

## **Bangor University**

DOCTOR OF PHILOSOPHY

Potential uses of red alder (Alnus rubra Bong) in silvopastoral systems

Mmolotsi, Ronnie M

Award date: 2004

Awarding institution: Bangor University

Link to publication

General rights Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
You may not further distribute the material or use it for any profit-making activity or commercial gain
You may freely distribute the URL identifying the publication in the public portal ?

Take down policy If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

POTENTIAL USES OF RED ALDER (Alnus rubra Bong) IN SILVOPASTORAL

SYSTEMS

By

MMOLOTSI, Ronnie M.

# B.Sc., FORESTRY (COLORADO STATE), M.Sc., FORESTRY (EDINBURGH)

# A THESIS SUBMITTED IN FULFILMENT FOR THE PHILLOSOPHIAE

# DOCTOR AT THE UNIVERSITY OF WALES, BANGOR, UK.



# SCHOOL OF AGRICULTURE AND FOREST SCIENCES,

# UNIVERSITY OF WALES, BANGOR,

# **UNITED KINGDOM**

**DECEMBER 2004** 



#### ABSTRACT

Studies on Alnus rubra, Bong. (Red alder) were carried out at Henfaes agroforestry experimental site, in North Wales to asses its potentials for use as a component of silvopastural systems. The specific objectives were: 1) to study the nitrogen fixing capability of red alder using nitrogen-15 natural abundance method, 2) to investigate the dynamics of fine roots and root nodules in red alder and their contribution to organic matter and nitrogen balance under silvopastoral systems, 3) to assess the timber quality of red alder wood in comparison with sycamore and finally, 4) to study the firewood and biomass energy value of red alder in comparison with sycamore. Literature reviewed showed that red alder, as a nitrogen fixing tree species, inputs large amount of nitrogen into natural ecosystems where it occurs. Reports also showed that it provides the much needed nitrogen when it is grown with other crops. The wood of red alder was reported to be good for furniture, studs, panelling and for making household utensils. There is, however, no information on the use of red alder in silvopasture. In the present study, using nitrogen-15 natural abundance method  $\delta^{15}N$  values close to zero were recoded in red alder plant parts except in root nodules. This indicates that a large proportion of nitrogen in red alder was fixed from the atmosphere. The highest fixed nitrogen was measured in leaves (91%) and the least (78%) in wood of red alder. Fixed nitrogen was higher during the summer and autumn (90% and 99%, respectively) than in winter and spring (85% and 64%, respectively). The  $\delta^{15}$ N values in root nodules were enriched with <sup>15</sup>N and this indicates that soil was the source of nitrogen for root nodules of red alder. Overall, it was estimated that 63.45 and 329.53 kg N ha<sup>-1</sup> was fixed by red alder in the agroforestry and forestry systems, respectively. Live and dead fine root weight densities of red alder were 2700 and 5400 kg ha<sup>-1</sup> and 360 and 790 kg ha<sup>-1</sup> in agroforestry and forestry, respectively. Live and dead root nodule weight density of red alder yielded 880 and 520 kg ha<sup>-1</sup> in agroforestry in 800 and 310 kg ha<sup>-1</sup> in forestry, respectively. The amount of organic matter potentially added to the soil due to senescent leaves and dead roots and root nodules was estimated at 4.0 and 9.1 t ha<sup>-1</sup> yr <sup>1</sup>, in agroforestry and forestry, respectively. These results showed that red alder has a potential to improve and maintain soil fertility in silvopasture. Fine root length density was also found to differ significantly between agroforestry and forestry treatments, between seasons and between depths but there was no difference between distances from the tree base of red alder. Overall, the results of the distribution of fine root length density showed that red alder is compatible with pasture. Wood mechanical properties of red alder were found to be significantly different from that of sycamore. Sycamore yielded higher wood density (0.64 g cm<sup>-3</sup>), modulus of rupture (90.24 MPa) and compression strength (36.49 MPa) than red alder (0.49 g cm<sup>-3</sup>, 73.48 MPa and 32.13 MPa, respectively). However, modulus of elasticity was higher in red alder (7614.64 MPa) than in sycamore (7430.05 MPa), although it was not significantly different. Based on the results of wood properties of red alder it was concluded that red alder is medium strength tree species with potential for furniture manufacturing and for ordinary non-structural uses such as panelling and studs. Although the calorific value of sycamore was higher than red alder, red alder wood gave significantly higher fuel value index (1637.77) than sycamore (1480.60) due to very high ash content of sycamore. Thus, it was concluded that red alder has a potential to provide a better bioenergy than sycamore for heating homes and generating electricity. On the basis of the findings of the present study it was finally concluded that red alder is a suitable tree species for incorporation in silvopasture.

#### ACKNOWLEDGEMENTS

I would like to thank foremost my supervisor, Dr. Zewge Teklehaimanot. This work would not have been possible without his invaluable guidance. He also offered personal support and coaching when things were tough. I also would like to thank Drs. J. B. Hall, J. Healy, M. Lukac, and Prof. Douglas Goldbold, of SAFS, UWB, for their advice during the planning phase of this work. Thank to Drs. M. Breese and Mike Hale who gave advice on the wood chapter. My sincere thanks go to Botswana College of Agriculture (BCA), who accorded me a study leave and funded this study. I thank in particular Prof S. K. Karikari, who supported my application for training. May I thank the SAFS, University of Wales, Bangor, for hosting this study and providing an enabling environment.

I had support from Mr John Evans, Llinos Hughes and Helen Simpson, technicians in SAFS, UWB and therefore would like to thank them. Andy Harris, Dr. Ian Harris, Gwyen Jones who assisted during data collection at Henfaes. Thank you to Jim Frith who processed my wood samples ready for testing. I want to thank also Dr G. Cadisch of the University of London and Dr E. Rowe (formally of SAFS) who gave advice on aspects of nitrogen-15. Thanks also to Jonathan Fear of University of London who analysed my plant samples for nitrogen-15.

Many thanks to my present and former students, W. Kasolo, J. Jatango, Dr. D. Earnshaw, F. C. Callisto, L. Anglaarie, M. Gonzalez-Pena, S. Sola, Drs B. Darko-Obiri, J. Obiri, F. Makhonda, M. Al-amin, S. Thenakoon, who made Bangor exciting. Thanks also to colleagues at BCA, Dr. S. Ngwako, Dr. D. Modise, M. Rampart, C. Patrick and Ishmael Kopong who assisted in one way or another in this study. Special appreciations go to my friends the world wide, R.Pheto, O. Oagile, M. Muskin, Diana Murata, Kuni Suzuki, Keneilwe Segopolo, Mr. and Mrs Segopolo, Thulaganyo Olefile, Mr and Mrs Linchwe and S. Keakopa. They have all been wonderful.

I think it fit to thank my wider family: my mother, Goitsemodimo, my grandmother, Mmammole, my brothers Abel and Andrew, my sisters, Basadi, Baboni, Kitso, Dimpho, Virginia who took care of my affairs while I was away in the UK. I cannot thank them enough.

My sincere thanks to my daughter Tsholo, son Vihape, my new born baby, Tangee, and my fiancée, Kuate, who endured the torture of my absence. They kept me dreaming!

# **TABLE OF CONTENTS**

# CONTENT

DECLARATIONii
ABSTRACTiii
ACKNOWLEDGEMENTSiv
TABLE OF CONTENTS v
LIST OF TABLES xi
LIST OF FIGURES xii
LIST OF PLATES xv
LIST OF SYMBOLS AND ABBREVIATIONSxvi
CHAPTER 1.0 INTRODUCTION 1
1.1 Alnus rubra (red alder)1
1.2 Potential uses of red alder2
1.2.1 Use of red alder for soil fertility improvement and maintenance
1.2.2 The potential of red alder to provide good quality timber4
1.2.3 Fuelwood potential of red alder4
1.3 Aim and objectives of the study
Specific objectives
1.4 Hypothesis of the experiments carried out in this study7
1.5 Thesis Structure
1.6 Study site
1.7 Research design
CHAPTER 2.0 LITERATURE REVIEW
2.1 The Biology of <i>Alnus rubra</i> 13
2.1.1. Taxonomy and systematics
2.1.2. Description of <i>Alnus rubra</i>
2.1.2.1 Seedlings
2.1.2.2 Habitat and form of mature trees14
2.1.2.3 Foliage15
2.1.2.4 Rooting habit15

2.1.2.5 Flowering and fruiting16
2.1.2.6 Phenology and life cycle
2.1.2.7 Seed biology and dispersal
2.2 Ecology
2.2.1 Geographical distribution and range19
2.2.2 Ecological requirements
2.2.3 Plant associations
2.3 Silviculture and management of red alder
2.3.1 Regeneration from seed
2.3.2 Regeneration from vegetative sprouts
2.3.3 Regeneration from planting with seedlings
2.3.4 Site preparation and vegetation management
2.4 Uses of red alder
2.4.1 The role of red alder in soil fertility improvement
2.4.1.1 Fine root turnover and fertility improvement
2.4.2 Nitrogen fixation and soil fertility improvement
i) Systems of Nitrogen fixation
ii) Nitrogen fixing plants
iii) Nitrogen fixation by red alder
iv) Mechanism of infection
v) Environmental factors affecting nitrogen fixation in root nodules
vi) Transfer of fixed nitrogen in the plant
vii) Transfer of fixed nitrogen to the soil
viii) Nodule distribution, dynamics and turnover
2.4.3 Measurement of nitrogen fixation
i) Total nitrogen method40
ii) Total nitrogen difference (TND) method40
iii) Ureides as a measure of biological nitrogen fixation41
iv) Acetylene reduction assay42
v) The nitrogen-15 incorporation method43
vi) The <sup>15</sup> N-labelled fertilizer method44
vii) Nitrogen-15 natural abundance method45
2.4.4 Methods of assessing root and nodule dynamics and turnover

2.4.6 Mechanical properties of wood	48
2.4.6.1 Specific gravity	49
2.4.6.2 Temperature and moisture content	50
2.4.6.3 Knots	50
2.4.6.4 Slope of the grain	51
2.4.7 The use of red alder for firewood and biomass energy	51
2.4.7.1. Biomass energy and wood fuel	52
2.4.7.2. Advantages associated with biomass energy	52
2.4.7.3. Fuel value of wood	53

# CHAPTER 3.0 CONTRIBUTION OF RED ALDER TO SOIL FERTILITY IMPROVEMENT AND MAINTENANCE IN SILVOPASTURAL SYSTEMS

3.1 Introduction
3.2 Methodology
3.2.1 Nitrogen-15 natural abundance in samples
3.2.2 Total biomass of red alder
3.2.3 Total fixed nitrogen accumulated and the rate of annual nitrogen fixation in
red alder61
3.2.4. Contribution of leaves, fine roots and root nodules to soil organic matter and
nitrogen content61
3.2.5. The C:N ratio62
3.3 Statistical analysis of results
3.4 Results
$3.4.1 \delta^{15}$ N values
3.4.2 Nitrogen content in plant parts
3.4.3 $\delta^{15}$ N and nitrogen content in root nodules of red alder and the soil of the
study site65
3.4.4 Fraction of nitrogen derived from the atmosphere in red alder
3.4.5 Total biomass of red alder
3.4.5.2 Below-ground biomass of red alder
(a) Live and dead root weight density and biomass68
(b) Live and dead nodule weight density71

3.5 Total fixed nitrogen accumulated and the rate of nitrogen fixation in red alder
trees74
3.6 Contribution of leaves, roots and dead root nodules to soil organic matter and
nitrogen content
3.6.1. The leaf C:N ratio76
3.7 Discussion
3.7.1 Delta 15N and natural abundance in selected plants at Henfaes
3.7.2 Variation between seasons in $\delta^{15}$ N and FNdfa
3.7.3 $\delta^{15}$ N in root nodules of red alder and soil of the study site
3.7.4 Nitrogen content of plant parts soil of the study site
3.7.5 Fine root and nodule weight density
3.8 Fixed nitrogen
3.9 Contribution of leaves, dead roots and root nodules to soil organic matter and
nitrogen content
3.10. The C:N ratio and decay rates in red alder

# CHAPTER 4.0 FINE ROOT LENGTH DENSITY DISTRIBUTION IN RED

ALDER
4.1 Introduction85
4.2 Methodology
4.3 Analysis of data
4.4 Results
4.4.1 Fine root length density of red alder according to tree planting density88
4.4.2 Fine root length density (RLD) of red alder in relation to season
4.4.3 Fine root length density of red alder in relation to soil depth90
4.4.4 Fine root length density of red alder in relation to distance from tree91
4.4.5 Fine root length of red alder according to root diameter size classes
4.5. Discussion
4.5.1 Fine root length density between treatments
4.5.2 Horizontal distribution of fine root length density
4.5.3 Vertical distribution of fine root density
4.5.4 Fine root length density in relation to season

WITH SYCAMORE	96
5.1 Introduction	96
5.2 Materials and methods	99
5.2.1 Determination of mechanical properties of wood	99
5.2.1.1 Harvesting of wood and processing	.99
5.2.1.2 Preparation of samples	.99
5.2.1.3 Density	100
5.2.1.4 Compression stress	101
5.2.1.5 Bending test	102
5.2.2 Determination of biomass energy values of wood	104
5.2.2.1 Wood harvesting and sample preparation	104
5.2.2.2 Checking the moisture content of the conditioned wood	104
5.2.2.3 Calorific values	105
5.2.2.4 Ash content	107
5.2.2.5 Fuel value index	108
5.3 Analysis of results	109
5.4 Results	109
5.4.1 Mechanical properties of red alder and sycamore	109
5.4.1.1 Wood Density	109
5.4.1.2 Modulus of rupture and elasticity	110
5.4.1.3 Compression stress	112
5.4.2. Biomass energy properties	113
5.4.2.1 Calorific value of wood at different wood moisture contents	113
5.4.2.2 Calorific value of wood at different tree planting densities	116
5.4.2.3 Ash content	117
5.4.2.4 Fuel value index	118
5.5 Discussion	119
5.5.1 Wood density in red alder and sycamore	119
5.5.2 Modulus of rupture and modulus of elasticity	120
5.5.3 Compression strength parallel to the grain	122
5.5.4 Calorific values of red alder and sycamore	122
5.5.5 Ash content and fuel value index	123

# CHAPTER 5.0 WOOD QUALITY OF RED ALDER IN COMPARISON

# CHAPTER 6.0 GENERAL DISCUSSION CONCLUSIONS AND

RECO	OMMENDATIONS	125
6.1	General discussion	125
6.2	CONCLUSIONS	129
6.3	RECOMMENDATIONS	130
	i. Refinement methods	. 130
	ii. Further work at the experimental site	131
REFE	RENCES	134

# LIST OF TABLES

## TABLE

PAGE

Table 1.1.	Average diameter at breast height and tree height (cm) in
	agroforestry and forestry of sycamore and red alder plots in
	different blocks in 200312
Table 2.1.	List of genera of microbes, which include
	nitrogen-fixing species31
Table 3.1.	Values of $\delta^{15}N$ (%/00±SE) in different plant parts (n=3) in four
	Seasons63
Table 3.2.	Mean nitrogen content (%±SE) of different parts of different
	plants (n=12)
Table 3.3.	Values of $\delta^{15}N$ (%/ <sub>00</sub> ±SE) and nitrogen content (%±SE)
	in root nodules of red alder and soils66
Table 3.4.	FNda (%) in red alder tree in relation to reference plants67
Table 3.5.	Biomass of leaves and wood of red alder68
Table 3.6.	Live root weight density (kg m <sup>-3</sup> ±SE) of red alder69
Table 3.7.	Dead root weight density (kg m <sup>-3</sup> ±SE) of red alder70
Table 3.8.	Total fixed nitrogen accumulated in red alder trees
Table 3.9.	Organic matter (kg ha <sup>-1</sup> yr <sup>-1</sup> ) and nitrogen (kg ha <sup>-1</sup> yr <sup>-1</sup> )
	addition to the soil in silvopastoral system76
Table 3.10	Decay rate and weight loss in <i>Alnus glutinosa</i>
Table 5.1.	Wood density (g cm <sup>-3</sup> ±SE of means) in red alder and
	sycamore planted at different densities110
Table 5.2.	Modulus of rupture and elasticity of red alder and sycamore

	planted at different densities 111
Table 5.3.	Compression strength of red alder and sycamore wood
	planted at different densities 112
Table 5.4.	Calorific values (kcal g-1) of firewood of sycamore at
	different moisture contents114
Table 5.5.	Calorific values (kcal g-1) of firewood of red alder at different
	moisture contents114
Table 5.6.	Ash content (%) for sycamore and red alder wood117
Table 5.7.	Fuel value index of sycamore at different moisture contents 117
Table 5.8.	Fuel value index of red alder at different moisture
	contents 118
Table5.9.	Calorific values of wood at different planting densities

