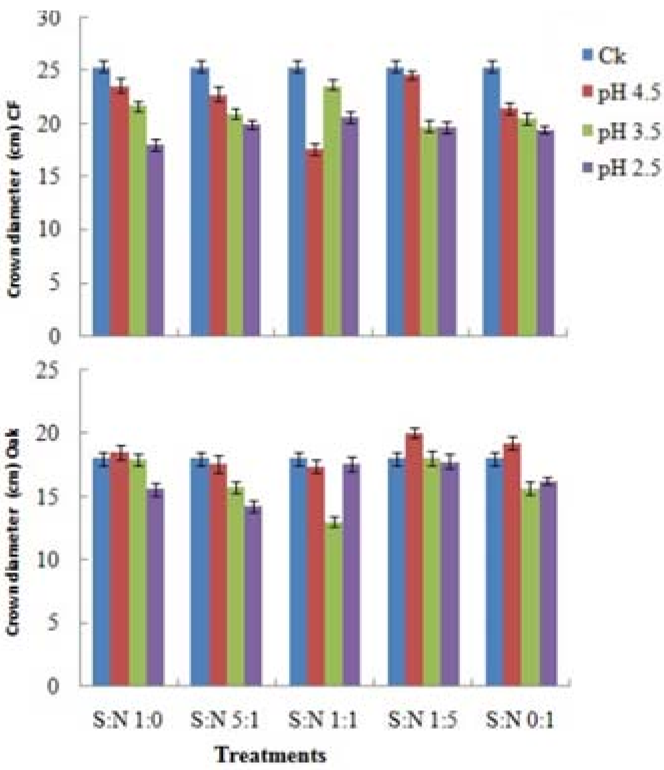

Fig. 4.2.1: Crown diameter variations of two species in spring under different concentrations of SAR

SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively , SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively , SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively , SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

Fig. 4.2.2 Comparison of new branches length of two species in spring

Comparatively, lowest growth rate was observed at pH 2.5 for both of species. This trend showed that pH less than 3.0 damaged more seriously as comparative to pH > 3. On the other hand, growth of new branches length for Chinese fir was highest in treatment 5, S:N 0:1, 11.81 cm at pH 4.5, even this value was greater from the seedlings planted in Ck treatment. Same trend was observed for Oak and recorded highest value in same treatment, S:N 0:1, 7.96 cm at pH 4.5. While minimum values of two species were less than Ck (Fig. 4.2.2).

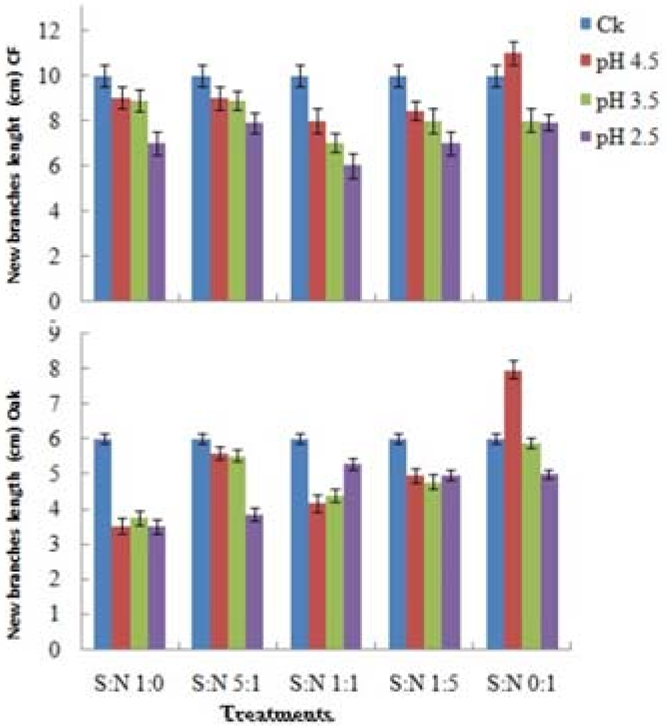

Fig. 4.2.2: New branches length variations of two species in spring under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively , SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively , SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively , SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.3 Comparison of height and DBH of two species in summer

4.3.1 Comparison of height of two species in summer

Seedlings of two species Chinese fir showed gradually positive pattern. Increment in all morphological characteristic was found for both species. Comparison of height of Chinese fir is presented in Fig 4.3.1.

Maximum height of Chinese fir's seedling was observed in treatment 5, S:N 0:1 , 38.91 cm at pH level 4.5. While minimum was 30.23 cm in treatment 1, had S:N 0:1 at pH 2.5. For Oak, maximum value of height obtained in summer was recorded in Ck, 52.16 cm, and minimum was 32.12 cm in treatment 5, had S:N 0:1, at pH level 2.5. As compared to Chinese fir, Oak seedling got bigger value of height but only in Ck. On the other hands, the average height of Chinese fir was not bigger but all the seedlings showed a maintained pattern not zigzag like Oak.

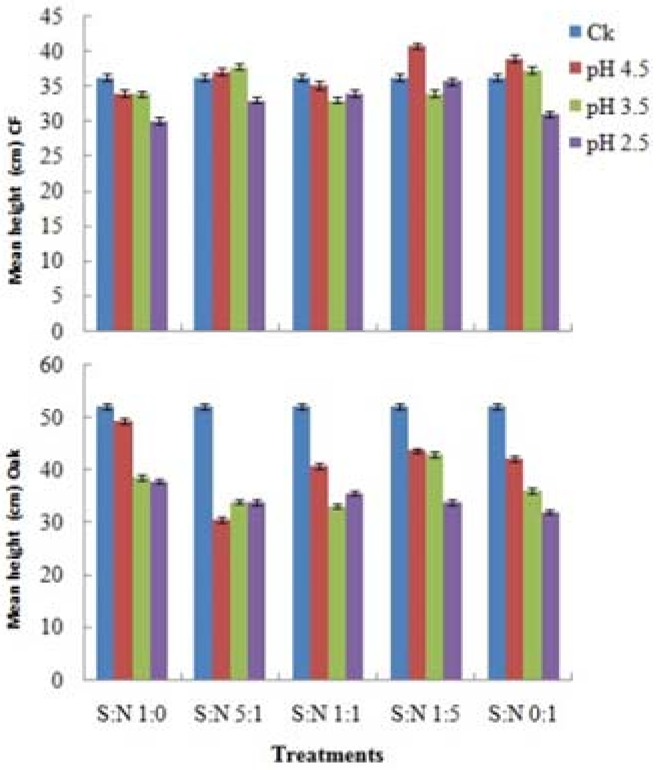

Fig. 4.3.1: Height variations of two species in summer under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.3.2 Comparison of DBH for two species in summer

We observed the increment in DBH of two species for second season (summer). Highest value of DBH was recorded in 0.54 cm in treatment 4, had S:N 1:5, at pH 4.5 in Chinese fir and minimum was .038 cm in treatment 5, had S:N 0:1, at pH 2.5. Maximum DBH value for Oak (0.39 cm) was recorded in treatment 1, had S:N 1:0, at ph 4.5, , while minimum 0.25 cm was recorded in treatment 2, had S:N 5:1, at pH 3.5. Overall the average DBH growth of Chinese fir was greater than oak.

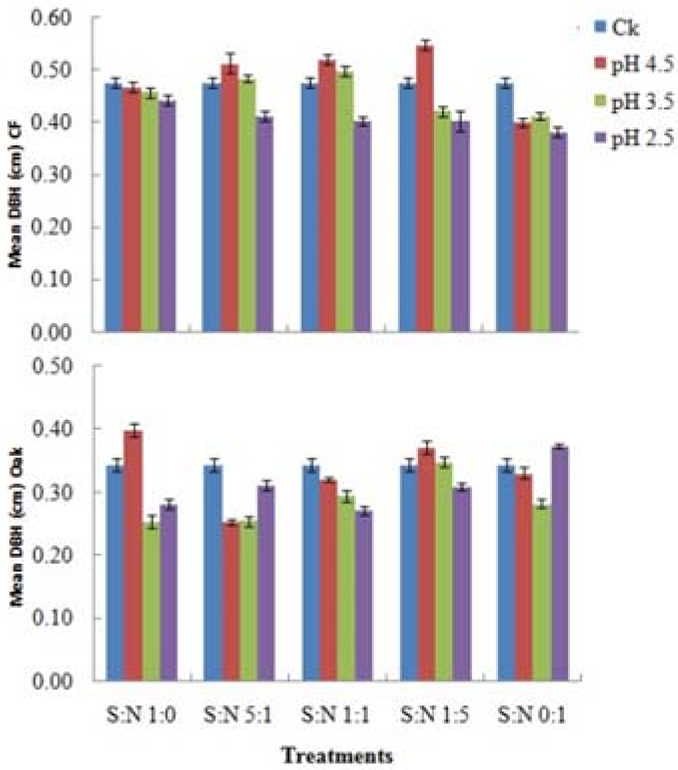

Fig. 4.3.2: DBH variations of two species in summer under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.4 Comparison of Crown diameter and new branches of two species in summer

4.4.1 Comparison of crown diameter for two species in summer

Total growth of crown diameter and new branches length for Chinese fir and Oak and variations between both species is presented in Fig. 4.4.1

With reference to crown diameter the Chinese fir performed better in treatment 5, had S:N 0:1, at pH 3.5, 50.12 cm, as compared to other acid concentrations and attained maximum growth 50.12 cm. Although, seedlings were grown in treatment 4 and 3 also showed a bit better performance only at pH 4.5 not less than this, while minimum value were recorded in treatment 5, S:N 0:1 under 2.5 pH. For Oak, highest value for crown diameter was found in Ck, 33.16 cm. Whereas minimum value i.e. 17.25 cm was recorded in treatment 5, S:N 0:1 under pH 2.5,.Comparatively minimum value of growth of two species was observed in all treatments under pH 2.5. This trend showed that pH less than 3.0 damaged more seriously as comparative to pH > 3.

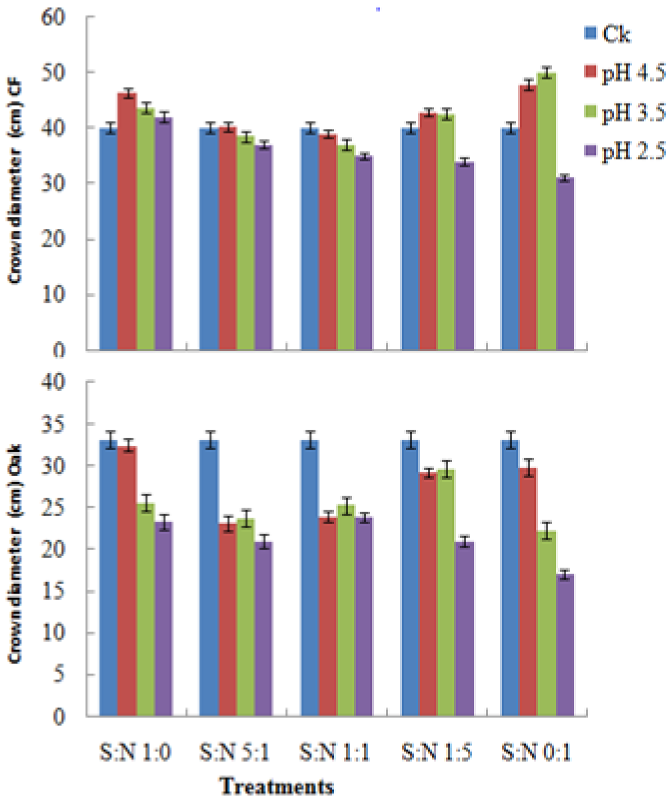

Fig. 4.4.1: Crown diameter variations of two species in summer under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.4.2 Comparison of crown diameter for two species in summer

On the other hand, growth of new branches length for Chinese fir was highest in treatment 5, S:N 0:1, 29.66 cm at pH 4.5,and minimum value was observed in treatment 3, had S:N 1:1, at pH2.5, i.e. 19.23 cm. Similar trend was observed for Oak and recorded highest value in same treatment, S:N 0:1, 16.30 cm at pH 4.5. While minimum values of two species were less than Ck. Overall, average crown diameter and new branches length was bigger in Chinese fir as compared to Oak under all treatments and pH levels (Fig. 4.4.2).