# LIST OF FIGURES

FIGURE	PAGE
Figure 1.1.	Distribution of consumption of wood energy by
	regions of the world5
Figure 1.2.	Map of the blocks and plots of sycamore and red
	alder at Henfaes10
Figure 1.3.	Mean monthly rainfall distribution at Henfaes during year
	2002 and 200311
Figure 1.4.	Mean monthly temperature distribution at Henfaes
	during year 2002 and 200311
Figure 2.1.	Global nitrogen cycling 29
Figure 2.2.	Systems of nitrogen fixation, energy source and
	estimates of amounts fixed30
Figure 2.3.	Infection and early organogenesis of nodule lobe in
	actinorhizal plants35
Figure 3.1.	Diagram showing sampling points (at 0.3, 0.75 and 1.0 m)
	from the tree base using logarithmic spiral
	technique60
Figure 3.2.	Mean values of $\delta^{15}N$ (%) <sub>00</sub> ±SE) for different species during
	the four seasons
Figure 3.3.	Live and dead root weight densities (kg m <sup>-3</sup> ±SE)
	in agroforestry70
Figure 3.4.	Live and dead root weight density (kg m <sup>-3</sup> ±SE) in

	forestry71
Figure 3.5.	Nodule weight density (kg m <sup>-3</sup> ±SE) according to distance
	from tree base in red alder agroforestryplots in 2003 72
Figure 3.6.	Nodule weight density (kg m <sup>-3</sup> ±SE) according to
	Distance from tree base in forestry plots in 200372
Figure 3.7.	Live and dead nodule weight density (kg m <sup>-3</sup> ±SE)
	according to season in forestry73
Figure 3.8.	Live and dead nodule weight density (kg m <sup>-3</sup> ±SE)
	according to season in the forestry plots in 200373
Figure 4.1.	Fine root length density (RLD) in agforestry and
	forestry
Figure 4.2.	Fine root length density in relation to season (cm cm <sup>-3</sup> ±SE)89
Figure 4.3.	Fine root density (cm cm <sup>-3</sup> ±SE) at different soil depths91
Figure 4.4.	Change in fine root length density (cm cm <sup>-3</sup> ±SE) with
	increasing distance from the tree
Figure 4.5.	Relationship between fine root length and size classes
Figure 5.1.	Relationship between moisture contents and calorific
	value in sycamore and red alder115
Figure 5.2.	Regression of calorific value against moisture content
	in sycamore115
Figure 5.3.	Regression of calorific value against moisture content in red
	alder

# LIST OF PLATES

PLATE	PAGE					
Plate 2.1.	Red alder stem showing white lenticels and epicormic					
	branches14					
Plate 2.2.	Leaves of red alder showing the in-rolling tooth and					
	tooth-lets					
Plate 2.3.	Red alder catkins17					
Plate 2.4.	Catkins of red alder form the previous year18					
Plate 2.5	Red alder root nodules showing orange-yellow live parts and					
	dead parts					
Plate 4.1.	Root washer					
Plate 4.2.	Root scanner and a computer87					
Plate 5.1.	Bolds of wood from sycamore (white in colour) and red alder					
	showing the distinct red colour stain100					
Plate 5.2.	An instron 5500R series shown during compression test102					
Plate 5.3.	Assembled ballistic bomb calorimeter, with a galvanometer					
	in the background107					

# LIST OF SYMBOLS AND ABBREVIATIONS

ANOVA	Analysis of variance
DEFRA	Department of Enviroment, Food and Rural Affairs
MOE	Modulus of elasticity
MOR	Modulus of rupture
FNdfa	Fraction of nitrogen derived from the atmosphere
SE	Standard error
RLD	Root length density
CV	Calorific value
AC	Ash content
FVI	Fuel value index
MPa	Mega Pascal

#### **CHAPTER 1.0**

#### INTRODUCTION

#### 1.1 Alnus rubra (red alder)

*Alnus rubra* Bong, belongs to the family *Betulaceae* (Johnson, 1967; Hosie, 1990). It is an early colonizing species after disturbances such as fire and clear felling (Binkley *et al.*, 1994). The tree is important for its nitrogen fixation and commercial timber. It grows fast and normally covers a site quickly. Red alder contributes to the nitrogen balance in the soil through root symbiosis with nitrogen fixing endophyte *Frankia* (Binkley *et al.*, 1994), which is a sporulating, filamentous actinomycete. For this reason, the tree species is classified as an actinorhizal plant (Rojas *et al.*, 2001). Annual nitrogen fixation rates for red alder have been estimated at 30 to 150 kg ha<sup>-1</sup>year<sup>-1</sup> in a wide range of ages (Binkley *et al.*, 1994).

In hospitable sites and forest, the tree has one straight stem with a slight taper (Hosie, 1990; Johnson 1967). However, when grown in the open, it is known to have a wide crown with branches starting close to the ground resulting in a conical shape (Hosie, 1990). The root system is reported to be shallow and wide spreading (Hosie, 1990). Red alder is found naturally growing in the forest region of the British Columbia in Canada and in the southern California coast, along the Pacific coast of USA to the southeast Alaska (Johnson, 1967; Hosie, 1967). It is found within a hundred miles from the ocean (Hosie, 1990).

Red alder is the largest alder tree and has the capacity to reach more than 20 metres in height and diameters of up to 60 cm (Hosie, 1990). The tree is known to mature at 60 to 70 years and rarely survives beyond 100 years (Niemiec *et al.*, 1995).

*Alnus rubra* is a nitrogen-fixing tree species commonly found in North America but adapted to grow in other parts of the world (Johnson, 1967). It is best developed in the alluvial flats, mesic and riparian zones. Red alder grows in habitats with western red cedar/western hemlock normally in alluvial flats, riparian and mesic zones and across differing elevation (Johnson, 1967; Rojas *et al.*, 2001). It is considered to be a major source of nitrogen for ecosystems where it is found.

#### 1.2 Potential uses of red alder

The potential of red alder to improve soil fertility, when grown with other trees in forestry plantations, has been reported by Binkley *et al.* (1994) and Wheeler *et al.* (1986). Experiments have been carried out to evaluate its contribution to soil fertility when grown with maize by Seiter *et al.*, (1999). The tree can potentially be used to increase soil fertility in a silvopastoral system though there is no report in the literature. A silvopastoral system is a type of agroforestry system where trees are grown on pasture, which is grazed by livestock. Red alder when grown to improve soil fertility in silvopastoral system may be managed to provide good quality timber at the end of the rotation. Red alder has been used to make pulp, furniture, cabinets, pallets and has also been reported to be satisfactory for oriented strandboard, studs and turned products (Lei *et al.*, 1997). The off-cuts from the mill and the branches may be used for firewood. The wood can also potentially be used as biomass energy in the production of

electricity. The above-mentioned uses have, however, not been studied in red alder when grown in silvopastoral system. Consequently, the aim of the present study is to evaluate the above aspects.

#### 1.2.1 Use of red alder for soil fertility improvement and maintenance

Soil fertility is one of the major factors limiting crop production (Sanchez and Logan, 1992). Commercial fertilizers, which may be used to correct this problem are expensive, may not be accessible and may cause pollution problems (Ladha and Reddy, 2003; Sprent and Sprent, 1990). Possible interventions to improve soil fertility include the incorporation of multipurpose nitrogen fixing tree species to provide nutrients through litter fall and nitrogen fixation (Govindrajan *et al.*, 1996; Roy *et al.*, 2003). Leaf nutrition content, nitrogen fixation properties and branching patterns are some of the important factors considered in the choice of multipurpose trees for soil fertility improvement (Akinnifensi *et al.*, 1999).

Improvement of soil fertility in agroforestry is a result of increased above and below ground organic matter inputs, nutrient cycling and nitrogen fixation depending on the tree species (Mafongoya *et al.*, 1998; Pandey *et al.*, 2000; Young, 1997). Agroforestry systems increase or maintain organic matter levels of soils (Young, 1997). Consistently increased organic matter promotes soil aggregation, which provides stability to the soil, improves soil structure and the microbial biomass (Killham, 1994). Nutrients become available to the crops through decomposition of tree prunings and litter (Mafongoya *et al.*, 1998). Red alder grown with other crops has been shown to have the capacity to improve levels of soil nitrogen and organic content (Binkley *et al.*, 1994; Seiter *et al.*, 1995; Tarrant and Trappe 1971).

Nitrogen fixation is an important part of recycling nitrogen in the ecosystem (Postgate, 1998). Nitrogen fixed through nodules found in leguminous and actinorhizal plants contributes to the total soil nitrogen. Non-nitrogen fixing plants benefit from the association when parts of the nitrogen fixing plants die and decompose in the soil (Killham, 1994). The mineralised nitrogen can then be available for absorption by the non-nitrogen fixing plants (Mafongoya *et al.*, 1998). Although *Alnus rubra* is a nitrogen-fixing tree species, its contribution to soil fertility improvement and maintenance in silvopastoral system has never been evaluated. No research has been carried out to quantify the amount of nitrogen fixed by red alder in silvopastoral systems using the nitrogen-15 natural abundance method.

#### 1.2.2 The potential of red alder to provide good quality timber

Wood quality is defined by the specific end-use of the solid wood in question. Whatever the requirements, the density, strength and appearance of wood are affected by management or silvicultural treatments applied to a stand. Aspects such as stocking density, intensive silviculture and site quality will finally affect the quality of the wood produced (Jozsa and Middleton, 1994; Lei *et al.*, 1997). Aspects on timber quality and strength however, have not been studied in red alder when grown in a silvopastoral system.

#### 1.2.3 Fuelwood potential of red alder

Biomass energy is a renewable resource and a large majority of people in the world depend on biomass energy. Biomass energy is known to provide about 14% of total energy worldwide, being the highest energy source in the majority of the developing world (Kataki and Konwer, 2002). Trosesso (2002) however, specified a lower value of seven percent. A greater proportion of biomass energy is used in developing countries and about 76 percent is obtained from charcoal and firewood (Fuwape, 1993; Trosesso 2002). The use of biomass for fuel ranges from about 3% in Australia to as high as 33% in China (Fuwape, 1993). In the developing world, wood energy represents approximately 15 percent of the total energy consumed (Trosesso 2002). Figure 1.1 below shows the distribution of wood energy consumption by regions of the world.

# Figure 1.1 Distribution of consumption of wood energy by regions of the world (Tresesso, 2002).



The British government supports the use of renewable energy to provide electricity and heat. This is part of its strategy for promoting sustainability and reducing the impact of climate change. The aim is for the power suppliers to source 10% of their power from renewable resources, which include biomass by 2010 (Upreti and van der Horst, 2004). Funding for biomass is made available from several sources such as Bio-energy Capital Grant Scheme, Woodland grant scheme, Department for Environment, Food and Rural Affairs (DEFRA) Energy crop scheme to mention a few to support renewable energy initiative (DEFRA, 2002). However, information on the energy values and fuel value index (FVI) of red alder when it is grown in silvopastoral systems is lacking. These need to be assessed in order to determine the potential of red alder for use in bio-energy production especially in the above-mentioned schemes in addition to willow, poplar and fibre plants in the UK.

#### 1.3 Aim and objectives of the study

The aim of the present study was to assess the potential multipurpose uses of red alder tree through quantification of nitrogen fixed by the species, and its contribution to Nbalance; and by evaluating other potential uses of its wood such as timber and fuelwood when it is grown in a silvopastoral system in comparison with a forestry system.

#### **Specific objectives**

- To study nitrogen fixation using nitrogen-15 natural abundance method in stands of *Alnus rubra*.

- To study the dynamics of fine roots and root nodules in red alder and their contribution to soil organic matter and the nitrogen-balance under silvopastural systems.
- To study the timber quality of red alder wood in comparison with sycamore.
- To study the firewood value of red alder in comparison with sycamore.

#### 1.4 Hypothesis of the experiments carried out in this study;

- 1. Nitrogen fixation, roots and root nodule dynamics of red alder contribute to increased nitrogen accumulation in the soil and to efficient nitrogen cycling in a silvopasture system.
- 2. Red alder trees grown at wide spacing in silvopasture have a different timber quality from those grown at close spacing in a forestry system.
- Red alder wood has good energy properties and has high potential for use in the generation of bio-energy.

## 1.5 Thesis Structure

The thesis has six chapters. Chapter 1 introduces background, objectives and hypothesis of the study, description of the study area, the thesis structure and research design. Chapter 2 reviews literature on soil fertility, nitrogen fixation, measurement of nitrogen fixation, fine root distribution, dynamics and turnover as well as timber and fuelwood quality of red alder and other related tree species. Chapter 3 reports the results of the present study on rate of nitrogen fixation in red alder and its contribution to soil fertility improvement and maintenance. Chapter 4 presents results of field study on fine root distribution and dynamics of red alder in silvopastoral systems in comparison with in forestry plots. Chapter 5 reports the results of investigation carried out to assess the wood quality of red alder in comparison with sycamore. Chapter 6 presents a general summary, conclusions and recommendations.

#### 1.6 Study site

The experiment was carried out in a 10 year-old red alder and sycamore silvopastoral experiment site at the University of Wales, Henfaes farm (Figure 1.2). It is one of the six experimental sites within the UK agroforestry network. The network sites were established to investigate the potential of silvopastoral agroforestry systems on UK farms. The six sites were established by the University of Wales, Bangor, the Macaulay Landuse Research Institute (Scotland), the Institute of Grassland and Environmental Research, and the Department of Agriculture of Northern Ireland (Teklehaimanot and Sinclair, 1993).

The network sites have sycamore (*Acer pseudoplatanus*) as a common tree species planted at 100, 400 and 2500 stems ha<sup>-1</sup> (forestry control) and have pure pasture as agricultural control. At the Hanfaes site red alder (*Alnus rubra*) has been introduced as an additional tree species planted at 400 stems ha<sup>-1</sup> and as a forestry control at 2500 stems ha<sup>-1</sup>.

The Henfaes research centre is located in Abergwyngregyn, Gwynedd, 12 km east of Bangor. The climate is hyperoceanic with an annual rainfall of about 1000 mm. The soil is mainly a fine loamy brown earth over gravel classified as Rheidol series (FAO-UNESCO, Dystric Cambisol). The parent material consists of postglacial alluvial deposits from the Aber River, comprising Snowdonian rhyolitic tuffs and lavas, microdiorites and dolerite in the stone fractions and lower Plaeozoic shale in the finer fractions. There is a shallow slope of about 1-2° on the deltaic fan with a northwest aspect at an altitude ranging from 4 to 14 metres above sea level. The water table of the area is located at 1 m to 6 m depth (Teklehaimanot and Martin, 1999; Teklehaimanot *et al.*, 2002). The rainfall and temperature of the site during the two years of the present study period 2002-2003 are given in Figures 1.3 and 1.4 respectively.

#### 1.7 Research design

Four treatments were used in the present research (Figure 1.2), and these included red alder 400 stems ha<sup>-1</sup> (agroforestry), red alder 2500 stems per ha<sup>-1</sup> (forestry control), sycamore 400 stems ha<sup>-1</sup> (agroforestry), sycamore 2500 stems ha<sup>-1</sup> (forestry control). Other plants growing within the area were also used as reference plants in the <sup>15</sup>N natural abundance studies. These included *Salix spp, Acer pseudoplatanus* and *Prunus spinosa*. The height and diameter of red alder and sycamore in the four treatments in 2003 are given in Table 1.1. Red alder had high and significant diameter and height than sycamore (P, 0.05). Diameter of agroforestry red alder plots was higher than that of forestry alder. Sycamore forestry yielded significantly taller trees than agroforestry and the diameter at breast height was the same (P, 0.05).



Figure 1.2. Map of the blocks and plots of sycamore and red alder at Henfaes. Each of the large plots with trees is 1 ha in area (Teklehaimanot *et al.*, 1997)



Figure 1.3: Average monthly rainfall (mm) at Henfaes research station in year 2002 and 2003.



Figure 1.4 Mean monthly temperatures (°C) at Henfaes research station in year 2002 and 2003.

Table 1.1Average diameter at breast height and tree height (cm) in<br/>agroforestry and forestry of sycamore and red alder plots in<br/>different blocks in 2003.

	Red alder					Sycamore			
	Silvopasture		Forestry		Silvopasture		Forestry		
Block	DBH	Height	DBH	Height	DBH	Height	DBH	Height	
Ι	16.7	847.8	13.4	963.3	9.45	617.9	8.8	739.0	
II	18.7	877.3	10.4	672.0	11.75	684.0	9.8	750.0	
ш	16.8	837.2	11.9	901.0	11.16	670.9	11.3	843.6	
Mean ±SE	16.7±1.6	854.1±8.5	11 <b>.9</b> ±1.7	845.0±6.7	9.9±1.6	657.6±9.8	10.0±1.4	777.5±9.3	

#### **CHAPTER 2.0**

#### LITERATURE REVIEW

#### 2.1 The Biology of Alnus rubra

#### 2.1.1. Taxonomy and systematics

Red alder is a member of the *Betulaceae* family (Johnson, 1967; Hosie, 1990; Niemiec *et al.*, 1995). Red alder is also called Oregon alder, western red alder and the Pacific coast alder. Red alder is one of the most common and important hardwood trees in the Pacific Northwest (Niemiec *et al.*, 1995) for soil fertility, timber and other products.

The number of species in this genus is not well known but about 30 species are known and found growing in north temperate regions and in central and South America. Some members of this family grow as shrubs for example mountain and sitka alder, whereas others such as red alder and European alder grow as large trees (Hosie 1990).

#### 2.1.2. Description of Alnus rubra

#### 2.1.2.1 Seedlings

Germination and growth of red alder occur well on moist mineral soil with full sunlight. Germination is epigeal. Since seeds are very small, it is important that the radicle reaches moist and nutritious substrate after germination to allow for successful establishment. While seedlings will tolerate partial sunlight, full sunlight is required for normal development and growth. On young trees, the bark is smooth and conspicuously patterned by long horizontal markings (Plate 2.1) (Harrington, 1990 and Niemiec *et al.*, 1995).



Plate 2.1 Red alder stem showing white lenticels and epicormic branches

#### 2.1.2.2 Habitat and form of mature trees

A mature tree of red alder grows up to 30 to 40 metres in height and 55 to 75 cm in diameter. The tree matures in 60 to 70 years and rarely lives above 100 years in age (Niemiec *et al.*, 1995). When growing in forest stands, red alder develops a clear and slightly tapered bole with narrow and domelike crown. Trees grown in the open tend to have broad conical crowns with highly tapered boles and often times with large forks and branches (Niemiec *et al.*, 1995). There is a red pigment on the bark that may stain freshly cut wood especially when it is wet. The wood is light, soft and moderate in strength (Hosie, 1990).

#### 2.1.2.3 Foliage

Alder has leaves that are alternate and grow singly in two or three ranks along the twigs. They are simple, usually oval and toothed and with two sizes of teeth (Plate 2.2) Leaves of red alder are oval to rhombic in shape, tapered from the middle to both ends (Hosie, 1990). They are dull green on the upper side, greyish beneath and hairy on the veins below with large lobe-like teeth with six inrolled teeth on each. Veins are impressed above with veinlets between veins forming a ladder-like pattern (Hosie, 1990).

Alder shoots have leaf scars that are raised on the thick parts of the twig with semicircular or triangular shapes with tree bundle scars. Alder twigs are slender to moderately stout with a triangular pith, and with terminal and lateral buds. Lateral buds are normally stalked and both types of buds have two to three overlapping scales or scales that meet along the edges (Hosie, 1990)

#### 2.1.2.4 Rooting habit

Red alder is reported to have an extensive fibrous root system. Root growth of the seedling is rapid and wide spreading and the tree attains large woody roots at an early age. Roots are known to commonly have ectomycorrhizae. The tree is importantly known to have roots that have root nodules that fix nitrogen (Harrington, 1990).

15

#### 2.1.2.5 Flowering and fruiting

Red alder reaches sexual maturity in 3 to 4 years of age. Dominant trees usually produce seed at 6 to 8 years of age. The tree is monoecious, and has separate male and female catkins, which develop in the previous years twigs. Staminate catkins occur in pendulous clumps and in later winter these elongate, changing from green to brown and from 2 to 3 cm to 7 to 8 cm. Red alder also has pistillate catkins that occur in clumps but borne upright. Pistillate catkins are 5 to 8 cm and reddish green when receptive. Flowering occurs in late winter or early spring and is followed by pollen shedding within a few days (Harrington, 1990; Niemiec *et al.*, 1995).

The fruit is a nutlet with a wing on each side and ripens in one season. The wing may appear to be circular, surrounding the fruit but in some cases does not exist. The fruits are borne on woody scales that are attached to a stiff central stem which form an oval fruiting body. In the autumn the scales open to release the fruit but the fruiting body remains in the tree (Plates 2.3 and 2.4) (Hosie, 1990).



Plate 2.2 Leaves of red alder showing the in-rolling tooth and tooth-lets.



Plate 2.3. Red alder catkins.



Plate 2.4. Catkins of red alder from the previous year.

#### 2.1.2.6 Phenology and life cycle

Red alder is a deciduous tree. Flowers of this tree begin to form in the spring before the leaves expand. Flowering in the Oregon and Washington have been reported to begin in late February and continuing into early May. Fruits ripen in late August and September (Haeussler, 1991). Seeds are then dispersed during the autumn and winter. Red alder tree is known to lose its leaves first in the autumn. The majority of the leaves are lost in October and by the beginning of November all leaves are lost (Haeussler, 1991).

#### 2.1.2.7 Seed biology and dispersal

Red alder is prolific and consistent in producing seed. It produces seeds annually with bumper crops every three to four years. Seeds of red alder are small, winged nuts borne in pairs and are on bracts of woody, cone like strobili. The strobili are 11 to 32 mm long and 8 to 15 mm wide. Dispersal of seed occurs in late September and most seeds are shed during late fall and winter. Since seeds are very light, dissemination by wind is effective. The seeds can be carried over long distances and this makes natural regeneration easy over an area (Harrington, 1990).

#### 2.2 Ecology

#### 2.2.1 Geographical distribution and range

Red alder extends from the southern parts of Alaska ( $60^{\circ}$  N) to southern California ( $34^{\circ}$  N) (Niemiec *et al.*, 1995). It is found growing within 200 km of the ocean (Harrington, 1990; Niemiec *et al.*, 1995). The tree is found at elevations, below 750 meters above sea level throughout the coast and the north cascades ranges. It is restricted to riparian areas and moist sites further south. Several isolated populations are known to exist in northern Idaho in the US (Niemiec *et al.*, 1995).

#### 2.2.2 Ecological requirements

The typical climate in the geographical range of red alder is mild and humid. Annual rainfall ranges from 400 to 5600 mm (Harrington, 1990; Niemiec *et al.*, 1995). Most of the precipitation in this area occurs as rain in winter with summers that are generally cool and dry (Harrington, 1990; Niemiec *et al.*, 1995). Red alder is known to grow well when annual rainfall is above 1000 mm or when roots have access to ground water. On the other hand the tree species does poorly under droughty conditions due to inadequate

rainfall, low moisture holding capacity of the soil or high evapotranspiration, combined or singly (Niemiec *et al.*, 1995).

Tolerable temperatures for red alder occur in the range of -30 to 46.1° C. Risks of mortality and damage from sunscald, heat and drought are high on the southern aspect especially inland on steep slopes (Niemiec *et al.*, 1995). Severe freezing and unreasonable frost hazards can also limit the performance of red alder resulting in poor quality stands. Periodic high winds can also reduce growth of red alder and such areas should be avoided.

Red alder is found in a wide range of soils but the most productive stands occur on deep well-drained loams and sandy loams soils especially those of marine and alluvial origin. Abundant soil moisture is important during the growing season. The plant will tolerate poor drainage and occasional floods during the growing season where soils prone to prolonged waterlogging may not be suitable. Soils with very low water holding capacity may result in excessive drought (Niemiec *et al.*, 1995).

#### 2.2.3 Plant associations

Red alder has been reported to grow in both pure and mixed stands (Harrington, 1990). Pure stands are normally found in stream bottoms and lower slopes. This tree is however widely distributed in mixed stands in the cover type red alder and as a major component in most other cover types of the North Pacific cover types (Harrington, 1990). Red alder is commonly found growing with Douglas fir (*Pseudotsuga menziesii*), western hemlock (*Tsuga heteropylla*), western red cedar (*Thuja plicata*),
Willow (*Salix species*), big leaf maple (*Acer macrophyllum*), Sitka spruce (*Picea sitchensis*) and grand fir (*Abies grandis*) (Harrington, 1990; Johnson, 1967).

Trees such as Pacific dogwood (*Cornus muttallii*), red wood (*Sequoia sempervirens*) are some of the examples of trees that are occasionally associated with red alder (Harrington, 1990). Shrubs including vine maple (*Acer circinatum*), red and blue alder (*Sambucus callicarpa, S. cerulean*) and salal (*Gaultheria shallon*) are commonly associated with red alder in the Pacific Northwest. Some herbaceous plants have also been reported to grow in association with red alder, examples of these being stinging nettle (*Urtica dioica*), Pacific water parsley (*Oenanthe sarmentosa*), blackberries (*Rubus laciniatus, R. leucodermis*) and California dewberry (*R. ursinus*)

#### 2.3 Silviculture and management of red alder

Since red alder was declassified as a weed and recognised as a good timber species, several silvicultural techniques have been applied throughout the life cycle of this tree. The tree is regenerated from seed, vegetative sprouts and through artificial planting (Niemiec *et al.*, 1995). Site preparations, planting and management of the stand through thinning and other silvicultural techniques are also undertaken (Niemiec *et al.*, 1995)

#### 2.3.1 Regeneration from seed

Seed of red alder is very light in weight, about 800 to 3000 seeds  $g^{-1}$  and will normally disseminate well in the site. Establishment from seed generally requires open

conditions and bare mineral soils (Harrington, 1990). Disturbance on the site, such as fire scratching of the ground normally results in plentiful germination of alder seed. The light environment resulting after disturbances is also reported to stimulate germination of red alder seeds (Haeussler and Tappeiner II, 1993).

#### 2.3.2 Regeneration from vegetative sprouts

*Alnus rubra* will sprout vigorously after cutting. Sprouting is feasible on young trees less than 10 years old with coppices of 4 to 6 years old cycles. Older trees do not sprout easily after cutting (Harrington, 1990; Niemiec *et al.*, 1995). Cuttings treated with root hormones at 22° C to 25° C have been successfully rooted (Harrington, 1990). However, cuttings that have not been treated have low success (Niemiec et al., 1995). Success has also been achieved with mound layering (Harrington, 1990). The seedlings are first coppiced and the base of the sprouts are covered with soil when they are a few months old (Harrington, 1990).

#### 2.3.3 Regeneration from planting with seedlings

Regeneration can be carried out using seedlings and this allows flexibility in site selection, and avails control on spacing and seed source compared with other methods (Niemiec *et al.*, 1995). Such seedlings will have advantage and will compete well with other vegetation. Seedlings should be 30 to 90 cm in height with basal diameter of at least 4 mm. The seedlings should be stocky rather than thin, with healthy buds or branches in the entire length of the stem, with a full undamaged fibrous root system and free of diseases (Niemiec *et al.*, 1995). Success of seedling in the site should consider

other factors such as extreme weather and other hazards, good site preparations and planting practices (Niemiec *et al.*, 1995).

#### 2.3.4 Site preparation and vegetation management

Red alder requires site preparation in areas where there is competing vegetation and where there is a lot of debris on the ground. Broadcast burning often provides adequate site preparation where there is a lot of slash and high shrub cover. Herbicides may also be applied before planting and such can make a difference between success and failure in establishment. Even distribution of mineral soil is important when seeds are to be used for regeneration. Such conditions can be provided by mechanical scarification, broadcast burning or piling and burning. Even germination of seeds may be encouraged by even scarification of the soil (Niemiec *et al.*, 1995).

Red alder stands establish at high densities and competition normally results in selfthinning with slow growth in diameter. Management of lower initial densities can result in increased diameter growth compared with unmanaged stands. It is advisable to remove smaller trees to favour the codominant and dominant trees. However crowding is important to maintain dominance, to reduce branching, forking and stem taper (Niemiec *et al.*, 1995).

Mixing red alder with other tree species is of interest since the tree is nitrogen fixing and adds organic matter to the soil. However, mixed cropping may be made difficult because red alder grows rapidly and may suppress growth of conifers (Niemiec *et al.*, 1995). Managing tree mixtures with red alder may include delaying of the planting of red alder for 3 to 6 years, maintaining a low proportion of red alder in the stand and managing mixtures in small patches similar to natural mixtures (Niemiec *et al.*, 1995).

#### 2.4 Uses of red alder

#### 2.4.1 The role of red alder in soil fertility improvement

Soil fertility is defined as the ability of soil to maximise plant productivity (Campbell, 1998). The decline in soil fertility is often due to overgrazing and deforestation (Campbell 1998). Nutrient losses in agricultural systems are due to volatilization, losses due to leaching, product removal, losses due to non-liable soil pools and losses due to erosion (Campbell, 1998; Laegreid *et al.*, 1999).

Soil fertility is an important component of crop production since nutrients from the soil are the major basis for food production (Laegreid *et al.*, 1999). Nitrogen is considered to be the most important nutrient supplied to the crop from the soil (O'Hara, 1998). It is essential for all life processes in the plant (Laegreid *et al.*, 1999). Often the availability of a suitable source of nitrogen is considered a major factor limiting crop productivity (Laegreid *et al.*, 1999; O'Hara, 1998).

Soil nutrients are used up during continuous cropping. Nitrogen, in particular, is the most mobile nutrient that is often lost through volatilisation by burning, for example, of crop residue or forest fires (Campbell, 1998). Losses of nitrogen also occur due to removal of products from a site such as harvesting and in cut and carry systems (Dulormne *et al.*, 2003, Campbell, 1998). Removal usually limits localised nutrient

cycling (Campbell, 1998). Nitrogen in the form of nitrates tends to be prone to leaching in very wet soils (Boddey, *et al.*, 2000; Laegreid *et al.*, 1999). Losses through ammonia volatilisation and denitrification also occurs (Boddey *et al.*, 2000). This situation can be corrected by addition of chemical and organic fertilizers. Chemical fertilizers, however, have disadvantages in that they may not be available in some areas, are expensive and can cause pollution to the environment. Inclusion of trees in farming systems may benefit the system through the addition of litter material. The mineralising litter material improves both the nutrient and the organic matter content of the soil (Teklehaimanot and Anim-Kwapong, 1996; Garcia-Montiel and Binkley, 1998). If a nitrogen-fixing tree is included in the system, additional nitrogen will be added to the soil due to its nitrogen fixing capacity (Govindrajan *et al.*, 1996).

The use of red alder to increase soil nitrogen content has been reported (Binkley *et al.* 1994; Seiter *et al.*, 1999; Tarrant and Trappe, 1971). In the Pacific Northwest the tree is grown with other non-nitrogen fixing trees in order to increase the nitrogen content of the soil (Binkley *et al.*, 1994). Research carried out using other alder species showed an improvement in the soil nutrition and structure (Hurd *et al.*, 2001; Myrold and Huss-Danell, 2003). Seiter *et al.*, (1999) reported increased nitrogen contents in soils where maize was grown with red alder compared to low nitrogen amounts in a maize monocrop. The increased rates of nitrogen in the soil were attributed to the high capacity of red alder to fix atmospheric nitrogen (Seiter *et al.*, 1999).

#### 2.4.1.1 Fine root turnover and fertility improvement

Fine root distribution and turnover are the mechanisms through which trees improve soil fertility (Joslin and Henderson, 1987; Kern *et al.*, 2004; Hendrick and Pregitzer, 1993). However, there is no information in the literature on roots of red alder and how they influence soil fertility.

Distribution and architecture of roots in the soil depends on a number of chemical, physical and biological factors (Fogel, 1983; Curt, *et al.*, 2001). Root systems change in response to the environment depending on the ease of obtaining necessary resources from the environment. The pattern of root development is the same as that of shoot and generally follows a sigmoidal shape. Generally there is low root death early in the growing season when conditions are favourable. As the season progresses, the natural senescence patterns follow and environmental conditions have an effect on mortality of fine roots and thus affecting the distribution in the growing gradient (Gregory, 1994).

It has been found that mobile and immobile nutrients influence root growth, which in itself also follows the moisture content of the different soil layers. Management option such as shoot pruning has an effect on root distribution and root amount (Bayala *et al.*, 2004). Root and shoots generally follow a functional equilibrium (Jones *et al.*, 1998; Gregory, 1994). Like other plant organs, fine roots are born; they age and die (Eissenstat and Yanai, 1997; Yavitt and Wright, 2001). The below ground biomass of the roots is highly dynamic with formation of new roots, death of old roots and decomposition of dead roots. In forests, between 30 and 90% of fine roots are lost and replaced annually (Killham, 1994).