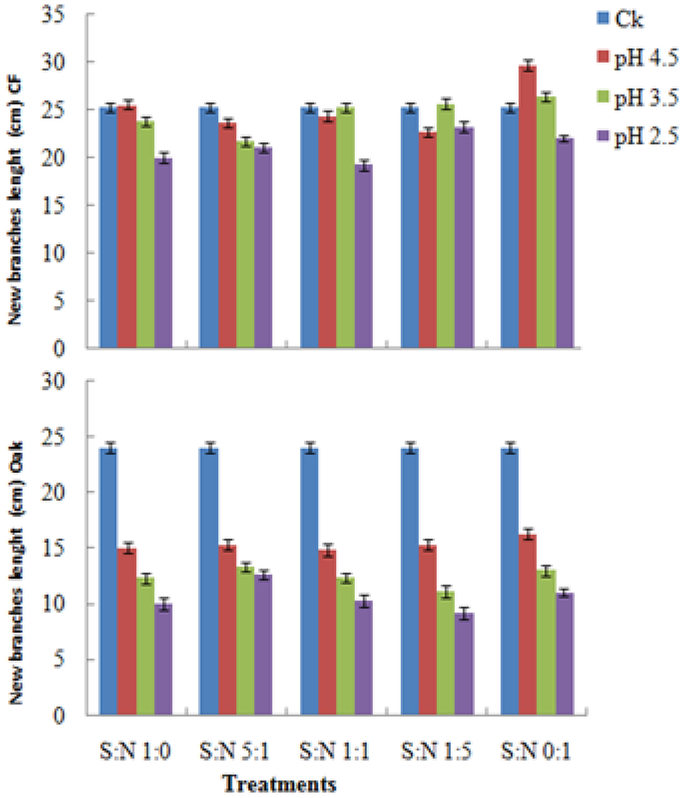

Fig. 4.4.2: New branches variations of two species in summer under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.5 Comparison of height and DBH of two species in autumn

4.5.1. Comparison of height for two species in autumn

Total growth in height of Chinese fir and Oak and variations between both species is presented in (Fig. 4.5.1).Statistically no significant difference was observed in two species at all treatment levels. In case of height growth all the mean values showed almost same height of two species. Chinese fir performed a bit better in all treatments while Oak attained maximum height only in Ck.

The maximum height for Chinese fir in autumn was observed in treatment 4, had S:N 1:5, 52.78 cm at pH 4.5. While minimum value 36.12 cm was recorded in treatment 2, had S:N 5:1,.

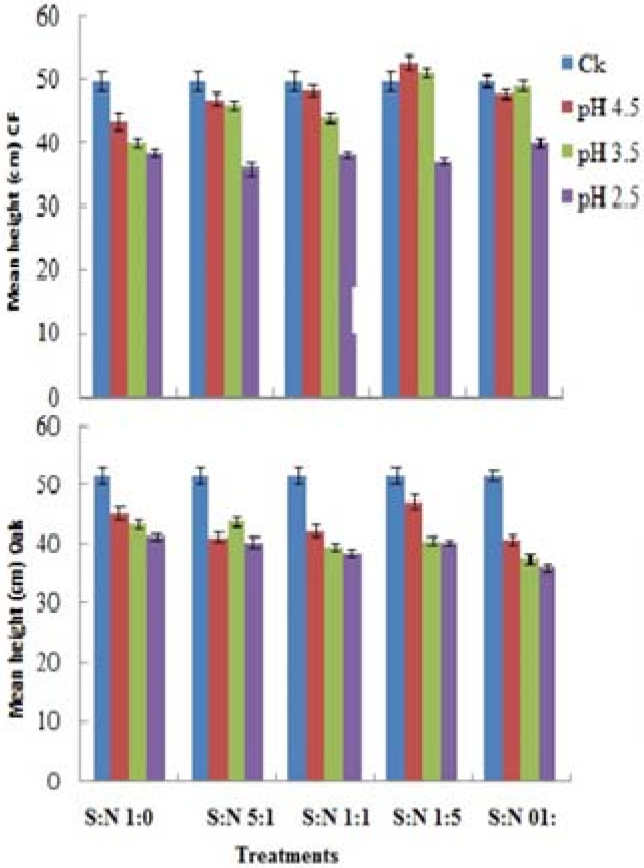

Fig. 4.5.1: Mean height variations of two species in autumn under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.5.2. Comparison of DBH for two species in autumn

For Oak maximum height was observed in Ck, 66.7 cm but treatment 4, had S:N 1:5, also showed better performance at pH 4.5 as compare to others. Seedlings were affected more severely in treatment 1, had S:N 0:1 and minimum value for height was recorded 36.12 cm at pH 2.5. In case of mean DBH, Chinese fir attained maximum growth in treatment 4, had S:N 1:5, 0.69 cm at pH 4.5 and minimum was found in treatment 2, had S:N 5:1, 0.49 cm at pH2.5. Maximum growth for Oak was in treatment 4, had S:N 1:5, 0.41 cm at pH 4.5 and minimum was in treatment 3, had S:N 1:1, 0.30 cm at pH 2.5. Mean max height and mean max DBH of two species were recorded in treatment 4, had S:N 1:5 at 4.5 pH (Fig. 4.5.2).

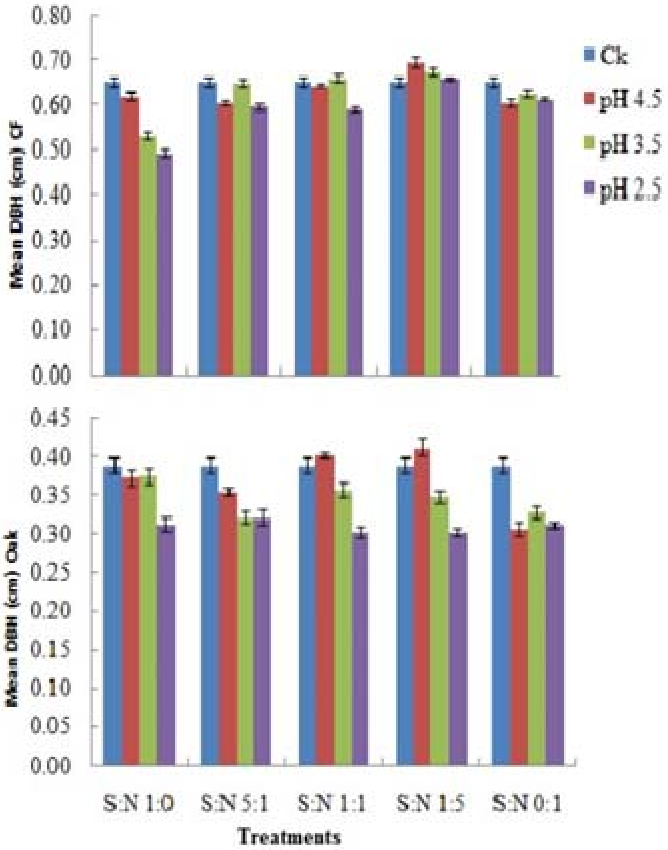

Fig. 4.5.2: Mean DBH variations of two species in autumn under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.6 Comparison of Crown diameter and new branches of two species in autumn

4.6.1. Comparison of crown diameter for two species in autumn

Total growth of crown diameter for Chinese fir and Oak and variations between both species is presented in Fig. 4.6.1.Mean maximum growth of crown diameter for Chinese fir was observed in Ck and treatment 4, had S:N 1:5, 47.58 cm at pH 4.5 and minimum value of crown diameter was recorded in treatment 3 i.e. 35.66 cm, had S:N 1:1, at pH 2.5. While Oak attained highest value of crown diameter in Ck, 33.5 cm and the minimum value was in treatment 5, had S:N 0:1, 16.29 cm at pH 2.5

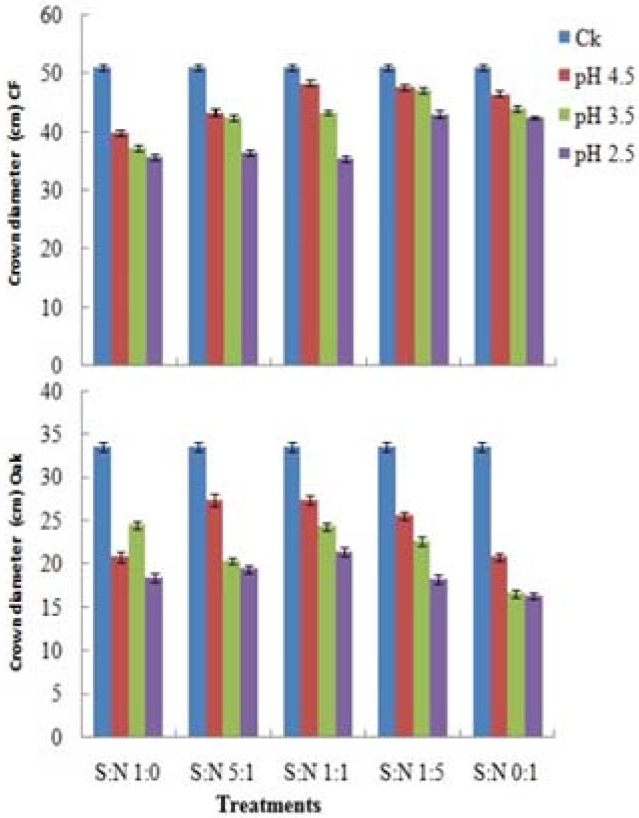

Fig. 4.6.1: Crown diameter variations of two species in autumn under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.6.2. Comparison of new branches length for two species in autumn

Data related to new branches length showed maximum growth of Chinese fir was recorded in treatment 4, had S:N 1:5, 33.66 cm at pH 4.5 and minimum was in treatment 5, had S:N 0:1, 26.54 cm at pH 2.5. For Oak it was 30.21 cm highest in treatment 4, had S:N 1:5 at 4.5 pH and minimum was 22.11 cm in treatment 3, had S:N 1:1 at pH 2.5 (Fig. 4.6.2).

.

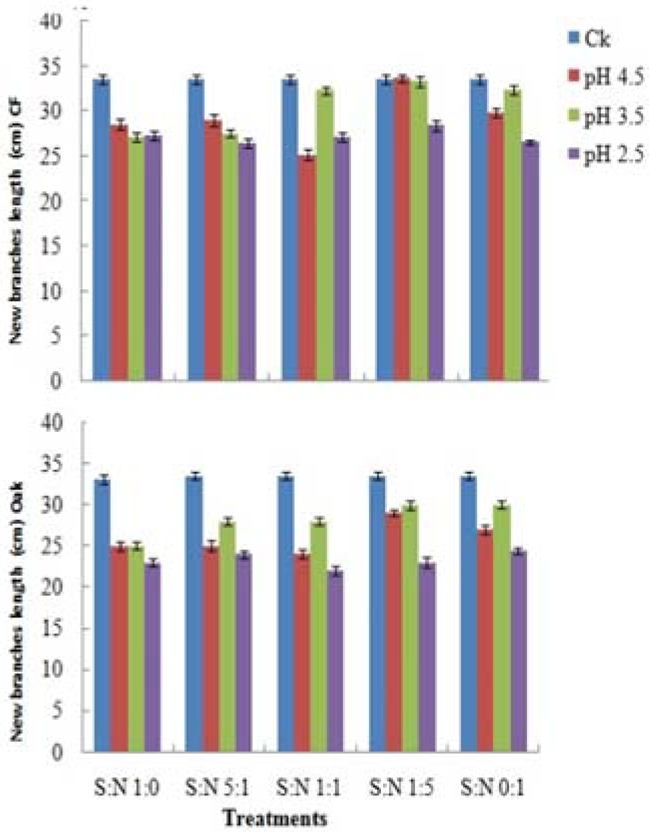

Fig. 4.6.2: New branches variations of two species in autumn under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.7 Comparison of height and DBH of two species in winter

4.7.1. Comparison of height for two species in winter

Total growth of height for Chinese fir and Oak and variations between both species is presented in Fig. 4.7.1.