Fine root dynamics is important for water absorption, contribution to net primary productivity and turnover for nutrient cycling (Joslin and Henderson, 1987; Munoz and Beer, 2001) (Figure 2.1). Fine root dynamics like fine root distribution is affected by biological, chemical and physical factors in the environment (Black *et al.*, 1998; Lopez *et al.*, 1998; Teskey and Hinckley, 1981; Tierney *et al.*, 2003). However, the understanding of the dynamics and control by soil conditions such as temperature, moisture, structure, resource levels and grazing are limited (Tierney *et al.*, 2003; Yavitt and Wright, 2001). Temperatures are strong cues for fine root dynamics in the temperate region, where root mortality occurs primarily in winter (Pregitzer *et al.*, 2000; Ruess *et al.*, 1998; Yavitt and Wright, 2001). In tropical dry areas, however, seasonal changes through the year result in changes in density and growth of roots. Roots mostly grow in the rainy season and die during the dry periods. During the dry period, root growth may be impeded by the hard clay soils. Roots also grow deep into the soil to capture moisture. Leaf flushes at the beginning of the growing period also act as a cue to fine roots in the tropics (Yavitt and Wright, 2001).

Fine root turnover in forest ecosystems is important and occurs at 2 to 5 times the rate that occurs above ground. Killham (1994) and Fogel (1983), stated root turnovers of 4-5 times greater than above ground litter. Turnover of roots is not only determined by the species, the soil environment is also critical. Soils that are poor tend to support roots with longer turnover times than those in nutrient rich soils (Killham, 1994). Distribution of fine roots in grey alder and *Discaria* was reported to be decreasing with distance away from the tree base (Rytter, 1989; Valverde and Wall, 2002).

#### 2.4.2 Nitrogen fixation and soil fertility improvement

#### i) Systems of Nitrogen fixation

Although nitrogen is one of the most widely distributed elements in nature being present in the atmosphere, lithosphere and the hydrosphere, there is very little of it available for plants in the soil (Merschner, 1995; Tisdale *et al.*, 1993). In the soil, nitrogen is in the form of  $NO_3^-$  or  $NH_4^+$  ions and is mobile circulating between the atmosphere, the soil and living organisms. Factors and processes involved in the turnover are physio-chemical such as the exudation from the roots and biological such as in the action of microbes in the soil e.g. *Nitrosomonas* (Tisdale *et al.*, 1993). Nitrogen is fixed from the inorganic form in the atmosphere and converted to the organic form through nitrogen fixation (Elkan, 1992). This is carried out by free-living microbes as well as symbiotic microorganisms in the soil, lightning and by the Harber-Bosch process which is high energy consuming process (Elkan, 1992; Sprent and Sprent, 1990) (Figure 2.1). Nitrogen is also deposited from pollutants that originate from fossil fuels, for example from car exhaust fumes.

The Harber-Bosch process is an industrial chemical process that facilitates the reaction of nitrogen and hydrogen under high pressures and moderately high temperatures and is used in the manufacture of nitrogen fertiliser (Merschner, 1995). This process is carried out under pressures ranging from 200-400 atmospheres and temperatures ranging from 400° to 650°C. A catalyst made mostly from iron enables the reaction to be carried out at relatively lower temperatures than it would otherwise be possible. The production of ammonia in this process reaches an equilibrium point but the continued production of ammonia is favoured as the product is continually cooled and removed.



Figure 2.1 Global nitrogen cycling (Laegreid *et al.*, 1999)

The costs of fossil energy and the worldwide demand for nitrogen fertilizer used for food production are the reasons for interest in biological nitrogen fixation (Merschner, 1995). Organisms that fix nitrogen all belong to the group called the prokaryotes. All these organisms, which reduce dinitrogen to ammonia, use the enzyme complex nitrogenase (Sprent and Sprent, 1990). Such organisms may be free-living or living in association with plants (Merschner, 1995; Postgate, 1998). Table 2.1 below shows the different organisms that exist as free living organisms in the soil. Some of the organisms listed in Table 2.1, however may be found in association with plants (Postgate, 1998). Many of the organisms need the supply of reduced carbon e.g. *Azobacter*, while others can reduce carbon dioxide (Sprent and Sprent, 1990).



## Figure 2.2 Systems of nitrogen fixation, energy source and estimates of amounts fixed (Merschner, 1995).

A large amount of nitrogen is fixed by miroorganisms living in symbiotic association with plants (Postgate, 1998). The symbiotic relations have been reported in legume and actinorhizal plants (Postgate, 1998; Schwencke and Caru 2001; Sprent and Sprent, 1990). Microorganisms living in association with plants benefit by acquiring carbon from the plant and at the same time supplying the plant with fixed nitrogen.

## Table 2.1. List of genera of microbes, which include nitrogen-fixing species

### (Postgate, 1998).

Microbes	Genus or type	Species (examples)
Strict anaerobes	Clostrodium	C. pasteurianum, C. butyricum
	Desulfovibrio	D. vulgaris, D. desulfuricans
	Methanosarcina	M. barkeri
	Methanococcus	M. thermolithotropicus
Facultative (aerobic when	Klebsiella	K. pneumoniae, K. oxytoca
not fixing nitrogen)	Bacillus	B. polymixa, B. macerans
	Enterbacter	E. agglomerans
	Citrobacter	C. freundii
	Escherichia	E. entermedia
	Propionibacterium	P. shermanii, P. petersonii
Microaerophiles (Normal	Xanthobacter	X. flavus, X. autotrophicus
aerobes) when not fixing	Thiobacillus	T. ferro-oxidans
nitrogen)	Azospirillum	A. lipoferum, A. braziliensis
	Aquaspirillium	A. perigrinum, A. fascicilus
	Methylosinus	M. trichosporum
	Bradyrhizobium	A.japonicum
	Herbaspirillium	H. seropedicae
	Burkholderia	B. brasilense
Aerobes	Azobacter	A. chroococuum, A. vinelandii
	Azotococcus	A. agilis
	Azomonas	A. macrocytogenes
	Beijerinckia	B. indica, B. fluminis
	Derxia	D. gummosa
Phototrophs (anaerobes)	Chromatium	C. vinosum
	Chlorobium	C. limicola
	Thiopedia	
	Ectothiospira	E. shapovnikovii
Phototrophs (facultative	Rhodospirillum	R. rubrum
phototrophs	Rhodopseudomonas	R. palusstris
(microaerophiles)	Plectonema	P. boryanum
	Lyngbya	L. aestuarii
	Oscillatoria	
	Spirulina	
Phototrophs (aerobic)	Anabena	A. cylindrical, A. inaequalis
	Nostoc	N. muscorum
	Calothrix	
	(7 other genera of	
	heterocystous blue-green	
	algae)	
	Gloeootheca	
		G. alpico

#### ii) Nitrogen fixing plants

There are two main types of symbiosis that occur between nitrogen-fixing bacteria and plants (Marschner, 1995; Wall, 2000). One occurs between *Rhizobium* and legume plants and the second one between *Frankia* and actinorhizal plants (Gualtieri and Bisseling, 2000; Wall, 2000). In each case, a new plant organ is formed and the bacteria differentiate within such, express the enzyme nitrogenase and fix nitrogen into ammonia (Wall 2000). The fixed compounds are then assimilated and transported to the other parts of the plant (Wall, 2000).

The symbiosis between legumes and *Rhizobia* occurs in more than 1700 species of the family Fabaceae in the sub-families Mimosoideae, Ceaesalpinoideae and Papilionoideae (Wall, 2000). The gram-negative bacteria in the family of Rhizobiaceae are *Rhizobium, Azorrhizobium, Sinorhizobium, Bradirhizobium* and *Mesorhizobium* (Binkley *et al.*, 1994; Sprent and Sprent, 1990; Wall, 2000). Legume nodules are characterized by a central infected tissue surrounded by nodule parenchyma and peripheral vascular bundles (Wall, 2000).

The symbiosis in actinorhizal plants, on the other hand, occurs on more than 220 plants, which are symbiotically associated with the filamentous actinomycete *Frankia* (Binkley *et al.*, 1994; Wall 2000). Examples of actinorhizal plants are found in genera *Alnus*, *Casuarina*, *Ceanothus*, *Eleagnus*, *Myrica*, *Hippophae*, *Prushia* and *Dryas* (Postgate, 1998). The actinorhizal nodules are characterized by central vascular bundle and peripheral infected tissue surrounded by cortical tissue parenchyma (Wall, 2000).

#### iii) Nitrogen fixation by red alder

Nitrogen fixation by *Alnus rubra* is accomplished by the symbiotic actinomycete of the *Frankia* genus living in the root nodules of the tree (Binkley *et al.*, 1994). Nodulation of *Alnus* begins when *Frankia* penetrates root hairs and enters the cortical cells of roots Binkley *et al.*, 1994). *Frankia* resides in enlarged cortical cells and then produces specialised cells called vesicles that are the site of the nitrogen-fixing enzyme, nitrogenase and nitrogen fixation. The alder roots, which are perennial, then continue to grow in size; where at the same time individual nodule lobes divide and produce a coral-like morphology (Binkley *et al.*, 1994) (Plate 2.5).



Plate 2.5. Red alder root nodules showing orange-yellow live parts and dead parts (black in colour).

#### iv) Mechanism of infection

*Frankia* is reported to infect host plants through two ways namely, (1) root hair infection or intracellular infection and (2) intercellular infection (Gualtieri and Bisseling, 2000; Obertello *et al.*, 2003; Schwencke and Caru, 2001). Figure 2.3 shows the stages through the infection process. The root hair infection process occurs in *Alnus, Myrica, Comptonia* and *Casuarina*, while the intercellular penetration is reported to occur in *Elaeagnus, Ceanothus* and *Cercocarpus* speices (Schwencke and Caru, 2001).

In the intracellular penetration, the first stage occurs by root curling and branching. The deformation is reported to occur mostly as a multiple swelling or lobbing of root tips with increased mucilage at the root surface favouring the binding of *Frankia* cells to the wall of the root hair (Schwencke and Caru, 2001). The cortical cells are induced to divide in response to the infection, therefore, producing a small protuberance called the prenodule (Schwencke and Caru, 2001). The pericycle opposite the protoxylem is mitotically activated to form a nodule primordium from which lateral root-like lobes are formed Obertello *et al.*, 2003; Schwencke and Caru, 2001).

The intercellular infection, however, occurs when *Frankia* hyphae penetrate the middle lamella between adjacent root epidermal cells (Obertello *et al.*, 2003; Schwencke and Caru, 2001). Within the root it branches and invades the intercellular spaces toward the nodule lobe primordium and here the intercellular colonisation occurs (Schwencke and Caru, 2001). The nodule lobe primordium is also started in the pericycle by the induction of the mitotic activity similarly to the intracellular infection but without the prenodule formation (Schwencke and Caru, 2001). The continuous growth of new lobes in both of the pathways lead to formation of typical coralloid structures of actinorhizal nodules, which are finally composed of several root-like structures, or lobes. In each lobe, the vascular bundle is central and the *Frankia* is located.



# Figure 2.3 Infection and early organogenesis of a nodule lobe in actinorhizal plants (source Obertello *et al.*, 2003).

#### v) Environmental factors affecting nitrogen fixation in root nodules

Nitrogen fixation, like plants, is affected by edaphic and environmental factors in the field (Giller, 2003; Giller, 2001; Teklehaimanot and Anim-Kwapong, 1996). These include soil acidity, soil compaction, oxygen and carbon dioxide balances in the soil and climatic conditions such as temperature, moisture levels and availability affecting nitrogen fixation (Dommergues, 1997; Griffths and McCormick, 1984; Martin *et al.*, 2003; Schwintzer and Tjepkema 2001, Winship and Tjepkema, 1982).

Acidic soil is known to affect nodulation and nitrogen fixation (Dommergues, 1997; Griffths and McCormick, 1984; Martin *et al.*, 2003). Griffths and McCormick, (1984), reported the highest nodulation between pH of 5.5 and 7.2 in *Alnus glutinosa*. However, below pH of 5.5 and above 7.2, nodulation was decreased (Griffths and McCormick, 1984).

An atmospheric level of oxygen is required for nitrogenase activity to take place in the root nodules (Schwintzer and Tjepkema, 2001). Reduced oxygen level and elevated carbon-dioxide may occur as a result of compacted soils and such conditions are reported to reduce nitrogenase activity (Schwintzer and Tjepkema, 2001; Tjepkema, 1978). Schwintzer and Tjepkema (2001) reported that some plants do adapt to conditions of elevated carbon-dioxide and performed the same as those that were in normal atmospheric oxygen. Winship and Tjepkema, (1982), reported that oxygen level of 15 kPa did not affect nitrogen fixation whereas nitrogenase activity was reduced at levels below 5 kPa and above 80 kPa.

Moisture conditions affect symbiotic nitrogen fixation (Dommergues, 1997). Water deficits for example will affect nitrogen fixation through influences on the host plant metabolism, nodulation and nodule formation (Dommergues, 1997). Excess moisture, however, tends to retard the activity of the *Frankia*. Floods are reported to have a negative effect on nitrogen fixation in *Alnus incana* (Kaelke and Dawson 2003). Red alder is reported to have shown a decline in nitrogenase activity after 24 hours of flooding but to have returned to normal after prolonged flooding (Batzli and Dawson, 1997). Sayed *et al.*, (1997) reported that dry soils reduced the effectiveness of the different *Frankia* strains tested and different strains behaved differently in soils maintain at field capacity.

The effect of temperature on nitrogen fixation is best explained by the patterns of acetylene reduction activity through the seasons (Binkley, *et al.*, 1994). The rates tend to rise in spring and summer when it is warm and decline into the autumn (Binkley *et al.*, 1994; Teklehaimanot and Martin, 1999). Hawkins and McDonald, (1994), reported high rates of nitrogen fixation at 20 to  $25^{\circ}$  C. Winship and Tjepkema (1982) reported that nitrogenase activity was reduced below  $16^{\circ}$  C and ceased below  $8^{\circ}$  C in root nodules of red alder. The authors reported that above  $30^{\circ}$  C nitrogenase activity ceased whereas between 16 and  $26^{\circ}$  C, nitrogenase activity was high (Winship and Tjepkema, 1982). Temperature is also known to affect infectivity of *Frankia* (Sayed *et al.*, 1997).

Nitrogen and phosphorus are known to affect n-fixation (Binkley *et al.*, 2003). Added phosphorus stimulated remarkable increases in nitrogen fixation through n-dilution and acetylene reduction assays (Binkley *et al.*, 2003). Nitrogen accumulated in the soil tends to have a negative effect on nitrogen fixation in symbiotic relationships (Binkley

*et al.*, 1994). The N status of the plant is thought to influence root growth, transport activity and nodule growth and activity (Parsons and Sunley, 2001). The precise signals that carry plant N status are unknown but are likely to be N-rich amino acids that are translocated from the shoot. In legume systems,  $O_2$  gas diffusion is closely regulated and can restrict nodule activity when N is available. In actinorhizal plants, control may be through carbohydrate availability or oxygen diffusion depending on availability (Parsons and Sunley, 2001).

#### vi) Transfer of fixed nitrogen in the plant

Once fixed, nitrogen is transferred from the microbe to the host by translocation in legumes and other  $N_2$  fixing plants. The nitrogen is bound to the organic carbon and is translocated in the form of amids. Non-nitrogen fixing plants can only access this nitrogen when the nitrogen fixing organism dies and is broken down by microorganisms. This transfer, via mineralization is less efficient than in the symbiotic relationship. The microorganisms that break the organic matter also bind part of the nitrogen that is mineralized thus making very little available to the plant (Killham, 1994).

#### vii) Transfer of fixed nitrogen to the soil

It has been reported that the nitrogen fixed is released into the soil from roots of the nitrogen fixing plant (Paynel *et al.*, 2001). The primary pathway for nitrogen transfer from the plant to the soil is, however, through decomposition of dead leaves, roots and nodules of the nitrogen fixing plant. Tree roots, therefore, play essential functions in the

supply of resources for growth and there is relatively poor understanding of how they function in their environment (Smit, *et al.*, 2000). The most important information lacking in terms of soil fertility is the role of fine root and nodule turnover in nitrogen transfer to the soil. The below ground biomass of roots is highly dynamic with new roots, death of old roots and decomposition of old roots. The death of old roots and nodules and their subsequent decomposition is the mechanism by which fixed nitrogen is made available in the soil. There is however, very little information on red alder regarding this aspect. Good knowledge of the dynamics and root and nodule turnover of red alder in soil is essential to understand the role red alder plays in soil fertility maintenance and improvement.

#### viii) Nodule distribution, dynamics and turnover

Very few studies have looked at the dynamics of in nitrogen-fixing trees and shrubs (Chesney and Nygren, 2002; Rytter, 1989; Valverde and Wall, 2002). Chesney and Nygren, (2002), reported nodule dynamics in *Erythrina poeppigiana*. Their studies were used to test the effect of aboveground pruning on the root nodule. The study was also carried out in the tropics, where conditions differ with those of the temperate regions. The density of nodules was reported to be decreasing with increasing depth (Rytter, 1989). Dulormne *et al.*, (2003), reported that *Gliricia sepium* nodules contributed more than 10 kg ha<sup>-1</sup> of N through nodule turnover.

#### 2.4.3 Measurement of nitrogen fixation

There are several methods that are available for measuring nitrogen fixation. Bergesen (1980) classified these methods as direct and indirect. The methods include total

nitrogen, acetylene reduction assay, nitrogen-15 dilution methods and the nitrogen-15 natural abundance method (Bergesen, 1980; Danso *et al.*, 1992). Ureides have also been used to evaluate nitrogen fixed in legume plants (Danso *et al.*, 1992). These methods are outlined below.

#### i) Total nitrogen method

This method involves the measurement of nitrogen accumulation over time to find out if there is an increase. The method assumes that the nitrogen fixed remains within the ecosystem. The method may result in underestimation of nitrogen fixed in areas where there is leaching of nitrogen in the soil. A modification of this method uses a time sequence plots at different locations to measure accretion that would be observed at a site over time. This method assumes that total nitrogen of each site will be similar for tree stands of the same age (Binkley *et al.*, 1994). Accretion of nitrogen in forest floors has been reported using this method (Newton *et al.*, 1968; Cole and Newton, 1986; Binkley *et al.*, 1994). Heilman and Ekuan (1982) reported accretion rates of 150 kg<sup>-1</sup>ha<sup>-1</sup> yr<sup>-1</sup> in 5 year-old red alder stands and rates as low as 60 kg ha<sup>-1</sup>yr<sup>-1</sup> in 2-4 year old red alder stands in Western Washington have been reported by Bormann and Gordon (1984).

#### ii) Total nitrogen difference (TND) method

Total nitrogen difference method is simple and is based on estimating how much of the total nitrogen (NT) in the plant was accumulated from the soil (NS) and the remainder is then attributed to fixed nitrogen (Danso *et al.*, 1992). The total nitrogen in a control

non-fixing plant is assumed to reflect the nitrogen acquired from the soil (Danso *et al.*, 1992). Use of different non-fixing tree species as controls may increase the accuracy of estimating nitrogen acquired from the soil and ultimately the biological nitrogen fixation (Danso, 1995). This method is recommended in low nitrogen soils because of small nitrogen contribution from the soil nitrogen relative to the fixed nitrogen (Danso *et al.*, 1992). In soils with high concentrations of soil nitrogen, soil nitrogen may contribute more to plant growth and therefore leading to significant reduction in biological nitrogen fixation. The method is, therefore, recommended only for sandy soils or low nitrogen soils (Danso *et al.*, 1992). Mariotti *et al.*, 1992; Parrotta *et al.*, (1994) used this method to estimate nitrogen fixed in *Casuarina equisetifolia*.

#### iii) Ureides as a measure of biological nitrogen fixation

Ureides are used in some grain legumes as a measure of biologically fixed nitrogen. The method is based on the composition of nitrogen compounds in the xylem sap (Danso *et al.*, 1992; Peoples *et al.*, 1996). The relative amounts of ureides in biologically fixed nitrogen in the xylem stream serves as an indicator of nitrogen fixed (Danso *et al.*, 1992). The method, therefore, serves to overcome problems of quantifying nitrogen from the soil separately (Danso *et al.*, 1992). Gathumbi *et al.* (2002) and Peoples *et al.* (1996) reported high ureide content in legume plants. Ureide content was found to be linearly positively correlated with natural abundance in tree legumes (Gathumbi *et al.*, 2002). It has also been reported, however, that the method is not always applicable (Danso *et al.*, 1992).

#### iv) Acetylene reduction assay

The acetylene reduction assay is a cheap, simple, rapid and sensitive method for measuring biological nitrogen fixation in nitrogen fixing plants by measuring nitrogenase activity (Hardy *et al.*, 1973; Singh, 1998; Tjepkema and Schwintzer, 1992). This method for assessing fixation uses incubation of detached nodules in enclosed vessel with acetylene to estimate the current rate of nitrogen fixation (Singh, 1998; Singh and Wright, 2003; Turner and Gibson, 1980). It is also possible to measure nitrogenase activity using intact plants with a flow through system (Singh and Wright, 2003).

The nitrogenase enzyme has high preference for acetylene over atmospheric nitrogen and the product of acetylene reduction, ethylene, is easy to measure. It is based on the ability of nitrogenase to reduce acetylene to ethylene, which is then measured by gas chromatography (Shear and Kohl, 1986). The rate of acetylene reduction is converted into an estimate of nitrogen fixation by the division that accounts for the greater electron requirement for nitrogen fixation per mol of atmospheric nitrogen. The nitrogen fixed can then be extrapolated to an annual and hectare basis by multiplying the total nodule biomass per hectare by the length of time that nodules are active (Teklehaimanot and Martin, 1999).

Acetylene reduction assay has become a common method of measuring nitrogen fixation (Turner and Gibson, 1980). Nitrogen fixation measured by this method has been reported in red alder (Binkley *et al.*, 1994; Teklehaimanot and Martin, 1999; Winship and Tjepkema, 1982). However this method is said to be imprecise because of

variations in nodule activity within and between plants, nodule activity through the season, nodule biomass per hectare, proper conversion from acetylene to nitrogen and errors that may arise from the effects of acetylene and ethylene on the physiology of symbiosis (Binkley *et al.*, 1994).

Another draw back with this method is the use of detached roots and nodules (Singh and Wright, 2003). The shoots are removed, roots are shaken and then they are soaked in water and such a process reduces nitrogenase activity (Singh, 1998; Singh and Wright, 2003). The effect of plant disturbance on nitrogenase activity is, however, expected to vary depending on the length of time that passes between the sampling and the measurement of nitrogenase activity (Singh and Wright, 2003). The use of intact plants with this method is limited to the flow-through system which is applicable only to pot grown plants (Singh and Wright, 2003). However, this method is still very popular with researchers and has been used to measure nitrogenase activity with *Alnus* plants (Binkley *et al.*, 1994; Teklehaimanot and Martin, 1999; Tripp *et al.*, 1979; Wheeler, *et al.*, 1981; Zitzer and Dawson, 1989).

#### v) The nitrogen-15 incorporation method

The third group of methods for estimating fixation rate uses the heavy, stable isotope  $^{15}$ N to directly trace nitrogen in plants (Bergersen, 1980). The first of these methods involves the exposure of roots to  $^{15}$ N<sub>2</sub> and the nodules then examined for the incorporation of  $^{15}$ N. In long-term experiments, nitrogen in the soil can be enriched with  $^{15}$ N. After some years lower ratios of the of  $^{15}$ N:  $^{14}$ N in the nitrogen fixing plant

relative to the non-nitrogen fixers can be used to calculate the proportion of nitrogen derived from the atmosphere (Bergersen, 1980).

This method also has problems that limit its use in the field. Exposure of roots and nodules to  ${}^{15}N_2$  is usually for a short period, making seasonal estimates difficult. The  ${}^{15}N_2$  gas must reach the nodule in a defined small volume. Thirdly, the  ${}^{15}N_2$  is costly and analysis in the mass spectrometer is time consuming (Shear and Kohl, 1986).

### vi) The <sup>15</sup>N-labelled fertilizer method

This method also known as the isotope dilution method involves labelling of the soil nitrogen pool using <sup>15</sup>N-enriched fertilizer to the soil on which nitrogen-fixing plants are to be grown (Danso *et al.*, 1992; Shear and Kohl, 1986). The <sup>15</sup>N abundance of the nitrogen fixing plant, the 15N abundance of the labelled pool of the available soil N and the total nitrogen in the fixing plant has to be known in order to calculate the amount of nitrogen fixed. The <sup>15</sup>N abundance of the non-fixing reference plant is also used to calculate the <sup>15</sup>N abundance of the available soil nitrogen pool (Shear and Kohl, 1986). This method assumes that the <sup>15</sup>N abundance of nitrogen taken from the soil by reference plants and nitrogen-fixing plants is the same (Shear and Kohl, 1986).

This method has its own limitations. The soil nitrogen dilutes the <sup>15</sup>N label of the added nitrogen and this dilution increases over time (Shear and Kohl, 1986). There is also a difficulty in the uniform application of the <sup>15</sup>N-labelled fertilizer to the volume of soil and as such it will decrease with soil depth (Shear and Kohl, 1986). Enriched tracers have been reported to be very expensive (Robinson, 2001). Several authors reported

quantities of biologically fixed nitrogen using this method (Cadisch, et al., 1989; Hansen and Vinter, 2001; Kakei and Clifford, 2000; Kurdali, 2000; Peoples et al., 1996, Rowe et al., 2001; Vinther and Jensen, 2000).

#### vii) Nitrogen-15 natural abundance method

In some areas, the natural ratios of <sup>15</sup>N: <sup>14</sup>N in the soil may differ enough from that of the atmosphere to allow for a calculation of nitrogen fixation by comparing the ratio in nitrogen fixing trees to adjacent non-nitrogen fixing trees (Binkley *et al*, 1994; Handley and Raven, 1992). Natural abundance method is based on the N-tracer method. N from the atmospheric N<sub>2</sub>, in addition to the combined soil nitrogen sources, is compared to the N of the non-nitrogen fixing plant that relies on soil-derived nitrogen (Hogberg, 1997; Vitousek *et al.*, 1989). The natural abundance of <sup>15</sup>N ranges from 0.01 to 4 atom percent. The <sup>15</sup>N abundance values are reported as  $\delta^{15}$ N (delta nirogen-15) in units of per mil <sup>15</sup>N (°/<sub>oo</sub><sup>15</sup>N) (Hogberg, 1997; Shear and Kohl, 1993).

This method, called nitrogen-15 abundance, was used in red alder but the authors concluded that natural variations in the ratios of the isotope were too inconsistent to allow for determination of nitrogen fixation (Binkley *et al.*, 1985). Domenach *et al.* (1989), working on *A. glutinosa* and *A. incana* reported that the trees fixed 110% and 75% nitrogen from the atmosphere, respectively.

The natural abundance method is valid when: (a) the soil delta-15N of the soil derived nitrogen is significantly different from the delta-15N of the atmospheric N, (b) control non-nitrogen fixing plants used must be growing in the same place and exploring

similar soil volumes, and (c) the isotopic values of the nitrogen originating from the  $N_2$ fixation process has to be determined using nodulated plants growing on an N-free media (Domenach *et al.*, 1989; Shear and Kohl, 1986). Leaves are the most practicable parts of the plant to collect and to use to estimate nitrogen fixation (Domenach *et al.*, 1989).

<sup>15</sup>N natural abundance is relatively precisely measured using the isotope ration mass spectrometry. Measuring nitrogen natural abundance is a challenge since the variation of the isotope may be very small (Domenach *et al.*, 1989; Shear and Kohl, 1993). According to Mulvaney (1993) the mass spectrometer is the most accurate instrument to measure ratio of isotopes.

#### 2.4.4 Methods of assessing root and nodule dynamics and turnover

Information on root and nodule dynamics and turnover is required in order to estimate the amount  $N_2$  fixation and its deposition in the soil. Böhm (1979) mentioned that root research under natural field conditions is still a step-child. He further argued that the reasons were primarily methodological (Böhm, 1979). Methods used then were tedious, time consuming and accuracy of data obtained was questionable Böhm (1979). More than twenty years later, working on roots is still considered to be tedious and time consuming; however, methods have been greatly improved (Livesley *et al.*, 2000; Scroth 2003; Smith, 2001; Smit *et al.*, 2000).

Several methods are available to study roots. Böhm (1979) elaborated some of the available methods. These are auger sampling, ingrowth cores and modified ingrowth

cores (Lukac and Godbold, 2001), pinboards (Oliveira *et al.*, 2000), trench profile techniques (Tomlinson *et al.* 1998) and core break methods (van Noordwijk *et al.*, 2000). Besides the methods above, Jose *et al.*, (2001) compared the minirhizotron and soil core methods in quantifying roots biomass. The minirhizotron method has been evaluated by Tierney *et al.*, (2001), who concluded that it is a suitable method for estimating fine root longevity. Smith (2001), Spek *et al.*, (1994) and van Noordwijk *et al.*, (1994) reported use of proximal root diameter with fractal rules to estimate root lengths. Destructive sampling such as soil coring and trenching can suitably be used to study root nodules. Chesney and Nygren (2002) have used root coring to study changes in root nodules due to branch pruning.

#### 2.4.5 The use of red alder as a source of timber

Red alder has a diffuse-porous, moderately dense and uniform textured wood (Harrington, 1990). It is used for making solid wood products such as furniture, cabinets, pallets and novelties. Composite products, which include plywood and flake board fibre-based products such as tissues and writing papers have been made from wood of red alder (Harrington, 1990; Resch, 1980).

Timber quality is assessed on wood with the end-use of wood in mind (Jozsa and Middleton, 1994). Moisture content, nominal specific gravity and temperature influence the strength of wood. High moisture content tends to reduce the strength of wood and the reverse is true. Tree species with high specific gravity have high strength values. A rise in temperature results in reduction in strength (Lavers, 1983). Specific gravity, modulus of rupture and modulus of elasticity are some of the measures undertaken in

wood to determine mechanical properties of wood (Desch and Dinwoodie, 1996; Lavers, 1983).

#### 2.4.6 Mechanical properties of wood

Bending properties of timber are important where it is used as beam, for example, in roof trusses, chair bottoms and table-tops (Desch and Dinwoodie, 1996). Static bending is the measure of strength of a material as a beam. In resting position, the upper part of a beam is in compression while the lower part is in tension and the mid-way part is in neutral. A load applied to the middle of the beam will deflect it out of the horizontal position. This causes shortening of fibres in the upper part and elongation of those on the lower side. As the load increases, failures develop on the upper side followed by tensile failure or horizontal shear in any order. The load that can be sustained by a beam depends on the distance between the points of support and the section area of the beam (Desch and Dinwoodie, 1996).

Timber used to make columns, props and chair legs need to have high strength in longitudinal compression (Desch and Dinwoodie, 1996). The authors, however, mention that timber often fails in bending rather than in compression. The resistance to crushing is important in uses such as wedges, bearing blocks, rollers and railway sleepers (Desch and Dinwoodie, 1996). Timbers with high density have high compression strength across the grain (Desch and Dinwoodie, 1996).

Few reports are available on the study of timber quality of red alder (Evans *et al.*, 2000; Gartner *et al.*, 1996; Harrington and DeBell, 1980, Lei *et al.*, 1997). Studies carried out on samples measuring 0.635 in width, 0.635 in depth and 8.89 cm length, using modified American standard for testing materials (ASTM) by Evans *et al.*, (2000) on red alder, showed variations in strength of modulus of elasticity (MOE) and modulus of rupture (MOR). The MOR was measured at about 55 MPa on the opposite and the lean side of the stem on wood at ten years old. MOE was measured at 7586 MPa and 8965 MPa on the opposite side and the lean side of red alder on wood ten years old. Both properties showed significantly higher values after the age of ten years. Studies carried out by Lei *et al.* (1997), showed MOE varying between 5000 and 8000 MPa where the MOR varied between 50 and 70 MPa (Lei *et al.*, 1997). The authors however, reported that rate of growth did not significantly alter the compression and bending properties of red alder wood.

#### 2.4.6.1 Specific gravity

Specific gravity of wood is the ratio of weight of wood to that of equal volume of water. In wood, this changes with shrinkage and swelling due to changes in moisture content (Desch and Dinwoodie, 1996; Lavers, 1983). Specific gravity is known to be a major factor that affects strength properties of wood (Zobel and Buijtenen, 1989). Species with wood of high specific gravity have high strength properties. Specific gravity or density can be determined using dry measure of volume and volume displacement methods. The samples to be used should be oven-dried at 105°C until a constant mass is reached which is at moisture content of zero. Wood density may also be measured at 12% since this is the moisture content where timber is at equilibrium with relative humidity of 65% in the atmosphere (Desch and Dinwoodie, 1996).

Evans *et al.* (2000), Gartner *et al.* (1996), Harrington and Debell (1980) and Lei *et al.* (1997), compared the specific gravity of wood from different alder provenances. The results showed a variation in specific gravity between 0.33 and 0.50, with an average of 0.40 (Harrington and DeBell, 1980; Lei *et al.*, 1997). The authors, however, concluded that there were no significant differences in specific gravity between tree growing at different growth rates and the different provenances of red alder.