In winter, we didn’t observe any significant increment difference about height of seedlings between both species as compared to previous all seasons. Height might be inhibited or stunted during whole season, same pattern was observed for Oak seedling with reference to DBH. However, maximum growth of height was observed in treatment 4, had S:N 1:5, 53.33 cm at pH 3.5 and minimum was in treatment 2, had S:N 5:1, 41.32 cm at pH 2.5. In case of Oak, the highest value of obtained height was recorded in treatment 5, had S:N 0:1, 46.6 cm at pH 4.5 and lowest value was found in treatment 3, had S:N 1:1, 33.42 cm at pH 2.5.

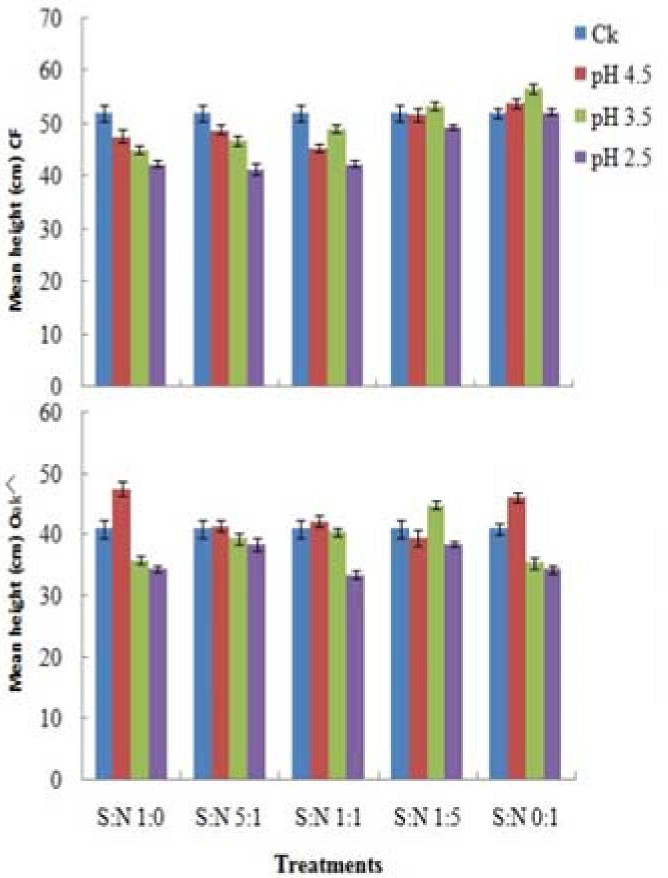

Fig. 4.7.1: Mean height variations of two species in winter under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.7.2. Comparison of DBH for two species in winter

For DBH, the maximum value of Chinese fir observed in treatment 4, S:N 1:5, 0.83 cm at pH 3.5 and minimum was in treatment 1, had S:N 1:0, 0.59 cm at pH2.5 and for Oak, it was maximum in S:N 1:0, 0.41 cm at pH 4.5 while minimum was in treatment 3, had S:N 1:1, 0.31 cm at pH 2.5. In winter, any significant increment difference was not observed about DBH of seedlings between both species as compared to previous all seasons. DBH might be inhibited or stunted during whole season, same pattern was observed for Oak seedling with reference to height (Fig. 4.7.2).

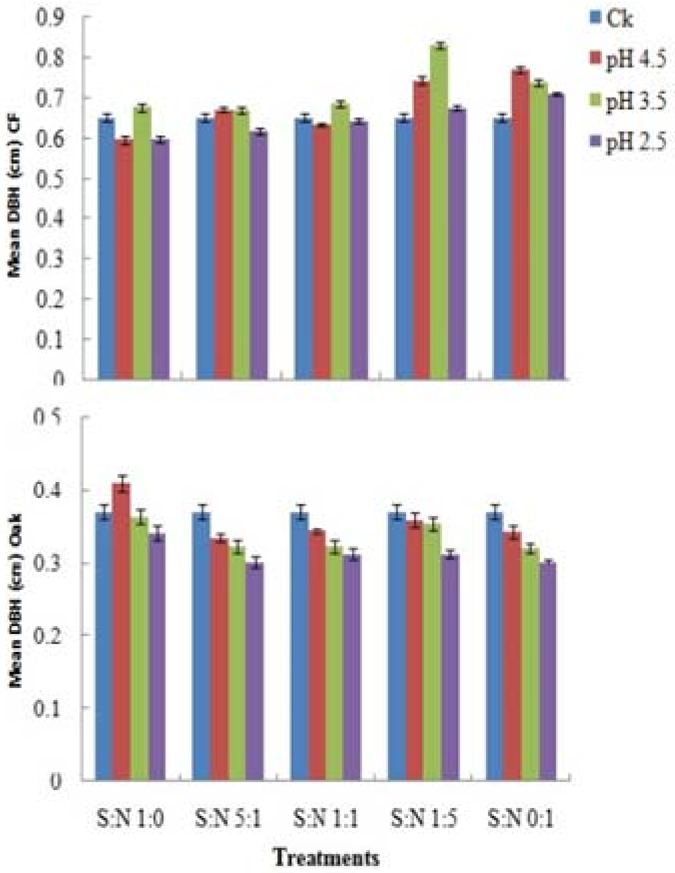

Fig. 4.7.2: Mean DBH variations of two species in winter under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0.

4.8 Comparison of Crown diameter and new branches of two species in winter

4.8.1. Comparison of crown diameter for two species in winter

The total growth of crown diameter for Chinese fir and Oak and variations between both species is presented in Fig.4.8.1.Mean maximum growth of crown diameter for Chinese fir was observed in treatment 4, had S:N 1:5, 51.25 cm at pH 3.5 and minimum value of crown diameter was recorded in treatment 2, had S:N 5:1, 41.33 cm at pH 2.5. While Oak attained highest value of crown diameter in treatment 2, had S:N 5:1, 25.87 cm at pH 3.5 and the minimum value was in treatment 5, had S:N 0:1, 14.87 cm at pH 2.5.

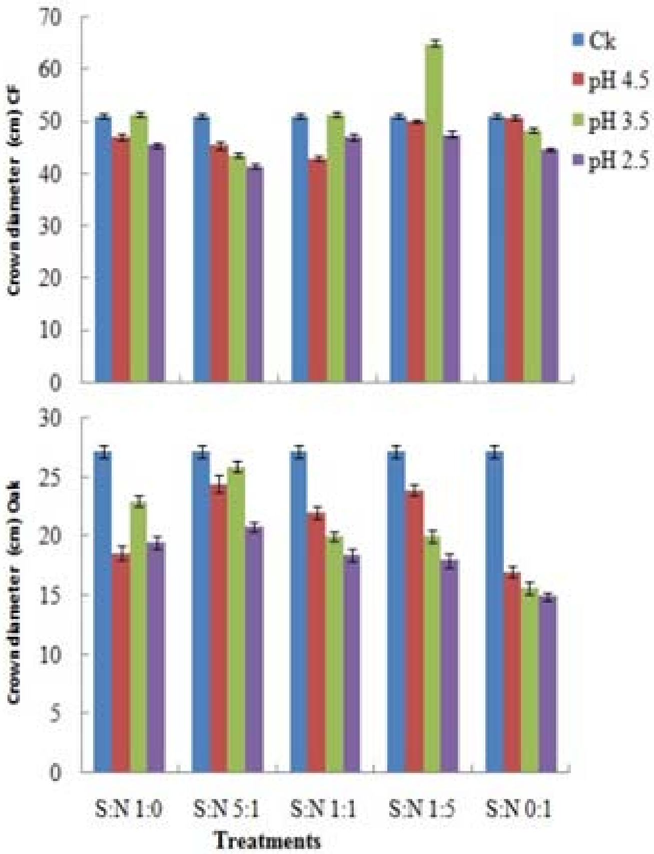

Fig. 4.8.1: Mean crown diameter variations of two species in winter under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

4.8.2. Comparison of new branches length for two species in winter

Data related to new branches length showed maximum growth of Chinese fir was recorded in treatment 5, had S:N 0:5, 23.95 cm at pH 4.5 and minimum was in treatment 4, had S:N 1:5, 14.54 cm at pH 2.5. For Oak, it was 21.21 cm highest in treatment 5, had S:N 0:1 at 4.5 pH and minimum was 13.3 cm in treatment 3, had S:N 1:1 at pH 2.5 (Fig. 4.8.2).

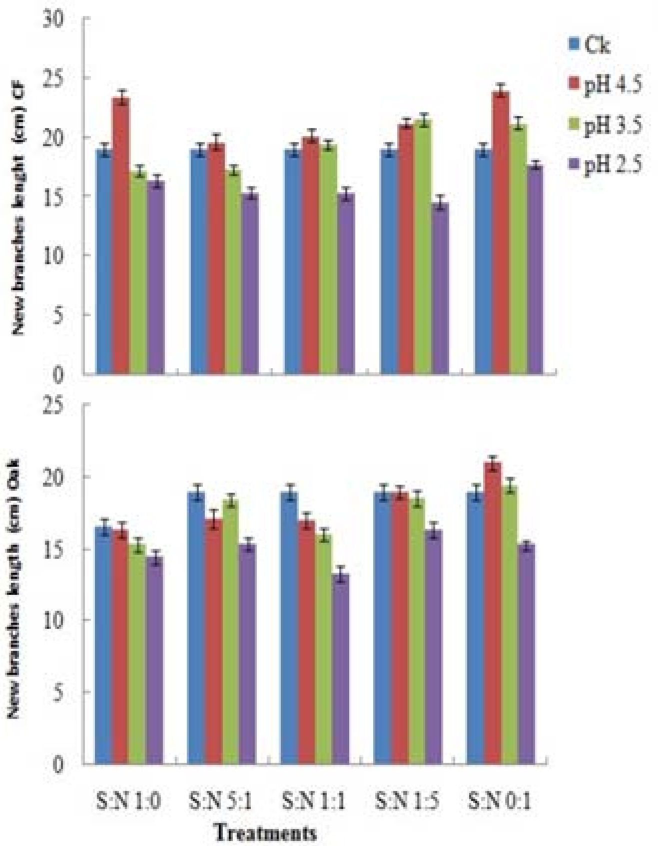

Fig. 4.8.2: New branches length variations of two species in winter under different concentrations of SAR
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

5 Study of root characteristics of Chinese fir by root scanner

5.1 Total growth in root length of Chinese fir for three different seasons

In every season we harvested randomly three different seedlings of Chinese fir from each treatment. Cut their fine roots and scan all of them with WinRHIZO 2004b (Regent Instrument Inc.), and it was installed on a Pentium PC (Compaq Deskpro 4000, with 64 Mb RAM, Compaq Computer Corporation, USA) attached to a flatbed scanner (HP ScanJet 4c, Hewlett-Packard Co., USA) with a transparency adapter (HP Transparency Adapter, Hewlett-Packard) (Regent Instrument Inc.),. Comparison of total root growth in all different treatment is presented in Fig. 5.1.1.