#### 2.4.6.2 Temperature and moisture content

Temperature influences the strength properties of wood (Desch and Dinwoodie, 1996). An increase in temperature results in a decrease in strength of wood. This effect tends to increase with increase in moisture content. Moisture content has a marked effect on the strength properties of wood. At high moisture contents, strength properties are considerably reduced. At moisture contents below 24%, strength values increase (Porter, 1981). Moisture tends to induce movements and bending properties in wood (Bengtsson, 2000; Bengtsson, 2001). High moisture content also tends to favour attack by pathogens (Desch and Dinwoodie, 1996). The exposure of wood during processing and use may have an effect on moisture content, temperature and finally an effect on specific gravity of wood (de Moraes *et al.*, 2004)

#### 2.4.6.3 Knots

Knots affect strength properties in wood (Bengtsson, 2000; Itagaki *et al.*, 1999). The varying type, size, positions and number of the knots affect strength at varying levels (Desch and Dinwoodie, 1996; Itagaki *et al.*, 1999; Porter, 1981). Such knots are a result

of silvicultural treatment such as spacing and pruning (Dunham, 1996; Forsberg and Warenso, 2001, Persson *et al.*, 1995). Knots affects strength properties since cells in the knots are not in line with cells in the rest of the wood (Desch and Dinwoodie, 1996). Knots in the edge of wood affect strength more than those found in the centre of wood (Bengtsson, 2000; Itagaki *et al.*, 1999; Porter, 1981).

#### 2.4.6.4 Slope of the grain

This refers to the deviation of line of fibres from a line parallel to the sides of piece of wood (Porter, 1981). This can result from the pattern of tree growth, such as taper, spiral grain or from the manner in which the timber is sawn (Forsberg and Warensjo, 2001; Kliger, 2001). Variation in grain angle between tree species is large (Forsberg and Warensjo, 2001). In some trees the angle increases towards the bark, while in others it is constant (Forsberg and Warensjo, 2001). Slope in grain is determined by genetic factors and factors affecting growth of the tree (Forsberg and Warensjo, 2001). Slope of grains in wood can result in marked reduction in strength (Macdonald and Hubert, 2002). However this varies depending on strength properties in question and the degree of the slope of the grains and does not affect the suitability of wood for panel and pulp production (Macdonald and Hubert, 2002).

#### 2.4.7 The use of red alder for firewood and biomass energy

Red alder has been used as fuelwood in home fireplaces and stoves. The wood residue has been used in mills to produce heat for drying and other processes (Harrington, 1990). The tree has been evaluated for use in biomass farms for energy conversion. Energy values close to the average 18605 kJ kg<sup>-1</sup> have been reported (Harker *et al.*, 1982; Tillman, 1987).

#### 2.4.7.1 Biomass energy and wood fuel

Firewood is a renewable energy resource. It is used worldwide to provide energy for domestic use. Firewood is a largely used fuel in the developing world since it is a cheap alternative to fossil fuels. Wood being a renewable resource is considered for power generation as a substitute for fossil fuels in both developing and developed world (Arbon, 2002).

#### 2.4.7.2. Advantages associated with biomass energy

Biomass includes a number of products such as straw, grasses, residues from agricultural operations and wood (Pitcher *et al.*, 1998). Pitcher *et al.*, (1998), highlighted several advantages that are associated with biomass as renewable energy resource as shown below:

- The resource is inexhaustible on a human time scale.
- Have very little emissions to the atmosphere.
- It is carried out locally and this allows for good capacity planning and future modifications as necessary.
- Reduces dependency on imports.
- Increases opportunities for biomass produced in agriculture.

#### 2.4.7.3. Fuel value of wood

Anatomical properties such as the structure of wood fibres, pathways for moisture content and physical characteristics such as moisture content, specific gravity, and void volume determine the fuel value of wood. Chemical properties of wood that determine the calorific value are hemicellulose and lignin content, proximate and ultimate contents (Murphey and Cutter, 1974; Tillman *et al.*, 1981). The average elemental composition of dry wood is known to depart little from 49.5% carbon, 6% hydrogen and oxygen 43.5% (Harker *et al.*, 1982).

Energy value of a material is the heat released on combustion of that substance in oxygen filled bomb at constant volume. The proximate analysis of the sample includes measurements of moisture content, volatile contents and ash contents of the charcoal. Moisture content in wood reduces the amount of heat liberated (Harker *et al.*, 1982; Murphey and Cutter, 1974). Other characteristics that may be used include drying rate, production of ambers, rate of burning, light source, ease of splitting and production of smoke (Abbot *et al.*, 1997; Groves and Chivuya, 1989).

Harker, *et al.*, (1982) reported gross calorific value of 18 608 kJ kg<sup>-1</sup> in red alder wood and an ash content of 0.4 % (dry weight basis) has been reported by Graboski *et al.*, (1979). Proe *et al.*, (1999), researched and reported on coppice and stem biomass production for the purposes of producing wood biomass for fuel. Coppiced alder trees showed very high mortality *circa* 50%. This, however, may not give a true performance of coppiced alder since coppicing was carried out within one year after planting.

#### **CHAPTER 3.0**

## CONTRIBUTION OF RED ALDER TO SOIL FERTILITY IMPROVEMENT AND MAINTENANCE IN SILVOPASTORAL SYSTEMS

#### 3.1 Introduction

Several reports have demonstrated the attractive role of nitrogen-fixing trees as sources of nitrogen and organic matter and their potential in soil fertility improvement and maintenance (Giller, 2003; Scroth and Sinclair, 2003; Teklehaimanot and Martin, 1999; Young, 1997). Red alder is one of the important nitrogen fixing tree species in temperate and boreal ecosystems (Binkley *et al.*, 1994; Rojas *et al.*, 2002). Red alder has been planted in mixture with non-nitrogen fixing trees to provide the much needed nitrogen in forest plantation in the USA (Binkley *et al.*, 1994). The tree has been experimentally planted with a maize crop in the Pacific Northwest for the same purpose (Seiter *et al.*, 1995). Experiments have been conducted with the tree on pasture in North Wales, UK for the same reason (Teklehaimanot and Martin, 1999). The incorporation of this tree species in agroforestry system is considered cheaper and environmentally sensible than the use of commercial fertilizers (Teklehaimanot and Martin, 1999).

Rates of nitrogen fixation in red alder have been previously assessed using the acetylene reduction assay method (Teklehaimanot and Martin 1999; Tripp *et al.*, 1979). Teklehaimanot and Martin (1999) and Tripp *et al.* (1979) reported significant variations in the diurnal and seasonal patterns of nitrogenase activity in red alder. Tripp *et al.*, (1979) reported mean  $C_2H_2$  reduction of 27.5 µmoles g<sup>-1</sup> dry nodule h<sup>-1</sup> with maximum

rates of activity in May and June. The authors estimated 62 kg ha<sup>-1</sup> of nitrogen was fixed annually (Tripp *et al.*, 1979). Teklehaimanot *et al.*, (1999) reported a lower average nitrogenase activity of 18  $\mu$ mol C<sub>2</sub>H<sub>2</sub> g dwt<sup>-1</sup> h<sup>-1</sup> in red alder in North Wales. However, the acetylene reduction assay method has been reported to have limitations and therefore may not give reliable results of nitrogen fixation (Minchin *et al.*, 1986).

Binkley *et al.* (1985) measured nitrogen fixation in red alder planted in forestry system using nitrogen-15 natural abundance method. Red alder grown in N-free media showed a natural abundance of  $-0.3^{\circ}/_{\circ\circ}$ . The authors, however, concluded that the method was not useful to quantify nitrogen fixed in N-fixing trees because of the absence of consistent patterns across locations. The natural abundance method has, however, been used successfully to measure nitrogen fixation in other alders and actinorhizal tree species (Tjepkema *et al.*, 2000; Domenach *et al.*, 1989; Hurd *et al.*, 2001; Sanborn *et al.*, 2002).

In addition to nitrogen fixation, nitrogen fixing trees provide organic matter to soil through decay of fine roots, nodules and leaf litter. So far no study has been carried out to quantify fine root turn-over in red alder and its contribution to soil organic matter and nitrogen balance. The contribution of fine root turn-over of other tree species in the recycling of nutrients and carbon in the rhizosphere and the consequences of these on soil nutrient balance has been reported by several authors (Chesney and Nygren, 2002; Curt *et al.*, 2001; Fownes *et al.*, 1991; Lehmann and Zech, 1998; Makkonen and Hilmisaari, 1999; Nygren and Ramirez, 1995; Ruess *et al.*, 1996; Tufekcioglu *et al.*, 1999). In addition to fine roots, root nodules of nitrogen-fixing trees die, decompose and as such contribute to the net balance of carbon and nitrogen in the soil. However,

studies of nodule weight per unit volume of soil under red alder and indeed in other nitrogen fixing plants has not been given the deserved attention. There are only a few articles existing in literature where authors have studied and reported the effects of different pruning regimes of other trees species on nodule distribution and dynamics in agroforestry systems (Chesney and Nygren, 2002; Dulormne, *et al.*, 2003; Fownes *et al.*, 1991; Nygren and Ramirez, 1995).

#### **Objectives**

- To assess the rate of nitrogen fixation in red alder using nitrogen-15 natural abundance method.
- 2. To estimate the contribution of red alder to soil nitrogen and organic matter content.

#### 3.2 Methodology

#### 3.2.1 Nitrogen-15 natural abundance in samples

Nitrogen-15 natural abundance measurements were made to determine the rate of nitrogen fixation in red alder. Three samples each of fine roots, nodules, leaves and wood of red alder (*Alnus rubra*) were randomly collected from a 10 year old red alder stand at Henfaes research statation, North Wales, UK for nitrogen 15 natural abundance studies. Reference species were selected among non-nitrogen-fixing tree species occurring in the same area with red alder. They were sycamore (*Acer pseudoplatanus*),
blackthorn (*Prunus spinosa*) and willow (*Salix sp*). Fine roots, leaves and wood samples, were collected from the reference plants. In addition 3 soil samples were taken from the site each season. Plant samples were collected in each season: autumn (September), winter (February), spring (April) and summer (July), starting in 2002. During the winter season no leaf samples were taken as all the species were deciduous. The plant samples were oven dried at 80°C for 2 days. A ball mortar mill was used to grind the samples to a fine powder. The <sup>15</sup>N/<sup>14</sup>N isotope ratio and total nitrogen content of the powdered samples were determined using an automated nitrogen analyser coupled to a mass spectrometer at the stable isotope facility in the Department of Agricultural Sciences of the Imperial College London.

Plant and soil material measured in minute quantities (milligrams), were weighed into tin capsules and dropped into a furnace at 1000°C while in an atmosphere of oxygen. The tin ignited and burned exothermically, and the temperature rose to about 1800°C, oxidising the nitrogen and the carbon in the sample. The combustion products passed through a bed of chromium trioxide at 1000°C and a helium carrier gas ensured complete oxidation. A layer of copper oxide followed by a layer of silver wool finished the oxidation removing any sulphur. The products (NO<sub>2</sub> and CO<sub>2</sub>) were then passed through a second furnace containing copper at 600°C where excess oxygen was absorbed and nitrogen oxides (NO<sub>2</sub>) were reduced to elemental nitrogen (N<sub>2</sub>). Water was then removed in a trap containing anhydrous magnesium perchlorate and carbon dioxide in a trap 'Carbosorb'. The gas stream was then passed into a gas chromatogram where components of interest were separated and then were bled into a Stable Isotope Mass Spectrometer (Europa Scientific 20-20) where nitrogen and carbon isotopes were ionised and then separated in a magnetic field. The isotopic levels of species were detected separately and from their ratios, the level of <sup>15</sup>N calculated. Calibration of the system was made using known standards allowing both total element and isotope content to be obtained from each sample. Results were reported in  $\delta^{15}N$  (‰) in each sample.

The standard procedure described by Shear and Kohl (1993) was used to calculate the fraction of nitrogen that is derived from the atmosphere (FNdfa) in each plant part of red alder (Formula 3.1).

FNdfa = 
$$\frac{\delta^{15}N_o - \delta^{15}N_i}{\delta^{15}N_o - \delta^{15}N_a}$$
(3.1)

Where FNdfa – fraction of nitrogen derived from the atmosphere  $\delta^{15}N_a$  - delta <sup>15</sup>N in red alder grown in nitrogen free media  $\delta^{15}N_o$  – delta <sup>15</sup>N in non-fixing reference plant sample  $\delta^{15}N_t$  – delta <sup>15</sup>N in red alder sample  $\delta^{15}N_a$  = -0.3 (from Binkley *et al.*, 1985)

## 3.2.2. Total biomass of red alder

Total biomass of red alder was measured in order to estimate the rate of nitrogen fixation and the amount of fixed nitrogen accumulated in red alder and to quantify the annual contribution of red alder to soil organic matter and nitrogen content as described below in sections 3.2.3 and 3.2.4, respectively.

Destructive sampling was used to measure the above ground biomass of red alder. Three representative trees of red alder were randomly chosen and felled. Samples were obtained from the stems, branches and leaves. Trees at the border of the stand were not selected to avoid edge effects. The sample trees were  $11.13\pm0.52$  m in height with a stem diameter at breast height of  $10.96\pm0.98$  cm. The stem, branch and leaf samples were separated and oven-dried at  $80^{\circ}$ C for 24 hours. The samples were then weighed and recorded.

Belowground biomass of fine roots and nodules was estimated using a modified quarter spiral trench technique (Figure 3.1), as described by Tomlinson *et al.* (1998). In this study, samples were taken instead of trench sampling, as the soil was shallow brown earth with gravel, which was difficult to dig. Spiral sampling enabled a large proportion of the soil cores to be excavated with only minimal damage to the tree. Soil cores of 30 by 20 cm and 15 cm depth were excavated at each sampling point from 9 trees from each of the agroforestry plots (400 stems per ha<sup>-1</sup>) and forestry plots (2500 stems per ha<sup>-1</sup>) treatment plots. Core samples were collected in each season (summer, autumn, winter and spring), starting in the summer of 2003. A total of 54 core samples were collected during each season. Again trees in the border were not selected to avoid edge effects. The core samples were labelled and stored in a cold store for approximately one week before being separated into roots, nodules and soil in the green house of the University of Wales Bangor, UK.

Wet sieving was used to separate roots and nodules from the soil. The fabric suspension bucket with hanging funnels with connected hush links beneath was used to separate the roots and nodules from the soil. A centrally powered valve water circulatory system allowed the roots and nodules to float from the bucket into the hanging funnels. All grass roots and large roots (tree roots more than 2 mm in diameter) were discarded. Red alder roots were distinguished from grass roots by their brown coloration (grass roots were white in colour). The fine roots and nodules were further separated into categories of live and dead roots and live and dead nodules. Dead roots were easily distinguishable from live roots by their black colour. Live nodules were yellowish orange and dead nodules were black in colour. All the samples were oven dried at 80°C for two days in a laboratory and then weighed and their weight density recorded. The weight density measurement was finally converted to weight per hectare (biomass).



Figure 3.1 Diagram showing sampling points (at 0.3, 0.75 and 1.0 m) from tree base using logarithmic spiral technique

## 3.2.3. Total fixed nitrogen accumulated and the rate of annual nitrogen fixation in red alder

The live fine root biomass data together with the data of above ground biomass (leaves and wood) as described above was used to estimate the total amount of nitrogen fixed per tree and finally the amount of fixed nitrogen accumulated per hectare. The following assumptions were made to calculate the annual fixation rate. The fine root biomass of both live and dead roots measured was assumed to be annual production. It was also assumed that annual wood increment of red alder was uniform over the 10 years. Thus, by taking the total wood biomass and dividing it by the number of years (10) the mean annual wood increment was calculated. Using mean annual wood increment and biomass of fine roots and leaves, the rate of nitrogen fixation per annum was calculated.

## 3.2.4. Contribution of leaves, fine roots and root nodules to soil organic matter and nitrogen content

Since only senescent leaves, fine roots and dead nodules of red alder pass to soil organic matter via decomposition, the biomass of leaves, fine roots (live and dead) and dead nodules and their nitrogen contents were used to estimate the potential contribution by red alder to the organic matter and nitrogen content of the soil. The wood biomass and live nodules were excluded form the calculation. Because red alder nodules are indeterminate, the turnover of live nodules may be longer than a year.

## 3.2.5 The C:N ratio

The C:N ratio in leaves of red alder was determined by diving the carbon content by the nitrogen content. Half of the leaf biomass in the leaf was assumed to contain 50% carbon. The C:N ratio was calculated for green leaves and senescent leaves. Senescent leaves were harvested during the autumn period before the leaves dropped to the ground.