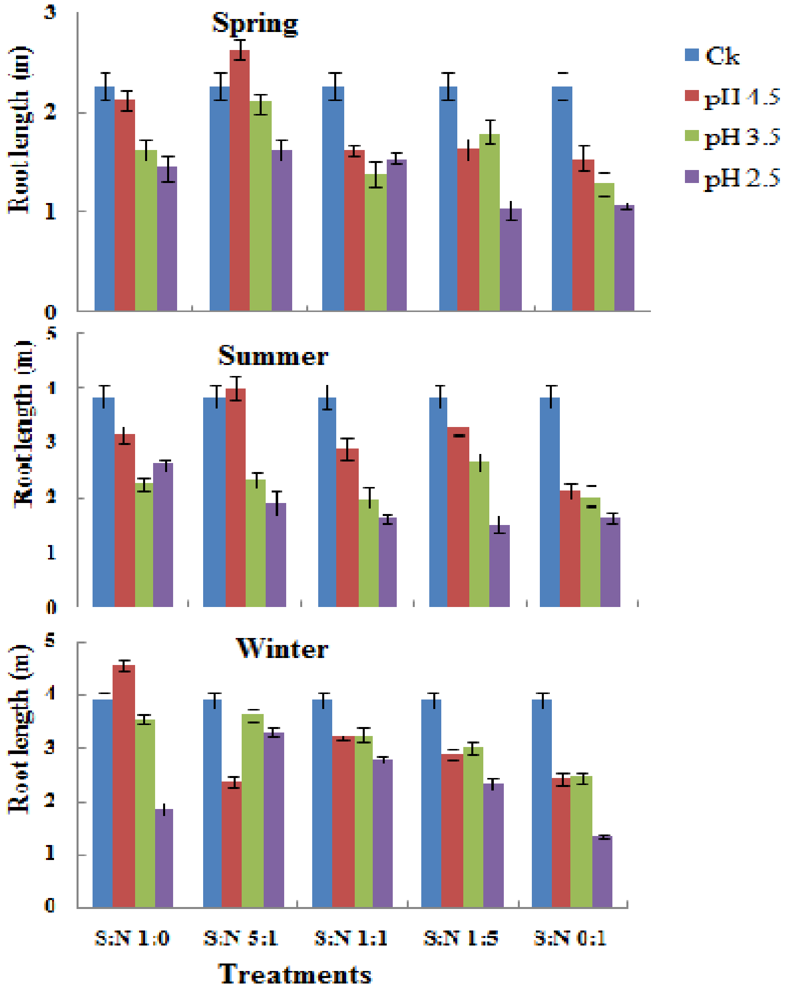

Fig. 5.1.1: Seasonal comparison of total root length growth in all different treatments

SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

In spring the root length for Ck was observed 2.25 m while from different concentration the maximum root length recorded in treatment 2, had S:N 5:1, 2.62 m at pH 4.5 it was higher than Ck and minimum for spring i.e. 1.6 m was found in treatment 5, had S:N 0:1, at pH 2.5. For second season summer Ck obtained 3.38 m, while highest value found in again treatment 2, had S:N 5:1, 3.98 m at pH 4.5 and minimum was in treatment 4, had S:N 1:5, 1.51 m. In autumn there was no such increase was observed in Ck as compared to previous both seasons (3.38 m) but in treatment 1, had S:N 1:0, got the maximum increment it was 4.53 m at pH 4.5 and the minimum value was 1.33 m at pH level 2.5.

5.1.2 Impact of different pH levels at root length of Chinese fir

Fig.5.1.2 presents the effects of pH levels at root length for Chinese fir for three different seasons including spring, summer and autumn.

Mean plot of root length for Chinese fir showed Ck contained pH 7 performed better in all seasons as compared to other 3 different pH levels included 4.5, 3.5 and 2.5. Except Ck, the pH level 4.5 showed better performance in all the concentrations of acid during whole experiment as compared to 3.5 and 2.5, pH level 2.5 performed worse and caused in the reduction of root length for all seasons in all acid concentrations.

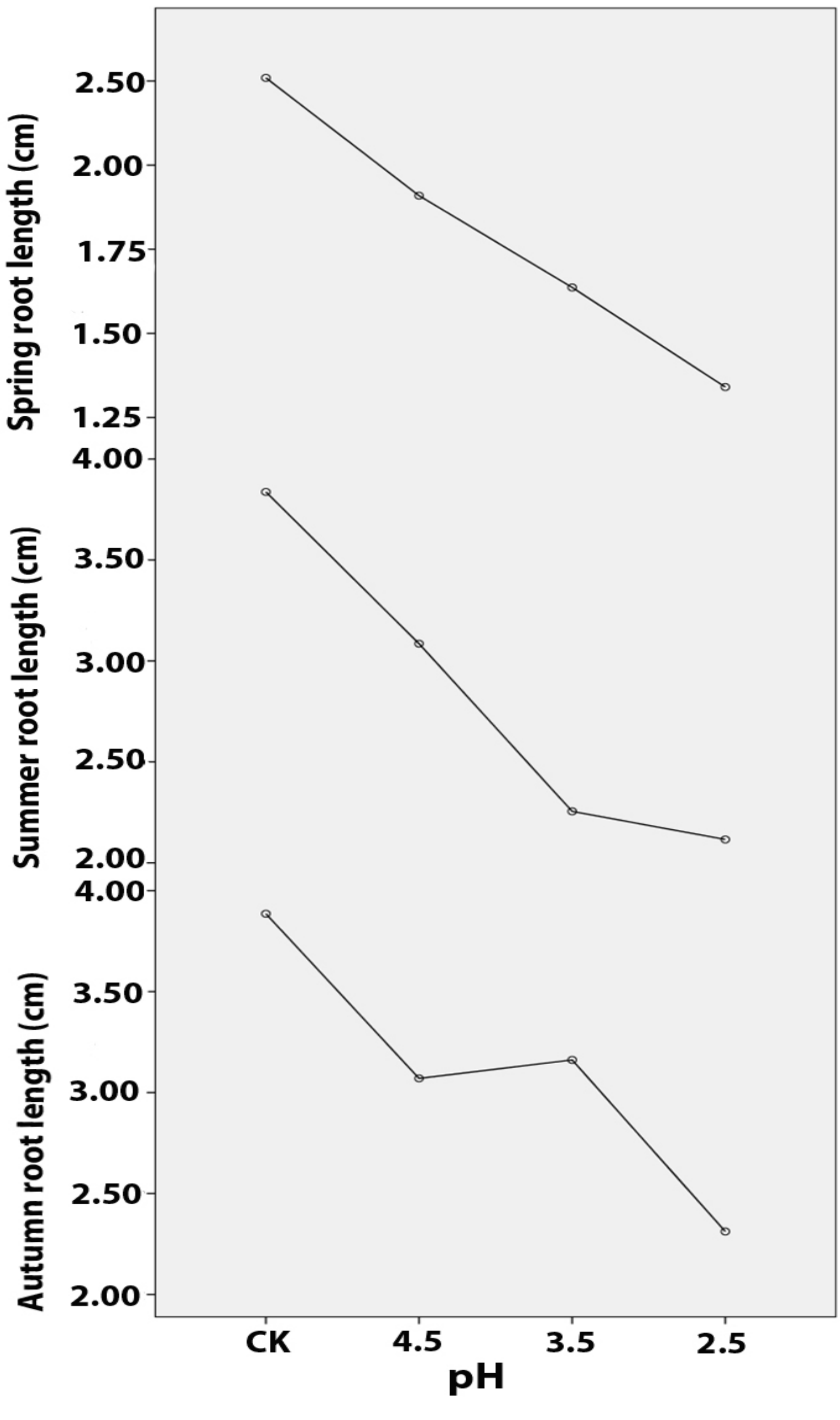

Fig. 5.1.2 Mean plot of root length for pH 4.5, 3.5, 2.5 and Ck contained pH level 7 for three different seasons

5.2 Total growth of root diameter of Chinese fir for three different seasons

In every season we harvested randomly three different seedlings of Chinese fir from each treatment. Cut their fine roots and scan all of them with WinRHIZO 2004b (Regent

Instrument Inc.), and it was installed on a Pentium PC (Compaq Deskpro 4000, with 64 Mb RAM, Compaq Computer Corporation, USA) attached to a flatbed scanner (HP ScanJet 4c, Hewlett-Packard Co., USA) with a transparency adapter (HP Transparency Adapter, Hewlett-Packard) (Regent Instrument Inc.),. Comparison of total root growth in all different treatment is presented in Fig. 5.2.1.

Data related to root diameter in spring showed the Ck obtained 0.269 cm^2 whereas treatment 2, had S:N 5:1, 0.316 cm^2at pH 4.5 and this value was also greater than Ck, while minimum value for spring related to root diameter was found in treatment 5, had S:N 0:1, 0.163 cm^2 at pH 2.5. For second season summer, the value for Ck was 0.426 cm^2 and treatment 2, has S:N 5:1, had value greater than Ck at pH 4.5, it was 0.582 cm^2 and minimum value for this season recorded in treatment 3, had S:N 1:1, 0.257 cm^2 at pH 2.5. Data related to root diameter showed maximum growth obtained for spring and summer in treatment 2, had S:N 5:1 at pH 4.5, and same pattern observed for minimum growth of two season in same treatment had concentration of acid 0:1 at pH 2.5. For winter root diameter was 0.480 cm^2 in Ck while maximum value found in treatment 1, had S:N 1:0, 0.591 cm2 at pH 4.5 and then minimum value was recorded in treatment 5, had S:N 0:1, 0.235 cm^2 at pH 2.5.

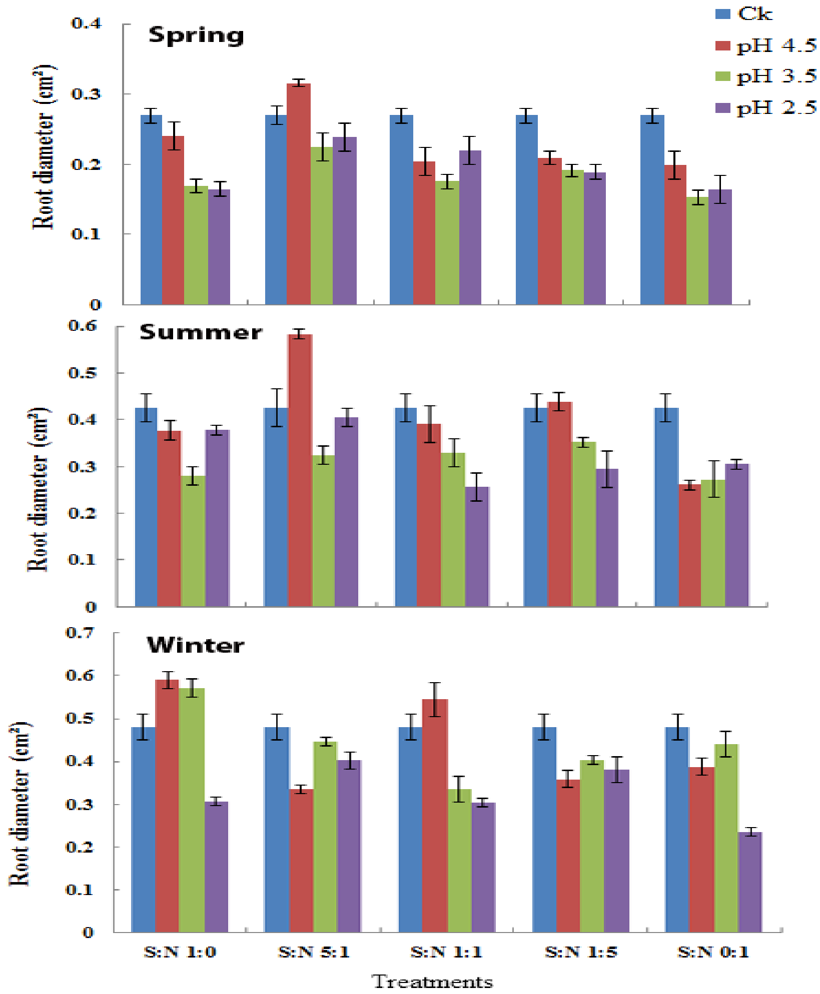

Fig. 5.2.1: Seasonal comparison of mean root diameter in all different treatments

SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

5.2.2 Impact of different pH levels at root diameter of Chinese fir

Fig. 5.2.2 presents the effects of pH levels at root diameter for Chinese fir for three different seasons including spring, summer and autumn.

Mean plot of root diameter for Chinese fir showed Ck contained pH 7 performed better in all seasons as compared to other 3 different pH levels included 4.5, 3.5 and 2.5. Except Ck, the pH level 4.5 showed better results in all the concentrations of acid during whole experiment as compared to 3.5 and 2.5, pH level 2.5 performed worse and caused in the reduction of root diameter for all seasons in all acid concentrations.

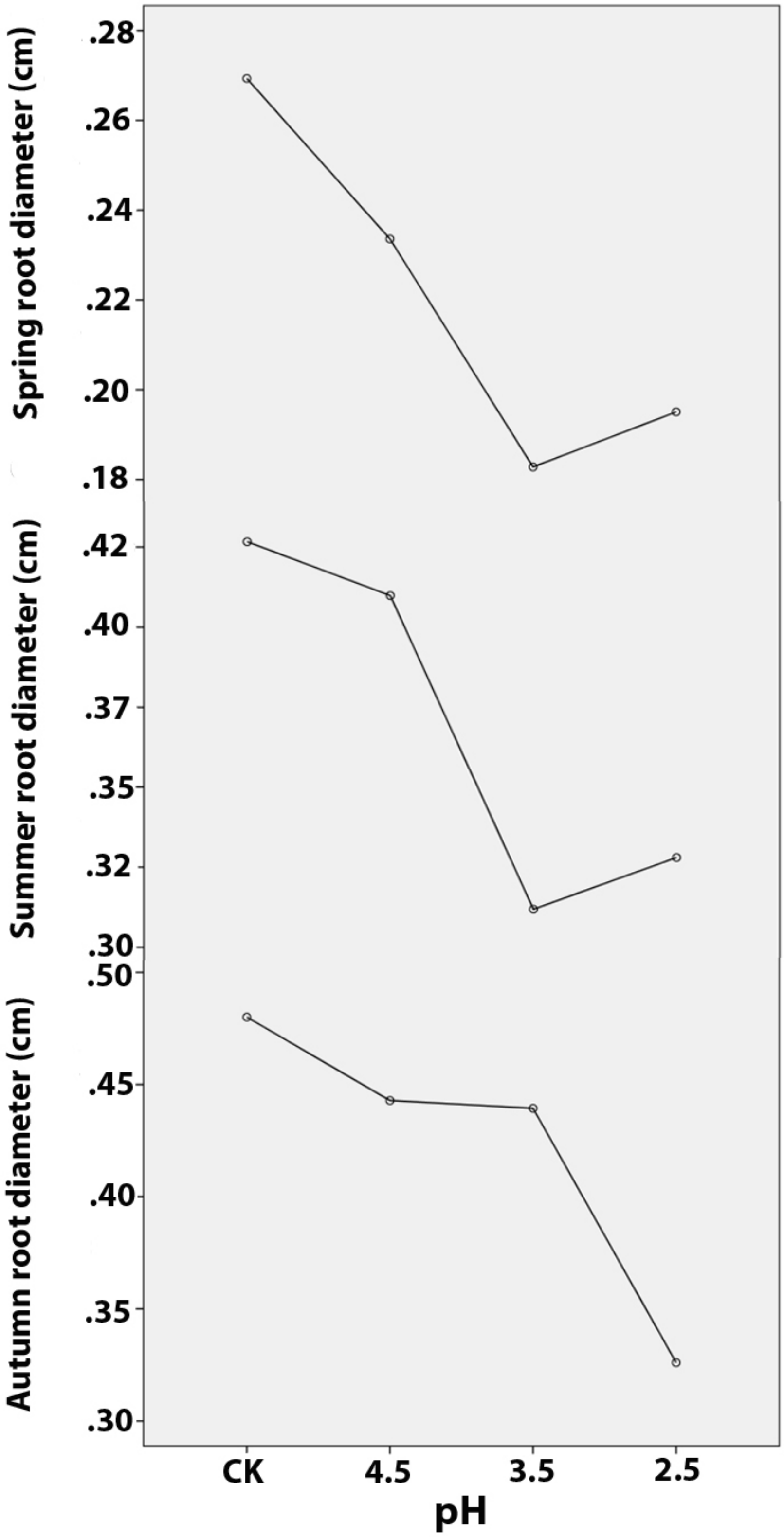

Fig. 5.2.2: Effect of different pH levels 4.5, 3.5, 2.5 and Ck on root diameter for three different seasons

5.3 Mean root surface area of Chinese fir for three different seasons

In every season randomly selected three different seedlings of Chinese fir from each treatment were harvested. Their fine roots were cut and scanned with WinRHIZO 2004b (Regent Instrument Inc.), and it was installed on a Pentium PC (Compaq Deskpro 4000, with 64 Mb RAM, Compaq Computer Corporation, USA) attached to a flatbed scanner (HP ScanJet 4c, Hewlett-Packard Co., USA) with a transparency adapter (HP Transparency Adapter, Hewlett-Packard) (Regent Instrument Inc.),. Comparison of total root growth in all different treatment is presented in Fig. 5.3.1.

In spring, value of root surface area for Ck was observed 635.57 cm^2 while from different concentration the maximum root length recorded in treatment 2, had S:N 5:1, 712 cm^2 at pH 4.5 it was higher than Ck and minimum was found in spring for treatment 4, had S:N 1:5, 304.12 cm^2 at pH 2.5. For second season summer, Ck obtained 1186.45 cm^2, while highest value found in again treatment 2, had S:N 5:1, 1437.31 cm^2 at pH 4.5 and minimum was in treatment 5, had S:N 0:1, 461.52 cm^2. In autumn, there was no such big increase was observed in Ck as compared to previous both seasons (1186.45cm^2) but in treatment 1, had S:N 1:0, obtained the maximum increment it was 1673.54 cm^2 at pH 4.5 and the minimum value was 500 cm^2 at pH level 2.5. During all three seasons we observed minimum value in the same treatment it showed it had severely disturbed the seedlings at pH 2.5

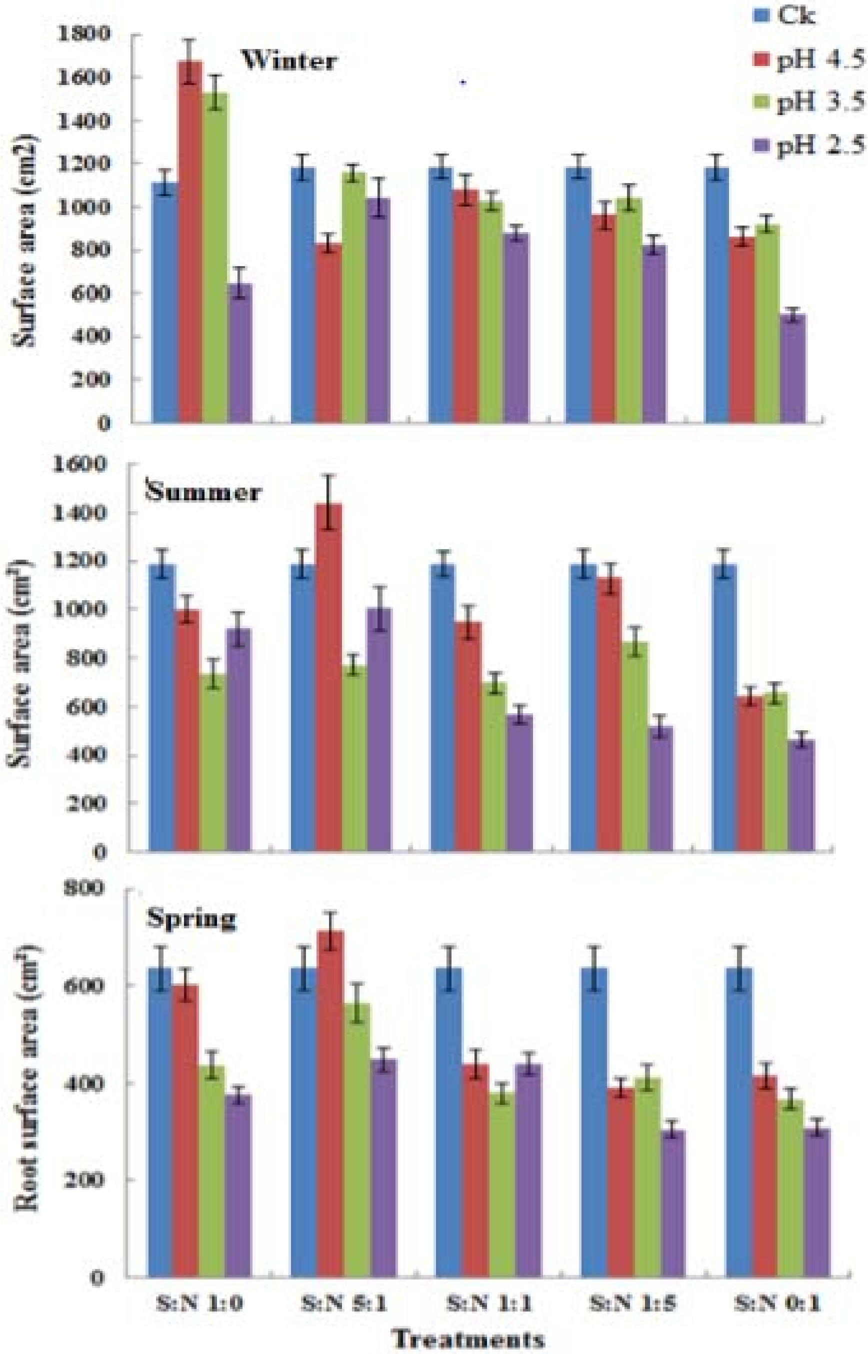

Fig. 5.3.1: Seasonal comparison of root surface area growth in all different treatments
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

5.3.2 Impact of different pH levels at root surface area of Chinese fir

Fig. 5.3.2 presents the effects of pH levels at root surface area for Chinese fir for three different seasons including spring, summer and autumn.

Mean plot of root diameter for Chinese fir showed Ck attained at pH 7 performed better in all seasons as compared to other 3 different pH levels included 4.5, 3.5 and 2.5. Except Ck, the pH level 4.5 showed better performances in all the concentrations of acid during whole experiment as compared to 3.5 and 2.5, pH level 2.5 performed worst and caused in the reduction of root diameter for all seasons in all acid concentrations.

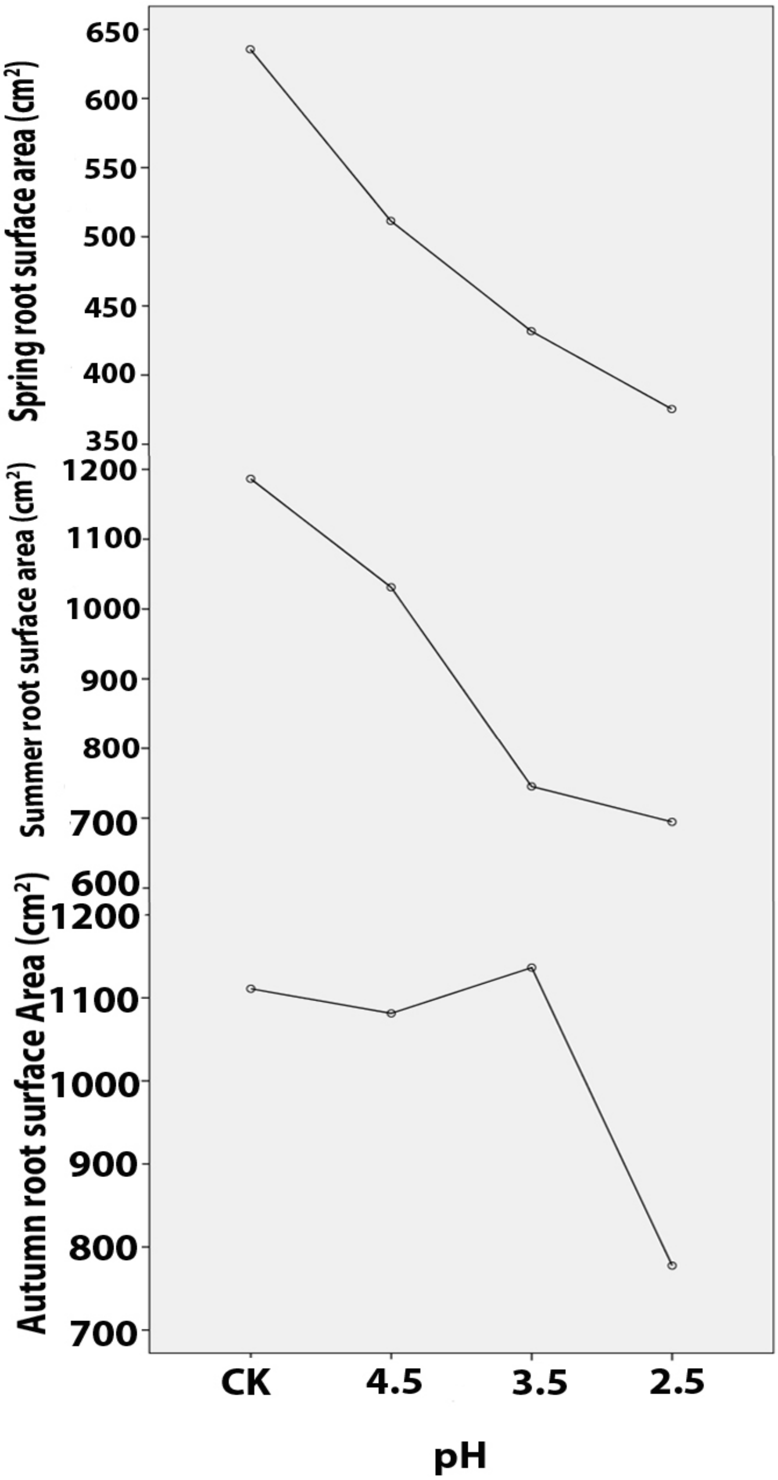

Fig. 5.3.2: Mean plot of root surface area for pH 4.5, 3.5, 2.5 and Ck contained pH level 7 for three different seasons