## 3.3. Statistical analysis of results

Comparisons between plant parts in  $\delta^{15}$ N, FNdfa and nitrogen contents were made using analysis of variance (ANOVA) of Minitab 13 statistical software package.

## 3.4 Results

## 3.4.1 $\delta^{15}$ N values

Table 3.1 shows  $\delta^{15}$ N values in different plant parts during the four seasons. There was significant difference (P<0.001) in  $\delta^{15}$ N values between plant species. The  $\delta^{15}$ N values of red alder were consistently close to zero whereas the reference plants showed highly enriched values of  $\delta^{15}$ N. This indicates that red alder is efficiently fixing atmospheric nitrogen and the reference plants are largely dependent on soil nitrogen. There were no significant differences in  $\delta^{15}$ N of the different plant parts of red alder. Also between the different parts of each plant species ANOVA showed also no significant differences in  $\delta^{15}$ N.

As shown in Figure 3.2,  $\delta^{15}$ N values for the summer and autumn seasons were negative for red alder indicating active nitrogen fixation during these periods. The average values for winter and spring seasons were positive but close to zero in red alder, which may indicate reduced nitrogen fixation in red alder during these periods.

Table 3.1. Values of  $\delta^{15}N$  (‰±SE) in different plant parts (n=3) in the four seasons

an a		Summer	Autumn	Winter	Spring	Mean
Willow	Leaf	4.19±0.08	4.30±0.63		5.27±0.36	4.59±0.27
	Wood	$3.59{\pm}0.07$	$4.46 \pm 0.48$	$3.23 \pm 0.25$	4.44±1.16	3.93±0.31
	Root	$2.91 \pm 0.04$	3.79±0.57	3.27±0.12	$3.89 \pm 0.94$	3.46±0.26
	Mean	3.56±0.18	4.18±0.30	$3.25 \pm 0.12$	$4.54 \pm 0.48$	$3.94{\pm}0.18$
Blackthorn	Leaf	3.11±0.05	4.09±0.54		-	$3.59 \pm 0.32$
	Wood	3.55±0.10	5.77±0.12	5.63±0.16	$5.52 \pm 0.41$	5.12±0.29
	Root	4.53±0.02	4.69±0.23	3.36±0.18	$3.53 \pm 0.67$	4.02±0.23
	Mean	3.72±0.21	4.85±0.30	4.50±0.52	4.52±0.56	4.38±0.19
Sycamore	Leaf	3.22±0.04	3.85±0.24	-	3.81±0.51	3.63±0.19
	Wood	4.59±0.06	3.01±0.36	$1.99{\pm}0.60$	5.25±0.16	3.71±0.41
	Root	$0.52 \pm 0.05$	1.49±0.21	3.39±0.16	4.66±0.21	2.51±0.49
	Mean	2.78±0.59	2.78±0.37	2.69±0.42	4.58±0.26	3.25±0.25
Alder	Leaf	0.23±0.06	-0.73±0.23	× <b>-</b>	0.71±0.43	$0.07 \pm 0.25$
	Wood	-0.34±0.06	0.01±0.18	1.40±0.36	1.32±0.65	$0.60 \pm 0.28$
	Root	$-0.02 \pm 0.02$	-0.12±0.48	$-1.06\pm0.30$	$2.09 \pm 0.80$	$0.21 \pm 0.40$
	Mean	$-0.04 \pm 0.08$	-0.28±0.19	0.17±0.59	1.37±0.38	0.31±0.19



Figure 3.2. Mean values of  $\delta^{15}N$  (‰±SE) in different plant species during the four seasons.

## 3.4.2 Nitrogen content in plant parts

There was no significant difference between species in nitrogen content (Table 3.2). Season also did not have significant effect on the nitrogen content. Nitrogen content was significantly highest (P<0.001) in leaves in all species in comparison to other plant parts.

# Table 3.2:Mean nitrogen content (%±SE) of different parts of different plants<br/>(n=12).

		Summer	Autumn	Winter	Spring	Mean
Willow	Leaf	2.47±0.01	2.69±0.25	( <del>_</del>	4.91±0.05	3.36±0.40
	Wood	0.79±0.01	1.02±0.53	0.64±0.09	1.17±0.60	$0.91 \pm 0.18$
	Root	1.16±0.01	1.09±0.18	0.96±0.15	0.77±0.19	$1.00 \pm 0.08$
	Mean	1.47±0.26	$1.60 \pm 0.32$	$0.80 \pm 0.11$	2.28±0.68	$1.61 \pm 0.22$
Blackthorn	Leaf	3.37±0.01	2.68±0.08	-	-	3.02±0.16
	Wood	0.58±0.01	0.54±0.05	$0.54 \pm 0.02$	0.54±0.09	$0.55 \pm 0.02$
	Root	$1.17 \pm 0.01$	1.48±0.42	$1.52 \pm 0.01$	2.10±0.39	1.57±0.16
	Mean	$1.71 \pm 0.42$	1.57±0.33	1.03±0.22	1.32±0.39	$1.45 \pm 0.18$
Sycamore	Leaf	3.12±0.01	2.82±0.16	-	2.56±0.12	2.83±0.10
	Wood	$0.56 \pm 0.01$	0.35±0.07	$0.61 \pm 0.07$	$0.54 \pm 0.09$	0.51±0.04
	Root	$1.18 \pm 0.01$	$0.72 \pm 0.04$	0.53±0.09	0.67±0.06	0.77±0.08
	Mean	1.62±0.39	1.30±0.39	0.57±0.05	1.26±0.33	1.24±0.17
Alder	Leaf	2.88±0.01	2.16±0.05		2.12±0.20	2.39±0.14
	Wood	$0.70 \pm 0.01$	0.33±0.02	0.84±0.05	0.44±0.04	0.58±0.06
	Root	0.84±0.01	0.68±0.06	0.40±0.09	0.79±0.11	0.68±0.06
	Mean	1.48±0.35	1.05±0.28	0.62±0.11	1.12±0.26	1.11±0.14

# 3.4.3. $\delta^{15}N$ and nitrogen content in root nodules of red alder and the soil of the study site

Values of  $\delta^{15}$ N and nitrogen content were similar in both red alder nodules and the soils samples (Table 3.3). Both red alder nodules and soil from the site were highly enriched with <sup>15</sup>N, and these were similar to the  $\delta^{15}$ N values found in the reference plants (Table

3.1). There were no significant effects of season in the values of  $\delta^{15}N$  and nitrogen content in both root nodules and soils.

Table 3.3:	Mean values of $\delta^{15}N$ (‰±SE) and nitrogen content (%±SE) in root
	nodules of red alder and soils (n=3)

		AUTUMN	WINTER	SPRING	Mean
nodule	$\delta^{15}N$	5.58±0.54	6.29±0.28	6.18±0.97	6.01
soil	$\delta^{15}N$	5.44±0.25	5.49±0.21	6.93±0.14	5.95
nodule	N%	2.05±0.03	$1.81 \pm 0.04$	$1.70 \pm 0.05$	1.85
soil	N%	0.25±0.001	0.33±0.01	0.30±0.01	0.29

## 3.4.4. Fraction of nitrogen derived from the atmosphere in red alder

Fraction of nitrogen derived from the atmosphere (FNdfa) in red alder was calculated in percentage by using the mean values of  $\delta^{15}$ N in the three non-nitrogen fixing reference plants (Table 3.4). The results indicate that nearly 85% of nitrogen in red alder was derived from the atmosphere. The highest proportion of nitrogen fixed (100%) was found in leaves and the least in wood (26%) and values of FNdfa were higher both in the summer and autumn than in winter and spring.

		Summer	Autumn	Winter	Spring	Mean
Willow	Leaf	88	100		82	93
	Wood	100	94	52	66	78
	Root	91	96	100	43	88
Blackthorn	Leaf	84	100	-	-	97
	Wood	100	95	71	72	85
	Root	94	97	100	38	87
Sycamore	Leaf	85	100	-	75	90
	Wood	100	91	26	71	72
	Root	67	90	100	52	82
Mean	Leaf	86	100	-	79	91
	Wood	100	93	49	70	78
	Root	84	94	100	44	86
Overall						
mean		90	99	85	64	85

 Table 3.4.
 FNdfa (%) in red alder tree in relation to reference plants

## 3.4.5 Total biomass of red alder

### 3.4.5.1 Above-ground biomass

Leaf and wood biomass per tree and per hectare derived by destructively sampling three representative red alder trees are given in Table 3.5.

## Table 3.5 Biomass of leaves and wood of red alder at different planting densities

Treatment	Density	Leaf biomass (kg)	Wood biomass (kg)
	One tree	1.06±0.20	21.25±5.31
Agroforestry	400 trees ha <sup>-1</sup>	424	8500
Forestry	2500 trees ha <sup>-1</sup>	2650	53125

#### 3.4.5.2. Below-ground biomass of red alder

## (a) Live and dead root weight density and biomass

Live root weight density showed a significant difference between treatments (P<0.001). Live root weight density in the forestry plots was twice that of the agroforestry plots. There was also significant seasonal difference in live root weight density (P<0.001). When data was analyzed separately for each treatment, however, seasonality did not have significant effect on the live root weight density in the forestry treatment, where it remained more or less constant throughout the four seasons (Figure 3.4). The significant difference was due to the sharp increase in live root weight density in the

agroforestry treatment between winter and autumn as shown in Figure 3.3. Mean live root weight density in agroforestry and forestry over the four seasons was  $0.27\pm0.01$  and  $0.54\pm0.03$  kg m<sup>-3</sup>, respectively (Table 3.6). These gave 2700 and 5400 kg ha<sup>-1</sup> of live root total biomass in agroforestry and forestry plots, respectively.

Planting density had also a significant effect (P<0.01) on dead root weight density. Mean dead root weight density in forestry ( $0.079\pm0.01$  kg m<sup>-3</sup>) was more than twice that of agroforestry ( $0.036\pm0.01$  kg m<sup>-3</sup>) (Table 3.7). These gave 790 and 360 kg ha<sup>-1</sup> of dead root total biomass in forestry and agroforestry, respectively. Dead root weight density remained more or less constant throughout the four seasons in both agroforestry and forestry (Figures 3.3 and 3.4, respectively). There was also no significant difference in either live or dead root weight densities between distances from the base of the tree (Tables 3.6 and 3.7).

Table 3.6. Live root weight density (kg	g m <sup>-3</sup>	'±SE)	of	red	al	de	r
---	-------------------	-------	----	-----	----	----	---

	Distance (m)	Winter	Spring	Summer	Autumn	Mean
Agroforestry	0.30	$0.11 \pm 0.01$	0.23±0.07	0.34±0.06	0.47±0.06	
	0.57	$0.20{\pm}0.04$	0.21±0.06	0.26±0.04	$0.41 \pm 0.04$	
	1.00	0.21±0.05	0.18±0.03	$0.30 \pm 0.04$	$0.40 \pm 0.10$	
	Mean	$0.17 \pm 0.02$	0.21±0.03	0.30±0.03	$0.43 \pm 0.04$	$0.27 \pm 0.01$
Forestry	0.30	$0.42{\pm}0.07$	0.52±0.13	$0.46 \pm 0.09$	$0.46 \pm 0.06$	
	0.57	$0.44 \pm 0.13$	$0.40 \pm 0.08$	$0.54 \pm 0.10$	0.65±0.09	
	1.00	0.69±0.18	$0.66 \pm 0.12$	$0.63 \pm 0.16$	$0.62 \pm 0.10$	
	Mean	$0.52{\pm}0.02$	$0.53 \pm 0.07$	$0.54{\pm}0.07$	$0.58 \pm 0.05$	$0.54{\pm}0.03$

Winter Spring Summer Autumn Mean Distance  $0.039 \pm 0.01$  $0.016 \pm 0.01$  $0.024 \pm 0.01$ Agroforestry 0.30  $0.049 \pm 0.02$  $0.039 \pm 0.01$  $0.029 \pm 0.01$  $0.040 \pm 0.01$ 0.57  $0.021 \pm 0.01$  $0.041 \pm 0.01$ 1.00  $0.045 \pm 0.01$  $0.047 \pm 0.03$  $0.045 \pm 0.01$  $0.036 \pm 0.01$  $0.041 \pm 0.01$  $0.035 \pm 0.01$  $0.038 \pm 0.01$  $0.030 \pm 0.01$ Mean  $0.064 \pm 0.01$  $0.117 \pm 0.02$ Forestry 0.30  $0.079 \pm 0.02$  $0.064 \pm 0.01$  $0.078 \pm 0.01$ 0.57 0.102±0.03 0.033±0.01 0.061±0.02  $0.076 \pm 0.01$ 1.00 0.074±0.02 0.092±0.03  $0.110 \pm 0.02$ Mean  $0.085 \pm 0.01$  $0.063 \pm 0.01$  $0.078 \pm 0.01$  $0.090 \pm 0.01$  $0.079 \pm 0.01$ 

Table 3.7. Dead root weight density (kg m<sup>-3</sup>  $\pm$ SE) of red alder



Figure 3.3: Live and dead root weight densities (kg m<sup>-3</sup>±SE) in the agroforestry plots in 2003.



Figure 3.4: Live and dead root weight density (kg m<sup>-3</sup>±SE) in the forestry plots in 2003.

## (b) Live and dead nodule weight density

Neither treatment nor season had significant effect on both live and dead nodule weight densities. Despite lack of significant differences, both live and dead nodule weight densities were higher in agroforestry than in forestry. There was also no significant difference in both dead and live nodule weight densities between distances from the tree (Figure 3.5 and 3.6). Both live and dead nodule weight densities showed different patterns according to variations in season in agroforestry and forestry systems as shown in Figures 3.7 and 3.8, respectively. Both live and dead nodule weight densities in forestry plots increased between winter and summer whereas in the agroforestry plots they decreased. Mean live and dead nodule weight densities over the four seasons in the

agroforestry plots were  $0.088\pm0.03$  and  $0.052\pm0.03$  kg m<sup>-3</sup> whereas they were  $0.080\pm0.02$  and  $0.031\pm0.01$  kg m<sup>-3</sup> in the forestry plots, respectively. These gave live and dead nodule total biomass of 880 and 520 kg ha<sup>-1</sup> in agroforestry and 800 and 310 kg ha<sup>-1</sup> in forestry, respectively.



Figure 3.5: Nodule weight density (kg m<sup>-3</sup> $\pm$  SE) according to distances from the tree base in alder agroforestry plots in 2003.



Figure 3.6: Nodule weight density (kg m<sup>-3</sup> $\pm$  SE) according to distances from the tree base in alder forestry plots in 2003.



Figure 3.7: Live and dead nodule weight density (kg m<sup>-3</sup>±SE) according to season in the agroforestry plots in 2003.



Figure 3.8: Live and dead nodule weight density (kg m<sup>-3</sup>±SE) according to season in the forestry plots.

## 3.5 Total fixed nitrogen accumulated and the rate of nitrogen fixation in red alder trees

Using total biomass of leaves, wood (Table 3.5) and fine roots (Tables 3.6 and 3.7) and their nitrogen contents (Table 3.2) and FNdfa (Table 3.4), the total amount of fixed nitrogen accumulated in red alder tree, excluding nodules, was calculated as shown in Table 3.8. The nodules were not included as there were no reference plants with nodules. Thus, FNdfa was not calculated for nodules. The results show that there was a total of 65.55 and 334.14 kg ha<sup>-1</sup> of fixed nitrogen accumulated in red alder trees and the rate of nitrogen fixation was 30.95 and 117.84 kg ha<sup>-1</sup> yr<sup>-1</sup> in the 400 trees ha<sup>-1</sup> (agroforestry plots) and 2500 tree ha<sup>-1</sup> (forestry plots) planting density, respectively (Table 3.8).

		N content (kg ha <sup>-1</sup> )	Total fixed N (kg ha <sup>-1</sup> )	N fixation rate (kg ha <sup>-1</sup> yr <sup>-1</sup> )
Agroforestry	Leaves	10.13	9.22	9.22
	Wood	49.30	38.45	3.85
	Live roots	18.36	15.78	15.78
	Dead roots	2.45	2.1	2.1
	Total		65.55	30.95
Forestry	Leaves	63.34	57.63	57.63
	Wood	308.12	240.33	24.03
	Live roots	36.72	31.57	31.57
	Dead roots	5.37	4.61	4.61
	Total		334.14	117.84

## Table 3.8. Total fixed nitrogen accumulated and rate of fixation in red alder

## 3.6 Contribution of leaves, roots and dead root nodules to soil organic matter and nitrogen content

Using the data of fine root (live and dead) and dead nodule biomass given in Section 3.4.5.2, leaf biomass given in Table 3.5 and their nitrogen contents given in Table 3.2, the contribution of red alder to soil organic matter and nitrogen in agroforestry and forestry systems was estimated as shown in Table 3.9. The author assumed that the amounts of fine roots and nodules of red alder recorded at the site were annual productions and they senescent and decompose within a year. The author also assumed that senescent leaves are deposited on the soil and decompose within a year. Thus, senescent nodules, fine roots and leaves were assumed to pass to soil organic matter via decomposition.