5.4 Mean root volume of Chinese fir for three different seasons

In every season, randomly selected three different seedlings of Chinese fir from each treatment were harvested. Their fine roots were cut and scanned all of them with WinRHIZO 2004b (Regent Instrument Inc.), and it was installed on a Pentium PC (Compaq Deskpro 4000, with 64 Mb RAM, Compaq Computer Corporation, USA) attached to a flatbed scanner (HP ScanJet 4c, Hewlett-Packard Co., USA) with a transparency adapter (HP Transparency Adapter, Hewlett-Packard) (Regent Instrument Inc.),. Comparison of total root growth in all different treatment is presented in Fig. 5.4.1.

Data related to root volume in spring showed the Ck obtained 14.51 cm^3 whereas treatment 2, had S:N 5:1, 15.57 at pH 4.5 and this value was also greater than Ck, while minimum value for spring related to root volume was found in treatment 4, had S:N 1:5, 7.41 cm^3 at pH 2.5. For second season summer, the value for Ck was 29.47cm^3 and treatment 2, has S:N 5:1, had value greater than Ck at pH 4.5, it was 41.48 cm^3 and minimum value for this season recorded in treatment 4, had S:N 1:5, 14.31 cm^3 at pH 2.5. Data related to root volume showed maximum growth obtained for spring and summer in treatment 2, had S:N 5:1 at pH 4.5, and same pattern observed for minimum growth of two season in same treatment had concentration of acid 1:5 at pH 2.5. For winter root volume was 37.26 cm^3 in Ck while maximum value found in treatment 1, had S:N 1:0, 51.24 cm3 at pH 4.5 and then minimum value was recorded in treatment 5, had S:N 0:1, 15.12 cm^3 at pH 2.5

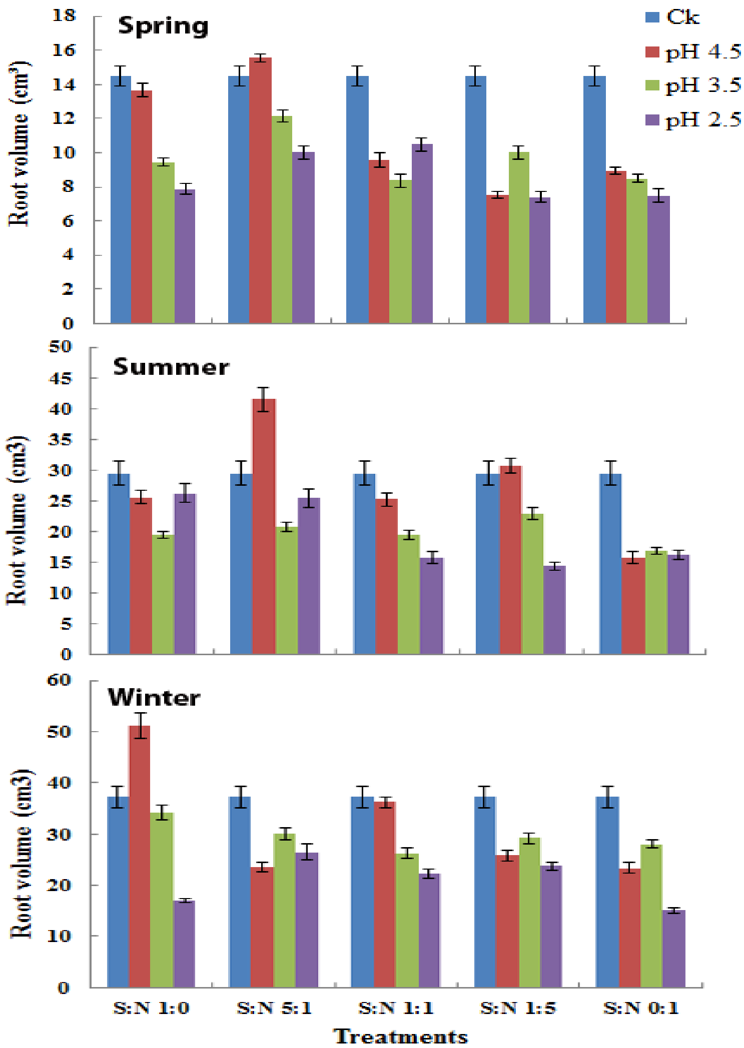

Fig. 5.4.1: Seasonal comparison of root volume growth in all different treatments
SAR 1,2,3 contained S:N 1:0 and pH 4.5, 3.5 and 2.5 respectively, SAR 4,5,6 contained S:N 5:1 and pH 4.5, 3.5 and 2.5 respectively, SAR 7,8,9 contained S:N 1:1 and pH 4.5,3.5 and 2.5, SAR 10,11,12 contained S:N 1:5 and pH 4.5,3.5 and 2.5 respectively, SAR 13,14,15 contained S:N 0:1 and pH 4.5,3.5 and 2.5 respectively while CK contained pH 7.0

5.4.2 Impact of different pH levels at root volume of Chinese fir

Fig.5.4.2 presents the effects of pH levels at root volume for Chinese fir for three different seasons including spring, summer and autumn.

Mean plot of root diameter for Chinese fir showed Ck contained pH 7 performed better in all seasons as compared to other 3 different pH levels included 4.5, 3.5 and 2.5. Except than Ck, the pH level 4.5 showed better performance in all the concentrations of acid during whole experiment as compared to 3.5 and 2.5, pH level 2.5 performed worse and caused in the reduction of root diameter for all seasons in all acid concentrations.

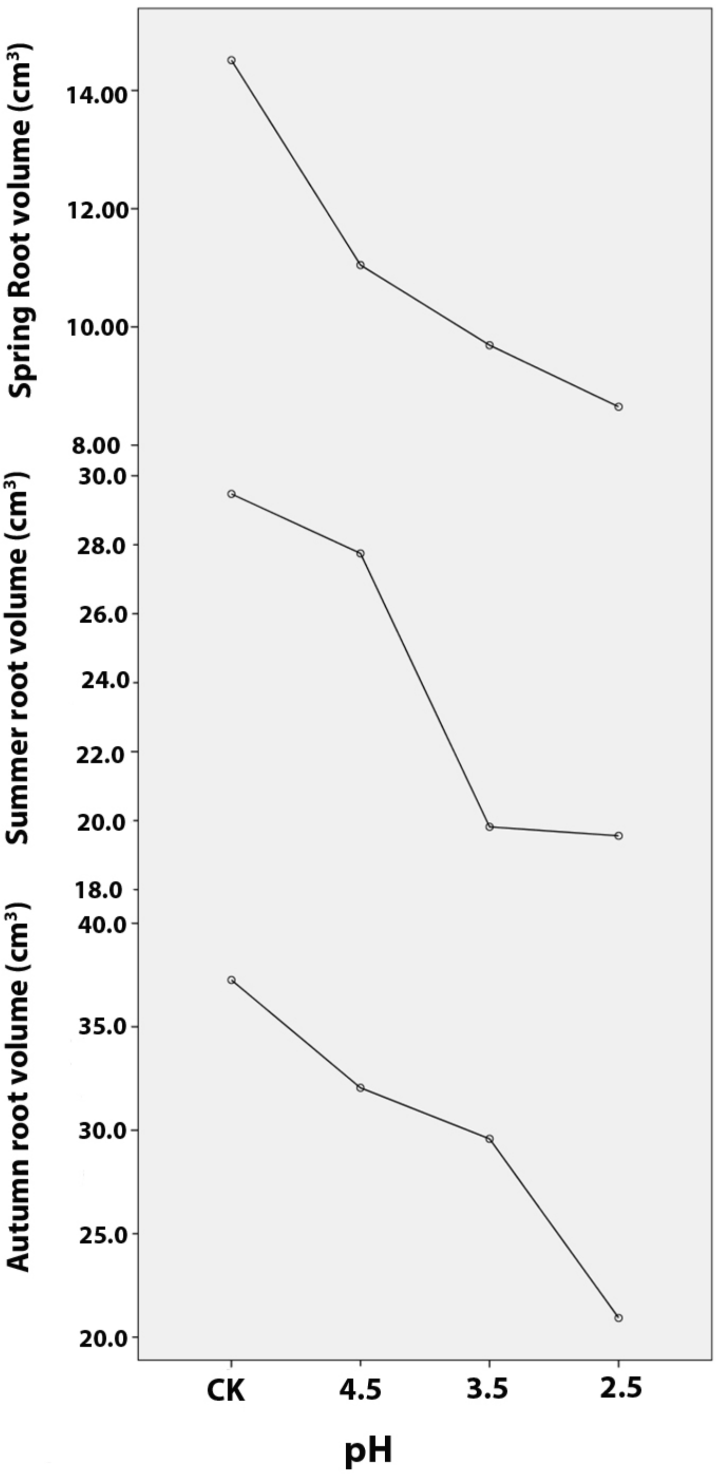

Fig. 5.4.2: Mean plot of root surface area for pH 4.5, 3.5, 2.5 and Ck contained pH level 7 for three different seasons

6. Discussion

No significant difference in plants height was observed during the whole experiment although, slightly significant was observed in some plants in different treatments. For mean growth of tree height for all 4seasons for Chinese fir and Oak, maximum and minimum values were observed SAR14 and SAR2 while for Oak maximum was in CK and minimum was SAR14. In spring season, average height was observed in the range of 23 to 28 cm, while the maximum relative height was recorded in SAR6 (S:N, 5:1 pH, 2.5), i.e. 27.66 cm. For summer, autumn, and winter season the maximum tree height was observed in SAR14 (S:N, 3.5 pH, 3.5) i.e.47.06 cm , 52.83 cm in SAR12 (S:N, 1:5 pH, 2.5) and 56.6 cm in SAR14(S:N, 1:5 pH, 3.5) . On the other hand, minimum relative height throughout the summer, autumn, and winter seasons was noted in SAR2 (S:N, 1:0 , pH , 3.5). While for Oak, in spring season, average height was observed in the range of 22 to 23 cm, while the maximum relative height was recorded in SAR13 (S:N, 0:1 PH, 4.5) i.e. 32.15 cm. For summer, autumn, and winter season, the maximum tree height, 52.16 cm, was observed in Ck, 51.66 cm and SAR9 (S:N, 1:1 pH, 2.5) 52.06 cm. On the other hand, minimum relative height throughout the summer, autumn, and winter seasons was noted in SAR14 (S:N, 0:1 , pH , 3.5), SAR14 (S:N, 0:1, pH,3.5) and SAR8 (S:N, 1:1 pH,3.5) i.e27.28 cm, 32.75 cm and 33.70 cm, respectively. For Oak, the maximum growth obtained in Ck during almost all seasons but reduction of plants height of two species observed in lower pH and more concentration affected directly plants height. Similar resulted were reported by Balasubramanian et al., (2007).

Plants during whole experiment showed variations with reference to relative tree height; these variations might be due to differences in acidity level. Similar trend on plant height reduction in many forests and field crops under simulated acid rain events has been previously reported in a number of studies (Sonia and Khan, 1996; Gadallah, 2000; Huang, et al., 2006).Simulated rain having pH 3.5 the function of cell expansion seemed to be more sensitive than the function of cell division and this has caused the reduction in plant height. While Dixon and Kuja (1995) reported increment in the plant height in sugar maple seedlings subjected to moderate levels of acid rain than seedlings receiving normal rain (pH 5.6) and attributed this to possible enhancement of photosynthesis after exposure to acid rain.

During the experiment, the highest value of seedlings height was observed in almost all season at pH 3.5, this finding have resemblance to the work of Lee, et al.,(1980). They reported that pH 3.0 marginally increased the total yield of corn, although it had foliar injury.

Mean DBH growth of Chinese fir seedlings throughout the four seasons. No significant difference in plants height was observed during the whole experiment although some plants in different treatments behaved slightly significantly. For mean growth of DBH for all the four seasons for Chinese fir and Oak, maximum and minimum values were observed SAR11 and SAR1 while Oak showed maximum value in SAR1 and minimum value was observed in SAR14.In spring season we noted average DBH growth between 0.38- 0.49 cm, while the maximum relative mean DBH growth was recorded in SAR10 (S:N, 1:5 pH, 4.5) 0.48 cm. For summer, autumn, and winter season the maximum DBH growth was observed in SAR11 (S:N, 1:5pH, 3.5) i.e. 0.56 cm , SAR11 (S:N, 1:5 pH, 3.5) i.e. 0.69 cm and SAR11(S:N, 1:5 pH, 3.5) i.e. 0.83 cm. On the other hand minimum mean DBH growth throughout the summer, autumn, and winter seasons was noted in SAR12 (S:N, 1:0, PH , 3.5), SAR2 (S:N, 1:0 PH, 3.5) and in SAR1 (S:N 1:0, PH 4.5) i.e. 0.46 cm, 0.53 cm and 0.596 cm, respectively. While for Oak, in spring season we noted average DBH growth in the range of 0.022 to 0.300 cm, while the maximum relative height was recorded in SAR8 (S:N, 1:1 pH, 3.5) i.e. 0.300 cm. For summer, autumn, and winter season the maximum DBH growth was observed in SAR1(S:N, 1:0 pH,4.5), , SAR9(S:N, 1:1 pH, 2.5) and SAR9(S:N, 1:1 pH, 2.5) i.e. 0.398 cm, 0.436 cm and 0.417 cm, respectively. On the other hand, minimum mean DBH throughout the summer, autumn, and winter seasons was noted in SAR9 (S:N, 1:1 , pH , 2.5) i.e. 0.270 cm, SAR4 (S:N, 5:1, pH,4.5) i.e. 0.329 cm and SAR14 (S:N, 0:1 pH,3.5) i.e. 0.279 cm.

For mean DBH growth, the minimum DBH growth values for all season was found in SAR2 and SAR1, that contained 1:0 ratios and pH < 3.5 of H_2SO_4 and HNO_3. Similar results were reported by Balasubramanian et al., (2007)

No significant difference in plants crown diameter was observed during the whole experiment of two species although some plants in different treatments behaved slightly significantly. For mean growth of crown diameter for all the 4seasons for Chinese fir and Oak maximum and minimum values were observed SAR8 and SAR7 while for Oak maximum was in CK and minimum was SAR14. In spring season of Chinese fir, average growth of canopy was found in the range of 10 to 25 cm, while the maximum relative mean growth of canopy was recorded in SAR5 (S:N, 5:1 pH, 3.5) i.e. 24.08 cm. For summer, autumn, and winter season the maximum growth was observed in SAR14 (S:N, 0:1pH, 3.5) i.e. 50 cm , SAR8 (S:N, 1:1 pH, 3.5) i.e. 48.41 cm and SAR11(S:N, 1:5 pH, 3.5) i.e. 60 cm. This growth pattern was also observed by Kuja (1994) who described that pH 3.2 and 4.3 treated sugar maple seedlings, tended to show greater growth than the pH 5.6 treated sugar maple seedlings. This trend toward increased growth may also be attributable to the higher nitrogen concentrations in the pH 3.2

and 4.3 treatments. On the other hand, minimum mean crown diameter growth throughout the summer, autumn and winter seasons was noted in SAR5 (S:N, 5:1 , pH , 3.5), SAR4 (S:N, 5:1 pH, 4.5) and in SAR5 (S:N 5:1, pH 3.5) i.e. 38.5 cm, 36 cm and 42.91 cm, respectively. In case of Chinese fir, minimum growth of plants canopy was observed in same treatment (S:N 5:1) that had the concentration of H_2SO_4 and HNO_3although every SAR had different pH level but concentration of acids behaved in negative way and caused reduction. Similar results were reported by Kabir et al. (2012)

While for Oak in spring season, we noted average canopy growth for between the range of 14 to 22 cm, while the maximum relative canopy growth was recorded in SAR11 (S:N, 1:5 pH, 3.5) i.e. 21.66 cm. For summer, autumn, and winter season the maximum crown diameter growth was observed in Ck, i.e33.16 cm, , 29.16 cm and , 27.15 cm, respectively. On the other hand, minimum mean crown diameter throughout the summer, autumn, and winter seasons was noted in SAR14 (S:N, 0:1 , pH , 3.5), SAR15 (S:N, 0:1, pH,2.5) and also SAR15 (S:N, 0:1 pH,2.5) i.e. 22.30cm,16.29 cm and14.87 cm, respectively. In case of Oak the highest growth of crown diameter for summer, autumn, and winter seasons were found in Ck treatment except spring season where it was in SAR11.Furthermore, the minimum values of crown diameter for summer, autumn, and winter seasons were noted within the same treatment that had the concentration of H_2SO_4 and HNO_3 (S:N, 0:1) but pH for all SAR were 3.5 and less than this. This trend showed the pH< 3.5 could reduce and affect the crown diameter; same results were observed by Gadallah, (2000) and Balasubramanian et al. (2007). The data revealed that low pH (3.5) of either sulfuric acid or the combination of H_2SO_4 and HNO_3more severely all parameters affected including number of leaves, shoot: root ratio, water contents of shoot reported by Imran and Meo (2014).

No significant difference in the length of new seedling branches was observed within the same season during the whole experiment, although some plants in different treatments behaved slightly significantly. For mean growth of new branches for all 4seasons for Chinese fir and Oak, maximum and minimum values were observed for SAR8 and SAR7 while for Oak maximum was observed in CK and minimum was SAR14. In spring season of Chinese fir, average length of new branches was observed in the range of 7 to 10 cm, while the maximum relative mean growth of new branches was recorded in SAR6 (S:N, 5:1 pH, 2.5) i.e. 9.16 cm. For summer, autumn, and winter season the maximum growth was observed in SAR9 (S:N, 1:1pH, 2.5), SAR10 (S:N, 1:5 pH, 4.5) and SAR13(S:N, 0:1 pH, 4.5) i.e. 30.4 cm, 33.66 cm and 23.95 cm, respectively. Better growth in branches could lead to increment in height

(McLaughlin et al. 1988) and similar resultswere reported by Dixon and Kuja(1995). Highest value for branch growth in all seasons was found in SARs having pH 4.5 and greater than this. This enhancement in length of branches depicts the slight fertilizer effect of nitrogen in case of nitric acid HNO_3 application (in milder acidic pH). The results are similar to those reported by Muhammad Asif Imran and Meo (2014). High nitrogen inputs can lead to growth stimulation (McLaughlin, et al., 1988; Dean and Johnson, 1992)

7. Conclusion

The total duration of green house experiment was almost 11 months. The whole experiment was divided into four different seasons with reference to collection of data. Due to short duration of this experiment, it is very difficult to make conclusions about the efficiency of both species with reference to morphological characters under simulated acid rain conditions. In this experiment, the effects of simulated acid rain within plant species with reference to all seasons and also comparison of both species regarding all parameters like plants Height, DBH, Crown diameter and new branches length were concluded.

For Chinese fir, maximum growth height, DBH, crown diameter and new branches length were observed in treatment 4, with S:N 1:5 and pH 4.5, almost during all seasons and reduction in seedling height was observed in treatment 1, with S:N 0:1 and pH 2.5. Higher concentrations of both acids had directly affected plant height. For mean DBH growth, for all the seasons the minimum DBH growth values were observed SAR2 and SAR1 which contained 1:0 ratios and pH < 3.5 of H_2SO_4 and HNO_3. For crown diameter, in case of Chinese fir, minimum growth of plants canopy was observed in the same treatment (S:N 5:1) that had the concentration of H_2SO_4 and HNO_3 although every SAR had different pH level but concentration of acids behaved in negative way and caused reduction in crown diameter.

While for Oak the maximum growth of height, DBH, crown diameter and new branches length was observed in treatment 4, with S:N 1:5 and pH 4.5 during almost all seasons. The minimum values of crown diameter for summer, autumn and winter seasons were observed within the same treatment that had the concentration of H_2SO_4 and HNO_3 (S:N, 0:1) but pH for all SAR was 3.5 and less than this. This trend showed us the pH< 3.5 could reduce and effect crown diameter. For New branches length, slightly better effects of simulated acid rain on both species were observed after exposure to simulated acid rain with higher concentration of HNO_3 and pH greater than 3.5. This enhancement in length of branches depicts the slight effect of nitrogen fertilizer in case of nitric acid HNO_3 application (in milder acidic pH). Data showed treatment 4, with S:N 1:5 and pH 4.5 had better affect on all the parameters of both seedlings as compared to others. It might be due to nitrogen effect in this treatment.

Data related to Chinese fir's root characteristics revealed that root length, root volume, root diameter and root surface area for all three seasons were highest in the treatment 2, had S:N 5:1 at pH level 4.5 and minimum recorded in treatment 5, had S:N 0:1 at pH 2.5 ,except than in autumn where it was observed highest in treatment 1, had S:N 1:0, at ph 4.5, and minimum was in treatment 5, had S:N 0:1, at pH 2.5. Overall all concentrations of both acids had stressed on all the root characteristics. Mean values for different level of pHs revealed that Ck (pH 7) behaved positively and all the parameters gained highest value in Ck; as compared to other pH levels 4.5, 3.5 and 2.5.

8. Future prospects

During our whole experiment we observed some plants stems and leaves were affected by fungus attack. Might be it was due to the anaerobic conditions.

Foliar application of simulated acid rain having HNO_3 could lead in the enhancement of branches length and root growth, by introducing nitrogen fixing bacteria in the soil it can be investigate more clearly by further study on this aspect.

References

1. Anatolaki, C. and R. Tsitouridou (2009). "Relationship between acidity and ionic composition of wet precipitation: a two years study at an urban site, Thessaloniki, Greece." Atmospheric Research 92(1): 100-113.
2. André, F., M. Jonard and Q. Ponette (2007). "Influence of meteorological factors and polluting environment on rain chemistry and wet deposition in a rural area near Chimay, Belgium." Atmospheric Environment 41(7): 1426-1439.
3. Arti, V., T. Ashish and A. Abdullah (2010). "Impact of simulated acid rain of different pH-levels on some major vegetable plants in India." Reports and Opinions 2: 38-40.
4. Ashenden, T. W. and S. A. Bell (1987). "The effects of simulated acid rain on the growth of three herbaceous species grown on a range of British soils." Environmental pollution (Barking, Essex : 1987) 48(4): 295-310.
5. Baez, A., R. Belmont, R. García, M. Torres and H. Padilla (2006). "Rainwater chemical composition at two sites in Central Mexico." Atmospheric Research 80(1): 67-85.
6. Balasubramanian, G., C. Udayasoorian and P. Prabu (2007). "Effects of short-term exposure of simulated acid rain on the growth of Acacia nilotica." Journal of Tropical Forest Science: 198-206.
7. Bardswick, W., W. Chan and D. Orr (1986). "A quality assurance program and quality assessment of the acidic precipitation in Ontario study (APIOS) deposition monitoring networks." Water, Air, and Soil Pollution 30(3-4): 981-990.
8. Bhattacharya, A., R. Mudgal and A. Taneja (2004). "Acid deposition and critical load analysis in Agra, India." Journal of hazardous materials 106(2): 157-160.
9. Caporn, S. J. M. and T. C. Hutchinson (1986). "The contrasting response to simulated acid rain of leaves and cotyledons of cabbage (brassica oleracea l.)." New Phytologist 103(2): 311-324.
10. CBEF. (2016). "Acid Rain Overview." from http://www.chesbay.org/acidRain/.
11. Çelik, M. B. and K. İbrahim (2007). "The relation between meteorological factors and pollutants concentrations in Karabük city." Gazi University Journal of Science 20(4): 87-95.
12. Chang,Y.-J.Z.H.-R. (2012). "The impact of acid rain on China’s socioeconomicvulnerability." Nat Hazards (2012) 64: 1671–1683.
13. Chesapeake Bay Ecological Foundation, I. (2016). "Acid Rain Overview." from http://www.chesbay.org/acidRain/.

14. Dean, T. J. and J. D. Johnson (1992). "Growth response of young slash pine trees to simulated acid rain and ozone stress." Canadian Journal of Forest Research 22(6): 839-848.
15. Dixon, M. J. and A. L. Kuja (1995). "Effects of simulated acid rain on the growth, nutrition, foliar pigments and photosynthetic rates of sugar maple and white spruce seedlings." Water, air, and soil pollution 83(3-4): 219-236.
16. Evans, L. S., D. C. Canada and K. A. Santucci (1986). "Foliar uptake of 15 N from rain." Environmental and experimental botany 26(2): 143-146.
17. Evans, L. S., K. F. Lewin and E. A. Cunningham (1982). "Air Pollution Effects on CropsEffects of simulated acidic rain on yields of field-grown radishes and garden beets." Agriculture and Environment 7(3): 285-298.
18. Feng, Z. (2000). "Ecological effects and control strategies of acid deposition on ecosystems in China." Yunnan Environ Sci 19: 1-6.
19. Francisco Sant'Anna-Santos, B., L. Campos da Silva, A. Alves Azevedo, J. Marcos de Araújo, E. Figueiredo Alves, E. Antônio Monteiro da Silva and R. Aguiar (2006). "Effects of simulated acid rain on the foliar micromorphology and anatomy of tree tropical species." Environmental and Experimental Botany 58(1–3): 158-168.
20. Gadallah, M. (2000). "Effects of acid mist and ascorbic acid treatment on the growth, stability of leaf membranes, chlorophyll content and some mineral elements of Carthamus tinctorius, the safflower." Water, air, and soil pollution 118(3-4): 311-327.
21. Grimm, J. W. and J. A. Lynch (2004). "Enhanced wet deposition estimates using modeled precipitation inputs." Environmental Monitoring and Assessment 90(1-3): 243-268.
22. Haiyan, W. and A. O. Stuanes (2003). "Heavy metal pollution in air-water-soil-plant system of Zhuzhou City, Hunan Province, China." Water, Air, and Soil Pollution 147(1-4): 79-107.
23. Harcourt, S. A. and J. F. Farrar (1980). "Some effects of simulated acid rain on the growth of barley and radish." Environmental Pollution Series A, Ecological and Biological 22(1): 69-73.
24. Huang, D.-Y., Y.-G. Xu, P. a. Peng, H.-H. Zhang and J.-B. Lan (2009). "Chemical composition and seasonal variation of acid deposition in Guangzhou, South China: comparison with precipitation in other major Chinese cities." Environmental Pollution 157(1): 35-41.

25. Huang, Y., Z. Li, X. Li, K. Wu and J. Sun (2006). "Impact of simulated acid rain on growth and nutrient elements uptake by Eucalyptus urophylla and Pinus massoniana." Ecol Environ 15: 331-336.
26. Jiang, P., Q. Xu and Y. Yu (2002). "Microbial biomass carbon as an indicator for evaluation of soil fertility." Journal of Zhejiang Forestry College 19(1): 17-19.
27. JSSB. (2014). "主要城市月降水量." From http://www.jssb.gov.cn/2015nj/nj09/nj0902.htm.
28. Kabir, M., Z. Iqbal and M. Sahfiq (2012). "Traffic density, climatic conditions and seasonal growth of samanea saman (Jacq.) Merr. on different polluted roads of Karachi city." Pak. J. Bot 44(6): 1881-1890.
29. Kuja, M. J. D. A. A. L. (1994). Effects of simulated acid rain on the growth,nutrition, foliar pigments and photosynthetic ratesof sugar maple and white spruce seedlings. Ontario ministry of environment andenergy.
30. Lee, J., G. Neely and S. Perrigan (1980). Sulfuric acid rain effects on crop yield and foliar injury. Final report, Environmental Protection Agency, Corvallis, OR (USA). Environmental Research Lab.
31. Leith, I., M. Murray, L. Sheppard, J. Cape, J. Deans, R. Smith and D. Fowler (1989). "Visible foliar injury of red spruce seedlings subjected to simulated acid mist." New phytologist 113(3): 313-320.
32. Lin, C., J. Liu, Y. Cai, B. Li, Z. Wang and B. Chen (2009). "Study on the relationship between meteorological conditions and acid rain in mid-eastern Fujian." Bulletin of environmental contamination and toxicology 83(2): 180-187.
33. Mandre, M. and J. Klyshejko (1995). "Effect of foliar treatment with simulated acid mists on biomass formation and nutrient composition of plants." Zeitschrift für Naturforschung C 50(7-8): 521-526.
34. McLaughlin, S., M. Adams, N. Edwards, P. Hanson, P. Layton, E. O'Neill and W. Roy (1988). Comparative sensitivity, mechanisms, and whole plant physiological implications of responses of loblolly pine genotypes to ozone and acid deposition, Oak Ridge National Lab., TN (USA).
35. Muhammad Asif Imran, S. H., Mumtaz Hussain, and M. N. C. A. A. A. Meo (2014). "Effect of Simulated Acid Rain (Sar) on Some MorphochemicalAspects of Mash (*Vigna Mungo* L.)." Pak. J. Bot 46(1): 245-250.
36. NADP (2012). "National Atmosphric Deposition Program."

37. Patrinos, A., M. Leach, R. Brown, R. Tanner and F. S. Binkowski (1989). "An acid rain study in the Washington, DC area." Journal of Applied Meteorology 28(9): 948-968.
38. Percy, K. E. and E. A. Baker (1990). "Effects of simulated acid rain on epicuticular wax production, morphology, chemical composition and on cuticular membrane thickness in two clones of Sitka spruce [Picea sitchensis (Bong.) Carr.]." New Phytologist 116(1): 79-87.
39. Ping, L., X. Fei, P. Jiayong, C. Yiping, P. Huaming and C. Shaohua (2011). "Discuss on Present Situation and Countermeasures for Acid Rain Prevention and Control in China [J]." Environmental Science and Management 12: 008.
40. Prado-Fiedler, R. (1990). "On the relationship between precipitation amount and wet deposition of nitrate and ammonium." Atmospheric Environment. Part A. General Topics 24(12): 3061-3065.
41. QLD. (2013). "Air Pollutants." from https://www.qld.gov.au/environment/pollution/monitoring/air-pollution/sulfur-dioxide/.
42. Ramlall, C., B. Varghese, S. Ramdhani, N. W. Pammenter, A. Bhatt, P. Berjak and Sershen (2015). "Effects of simulated acid rain on germination, seedling growth and oxidative metabolism of recalcitrant-seeded Trichilia dregeana grown in its natural seed bank." Physiologia Plantarum 153(1): 149-160.
43. Romero, H., M. Ihl, A. Rivera, P. Zalazar and P. Azocar (1999). "Rapid urban growth, land-use changes and air pollution in Santiago, Chile." Atmospheric Environment 33(24): 4039-4047.
44. Silva, L. C., M. A. Oliva, A. A. Azevedo, J. M. Araújo and R. M. Aguiar (2005). "Micromorphological and anatomical alterations caused by simulated acid rain in Restinga plants: Eugenia uniflora and Clusia hilariana." Water, Air, and Soil Pollution 168(1): 129-143.
45. Singh, A. K., G. Mondal, S. Kumar, K. Singh, K. Kamal and A. Sinha (2007). "Precipitation chemistry and occurrence of acid rain over Dhanbad, coal city of India." Environmental monitoring and assessment 125(1-3): 99-110.
46. Singh, B. and M. Agrawal (2004). "Impact of simulated acid rain on growth and yield of two cultivars of wheat." Water, Air, and Soil Pollution 152(1-4): 71-80.
47. Sonia, R. and M. Khan (1996). "Effect of simulated acid rain on Cicer arietinum var. Pant G-114." Journal of Environmental Pollution 3(3-4): 197-201.
48. Tamm, C. O. and L. Hallbäcken (1986). Changes in soil pH over a 50-year period under different forest canopies in SW Sweden. Acidic Precipitation, Springer: 1391-1395.

49. Tang, A., G. Zhuang, Y. Wang, H. Yuan and Y. Sun (2005). "The chemistry of precipitation and its relation to aerosol in Beijing." Atmospheric Environment 39(19): 3397-3406.
50. Tripathi, A. and M. Gautam (2007). "Biochemical parameters of plants as indicators of air pollution." Journal of Environmental Biology 28(1): 127.
51. Troiano, J., L. Heller and J. Jacobson (1982). "Effect of added water and acidity of simulated rain on growth of field-grown radish." Environmental Pollution Series A, Ecological and Biological 29(1): 1-11.
52. USGS. (2016). "Acid Rain." from http://water.usgs.gov/edu/acidrain.html.
53. Van Huylenbroeck, J., A. Piqueras and P. Debergh (2000). "The evolution of photosynthetic capacity and the antioxidant enzymatic system during acclimatization of micropropagated Calathea plants." Plant Science 155(1): 59-66.
54. Vautz, W., A. Busch, W. Urfer and D. Klockow (2003). "A statistical approach to estimate spatial distributions of wet deposition in Germany." Water, air, and soil pollution 145(1-4): 215-238.
55. Viet, P., V. Tuan, P. Hoai, N. Anh and P. Yen (2001). Chemical composition and acidity of precipitation: A monitoring program in northeastern Vietnam. Acid rain 2000, Springer: 1499-1504.
56. Wagh, N., P. V. Shukla, S. B. Tambe and S. Ingle (2006). "Biological monitoring of roadside plants exposed to vehicular pollution in Jalgaon city." Journal of Environmental Biology 37(2): 419-421.
57. Walna, B., S. Drzymała and J. Siepak (2000). "The impact of acid rain on potassium and sodium status in typical soils of the Wielkopolski National Park (Poland)." Water, air, and soil pollution 121(1-4): 31-41.
58. Wang, C., P. Guo, G. Han, X. Feng, P. Zhang and X. Tian (2010). "Effect of simulated acid rain on the litter decomposition of Quercus acutissima and Pinus massoniana in forest soil microcosms and the relationship with soil enzyme activities." Science of the Total Environment 408(13): 2706-2713.
59. Wang, T., F. Jiang, S. Li and Q. Liu (2007). "Trends in air pollution during 1996-2003 and cross-border transport in city clusters over the Yangtze River Delta region of China." Terrestrial Atmospheric and Oceanic Sciences 18(5): 995.
60. Wertheim, F. S. and L. E. Craker (1987). "Acid rain and pollen germination in corn." Environmental Pollution 48(3): 165-172.

61. Xiaoqin Wang , Z. L., Li Niu , and B. Fu (2013). "Long-term effects of simulated acid rain stress on a staple forestplant, Pinus massoniana Lamb: a proteomic analysis." Trees (2013) 27:297–309 27:297–309: 297–309.

62. Xinmin, Z., C. FaHe, W. Shulan, S. XinZhang and H. Mei (2010). "Research progress of acid precipitation in China." Research of Environmental Sciences 23(5): 527-532.

63. Y. KOHNO, H. M., and T. KOBAYASHI (1992). Effect of simulated acid rain on the growth of Japanese conifers grown with or without fertilizer. C. Central Research Institute of Electric Power lndustr 3.

诚樸雄偉
樹木樹人