The total amount of organic matter added to the soil due to senescent leaves and fine roots and dead nodules was estimated at 4.0 t ha<sup>-1</sup> yr<sup>-1</sup> and 9.1 t ha<sup>-1</sup> yr<sup>-1</sup> in agroforestry and forestry, respectively (Table 3.9). The total amount of nitrogen that could potentially be added to the soil as a result of decomposition of senescent leaves, root and dead nodules was estimated at 40.56 kg ha<sup>-1</sup> yr<sup>-1</sup> and 111.17 kg ha<sup>-1</sup> yr<sup>-1</sup> in agroforestry and forestry, respectively (Table 3.9). Out of the total nitrogen added to the soil, 27.1 kg ha<sup>-1</sup> yr<sup>-1</sup> and 93.81 kg ha<sup>-1</sup> yr<sup>-1</sup> (in agroforestry and forestry systems, respectively) were due to nitrogen fixation by red alder from the atmosphere.

Systems	Addition to the	Leaves	Fine roots	Dead	root	Total
	soil			nodules		
Agroforestry	Organic matter	424	3060	520	Managaran	4004
	addition					
	Total Nitrogen	10.13	20.81	9.62		40.56
	addition					
	Fixed nitrogen	9.22	17.88	-		27.1
	addition					
Forestry	Organic matter	2650	6190	310		9150
	addition					
	Total Nitrogen	63.34	42.09	5.74		111.17
	addition					
	Fixed nitrogen	57.63	36.18	-		93.81
	addition					

Table 3.9.Organic matter (kg ha<sup>-1</sup> yr<sup>-1</sup>) and nitrogen (kg ha<sup>-1</sup> yr<sup>-1</sup>) addition to<br/>the soil.

## 3.7 The Leaf C:N ratio

The C:N ratio of alder green leaves was calculated with the mean of  $23.02\pm2.1$ . There were significant differences (P>0.05) in C:N ratio through the seasons with lowest at 17.54 calculated in leaves sampled in the summer where the highest was calculated at 27.78 in the winter period. The mean C:N ratio of senescent leaves was calculated at 21.91±2.03. There was no significant differences (P<0.05) in the leave C:N ratio between the senescent leaves and green leaves.

## 3.7. Discussion

## 3.7.1 Delta 15N and natural abundance in selected plants at Henfaes

The investigation carried out at Henfaes research station, North Wales, which was the subject of this report, showed that red alder <sup>15</sup>N signatures were close to the nitrogen in the atmosphere. This means red alder fixed most of the nitrogen found within its biomass from the atmosphere. The  $\delta^{15}$ N values measured in reference plants at the site, however, showed consistently enriched values of <sup>15</sup>N in the samples of leaves, wood and roots. This shows that the reference plants heavily depend on soil nitrogen. The values measured in red alder showed negative or values close to zero. The depleted values in red alder indicate that a signature of <sup>15</sup>N which is similar to the nitrogen in the atmosphere as such showing that the atmosphere is source of the nitrogen in red alder. Binkley *et al.* (1985) reported  $\delta^{15}$ N values of *A. rubra*, which were consistent with results obtained in the present study. Studies carried out on other actinorhizal plants yielded similar negative delta values for nitrogen-15 (Domenach *et al.*, 1988; Tjepkema *et al.*, 2000).

The calculations were taken a step further to find out the proportions of nitrogen that was fixed from the atmosphere (FNdfa) as described in the methodology section of this chapter. This was carried out using  $\delta^{15}N$  values obtained in the non-nitrogen fixing reference plants grown in the same area with red alder. Comparison using the  $\delta^{15}N$  values in root, wood and leave parts showed that a significant part of nitrogen within red alder was fixed from the atmosphere. The lowest amount of 26% was found when comparisons were made with sycamore wood during winter 2003. The highest FNdfa

was calculated to be 100% and this was evident when using leaves in autumn of the three reference trees and roots, wood blackthorn and sycamore in the summer, and roots in the winter (Table 3.4). Although the results reported by Binkley *et al.* (1985) showed relatively similar depleted values of  $\delta^{15}$ N in red alder, the authors could not conclude that red alder fixed most of the nitrogen within the plant (Binkley *et al.*, 1985). Nitrogen fixation estimated using <sup>15</sup>N enriched nitrogen fertilizer yielded relatively lower fixed nitrogen at 21% in red alder (Khurdali, 2000). Work reported elsewhere, using nitrogen-15 natural abundance method, however, showed similar results to that of the present study. For example, Domenach, *et al.* (1989) reported that *Alnus incana* fixed 75%, and *Alnus glutinosa* 97% of the nitrogen found in their leaves.

## 3.7.2 Variation between seasons in $\delta^{15}$ N and FNdfa

The results of the present study showed that  $\delta^{15}$ N and FNdfa varied between seasons. Red alder yielded low  $\delta^{15}$ N values (less than zero) in the summer and autumn and positive but low values in the winter and spring periods. FNdfa also showed remarkable decline in the winter and spring periods. Watt *et al.*, (2003) reported  $\delta^{15}$ N and FNdfa in broom (*Cytisus scoparius* L.) to be high with high temperatures during mid-summer and depressed in spring. The authors also reported that FNdfa were low after midsummer due to moisture shortage (Watt *et al.*, 2003). FNdfa values were reported to remain low in the winter when temperatures very low.

Comparison with nitrogen fixation estimates carried out using acetylene reduction assay method also showed that mean nitrogenase activity in red alder nodules was high in summer and autumn when temperature and moisture regimes were favourable and significantly reduced in the winter periods probably due to low temperature (Teklehaimanot and Martin, 1999; Tripp *et al.*, 1979). The daily variation in acetylene reduction was also depicted by elevated activity at midday when temperatures were high and dropped with declining temperatures at night (Tripp *et al.*, 1979). Therefore, favourable environmental conditions such as available soil moisture, optimum temperature and light regimes may be contributing to the high proportions of nitrogen fixation occurring in the summer and autumn periods in red alder. Such conditions tend to favour metabolism in plants, which in turn favours nitrogen fixation in plants (Hawkins and McDonald, 1994; Sayed *et al.*, 1997). Sayed *et al.* (1997) found that optimal temperature for *Frankia* in *Alnus spp* was 25°C and performed very poorly at a higher temperature of  $37^{\circ}$ C.

## 3.7.3 $\delta^{15}$ N in root nodules of red alder and soil of the study site

Root nodules of red alder and soil of the study site showed enriched values of nitrogen-15. This shows that nodules depend on soil nitrogen. The results of the present study are also consistent with those reported by Tjepkema *et al.*, (2000) who reported that nodules of *Alnus glutinosa* were consistently enriched in <sup>15</sup>N relative to the rest of the plant parts. The authors also reported that the nodules of *Parasopania rigida* were enriched in <sup>15</sup>N values but root nodules of other actinorhizal plants such as *Casuariana cunninghamiana*, *Datisca glomerata*, *and Myrica gale* were depleted in <sup>15</sup>N (Tjepkema *et al.*, 2000). The values of  $\delta^{15}$ N of the nodules in the present study were dissimilar to those reported by Domench *et al.*, (1988) who reported negative delta nitrogen values in actinorhizal nodules. The findings in the present study of enriched  $\delta^{15}$ N values (5.95 °/<sub>00</sub>) in the study site soil samples are consistent to those reported in the literature. Kreibich and Kern (2000) reported that  $\delta^{15}$ N on forest soils ranged between 5.0 and 5.8°/<sub>00</sub>. Piccolo *et al.* (1996) reported slightly higher  $\delta^{15}$ N values between 8 and 23 in forest soils in Brazil. Vitousek *et al.*, (1989) also measured negative <sup>15</sup>N values in soils in a young site and values of up to 4.2°/<sub>00</sub> in an old site in Hawaiian rainforest. Values of  $\delta^{15}$ N, as high as 10°/<sub>00</sub>, were reported in soils in Kenya by Gathumbi *et al.* (2002). Soil  $\delta^{15}$ N has also been found to be higher than  $\delta^{15}$ N in roots, wood and leaves of plants in Tanzania (Hogberg, 1997). The enrichment of soils may be due to ammonia volatilization during decomposition, plant uptake or leaching of NO<sub>3</sub><sup>-</sup>. These processes tend to enrich the remaining N with <sup>15</sup>N and the preferential selection for <sup>14</sup>N over <sup>15</sup>N during nitrification or denitrification (Hogberg, 1997; Piccolo *et al.*, 1996).

## 3.7.4 Nitrogen content of plant parts and soil of the study site

The results of the present study showed that leaves contained high nitrogen content ranging from 2.39% in red alder to 3.36% in willow. Hurd *et al.* (2001) reported similar quantities of nitrogen in leaves of sitka alder. Wood samples yielded the lowest nitrogen content of less than 1%. This was expected as wood is more ligneous than leaves and roots. The nitrogen content of root nodules of red alder was 1.85% and this was lower than 3.28% reported for *Alnus incana* by Hurd *et al.*, (2001). This may be due to difference in species and age of trees. Nitrogen content of all plant parts was higher than that of the soil, which was 0.29% and this was similar to nitrogen content of soils of other sites in North Wales (Emmett *et al.*, 1995).

#### 3.7.5 Fine root and nodule weight density

In the present study fine root weight density was observed to be higher in the autumn and summer seasons than in the winter and spring in both the agroforestry and forestry plots. The high value of root weight density in the autumn and summer may be attributed to the conducive environmental conditions of high moisture content and optimal temperatures during these seasons. This finding is in agreement with results published in the literature (Makkonen and Helmissaari 1999). Baddeley and Watson (2004) studied seasonal patterns in fine root production and mortality in Scotland and reported that the highest occurrence of roots and net production was in the summer period.

There was significant difference in both live and dead root weight densities between treatments with forestry plots giving the highest values. This may be due to higher tree planting density in forestry where roots of adjacent trees may overlap. The reduced root weight density in agroforestry may also be due to the effect of soil compaction by livestock and competition with roots of pasture. Sheep (more than 12 ewes plus their lambs per hectare) grazed in the agroforestry plots every year from spring to autumn (Teklehaimanot *et al.*, 2002). This may have compacted the soil as such increasing the bulk density of the soil, which in turn may affect fine roots in agroforestry (Bezkorowanjnyj *et al.*, 1993). Red alder has also been reported to have shallow, fibrous and laterally wide spreading root system when it is grown alone as in forestry (Binkley *et al.*, 1985). However, the presence of ryegrass in agroforestry, which has a dense, shallow and fibrous root system may compete and affect the fine root weight density of red alder.

There was no significant difference in either live or dead nodule weight density between treatments, seasons and distance from tree in both agroforestry and forestry. However, different patterns of nodule weight density with respect to season and distance from the tree were observed between agroforestry and forestry. This may be due to the very high seasonal variability and uneven distribution of root nodules in samples collected as evidenced by very high standard errors in both live and dead nodule weight densities. A significant proportion of samples lacked root nodules. High standard errors in root nodule counts were observed in work done on 2 and 8 year old *Erythrina poeppigiana* in Costa Rica by Chesney and Nygren (2002).

### 3.8 Fixed nitrogen

The results of the present study showed that there was a total of 65.55 and 334.14 kg ha<sup>-1</sup>of fixed nitrogen accumulated in red alder trees in agroforestry and forestry, respectively. This includes fixed nitrogen in leaves, wood and roots. When leaves were taken alone, as most of the studies on nitrogen fixation by previous workers are based on leaves, the results of the present study showed that the amount of nitrogen derived from the atmosphere in leaves was 9.22 and 57.63 kg N ha<sup>-1</sup> in agroforestry and forestry, respectively and this is comparable with reports in the literature for alder and other tree species. According to Sylla *et al.* (2002) the contribution of nitrogen derived from fixation in leaves of *Pterocarpus lucens* in Senegal reached 28.9 kg N ha<sup>-1</sup> in ferruginous soil. Coté and Camire (1984) estimated fixed- N<sub>2</sub> in leaves to be 53 kg N ha<sup>-1</sup> in both mixed and pure stands of alder grown in Canada. Sougoufara *et al.* (1990) estimated fixed nitrogen in leaves to be 40-60 kg N ha<sup>-1</sup> for *Casuarina equisetifolia*.

The results of the present study also showed that the rate of nitrogen fixation by red alder was 30.95 and 117.84 kg ha<sup>-1</sup> yr<sup>-1</sup> in agroforestry and forestry, respectively. This finding is similar with values reported for red alder forestry stands in the literature. Binkley *et al.* (1994) reported that the rate of nitrogen fixation by red alder ranged between 60 and 150 kg ha<sup>-1</sup> yr<sup>-1</sup> and these are based on acetylene reduction assay.

## 3.9 Contribution of leaves, dead roots and root nodules to soil organic matter and nitrogen content

According to the results of the present study, senescent leaves, fine roots and dead root nodules could potentially contribute 4.0 t ha<sup>-1</sup> yr<sup>-1</sup> organic matter to the soil in agroforestry and 9.1 t ha<sup>-1</sup> yr<sup>-1</sup> in the forestry systems. The amount of organic matter deposited in the soil from senescent leaves alone, because most of the previous studies are based on leaves only, was estimated between 0.42 and 2.65 t ha<sup>-1</sup> yr<sup>-1</sup>. Comparable amounts of organic matter addition to soil in the form of annual prunings of red alder of 0.9 - 4.5 t ha<sup>-1</sup> yr<sup>-1</sup> (depending on planting density) have been reported by Seiter *et al.* (1999).

The organic matter quantities estimated could potentially contribute 27.10 and 93.81 kg fixed N ha<sup>-1</sup> yr<sup>-1</sup> to the soil in agroforestry and forestry systems, respectively. This contribution is comparable to figures reported in the literature. The values in the literature vary between 13 and 164 kg fixed N ha<sup>-1</sup> yr<sup>-1</sup>. The lowest accretion of fixed nitrogen by red alder (13 kg ha<sup>-1</sup> yr<sup>-1</sup>) to forest soil was reported by Berg and Dorksen (1975) in Western Oregon, USA. Luken and Fonda (1983) measured the highest fixed nitrogen accretion due to red alder (164 kg ha<sup>-1</sup> yr<sup>-1</sup>) in forest soils of Washington, USA.

### 3.10 The C:N ratio and decay rates in alder

The C:N ratio was low in the leaves of red alder with differences between senescent leaves and green leaves. Gwozdz (2003) reported C:N ratio of 27.88 in senescent leaves of red alder. The low C:N ratio in red alder leaves may have impact on decomposition on the rate of decomposition and subsequent nitrogen additions to the soil. However there is little information on C:N ratio and rate of decomposition in leaves of *Alnus* species. Compton and Cole (2001), reported that red alder had a decay rate, k of -0.26 and with 8% of the organic matter lost in a period of 4 months. Different rates of decay have been reported in *Alnus glutinosa* as shown in the Table 3.10 below. Differences in decay may be influenced by other factors such as leaf nutritional quality, colonisation by microorganisms and decay rates are expected to change on temporal basis (Compton and Cole, 2001).

## Table 3.10 Decay rate and weight loss in Alnus glutinous

k	Weight loss (%)	Source
-0.0097	37.75	Lopez et al., 2001
-0.0161	*	Canhoto and Graca (1996)
-0.908	*	Pereira et al., (1998)

\* Data not available

### **CHAPTER 4.0**

## FINE ROOT LENGTH DENSITY DISTRIBUTION AND DYNAMICS IN RED ALDER

### 4.1. Introduction

Distribution of fine roots is an important factor influencing competition between trees and inter-planted crops in agroforestry systems (Gautam, *et al.*, 2002; Scroth, 2003). Lower tree surface root density would be an attractive feature because it would probably reduce the competition between crop plants and trees. In agroforestry systems in general, fine root distribution depends on plant species, age of the tree, planting density and management practices, among other factors (Tomlinson *et al.*, 1998; Bayala *et al.*, 2004). The spatial distribution of fine roots is, therefore, an important characteristic of trees to understand in order to improve the management and sustainability of agroforestry systems.

In silvopastoral systems many perennial grasses that establish a dense, shallow, fibrous root system compete severely with trees. This may consequently influence fine root distribution of trees. Grazing animals also may influence tree root distribution as a result of soil compaction and browsing of the tree component (Bezkorowajnyj *et al.*, 1993). No study has, however, been reported so far on fine roots of red alder in general and in particular on the effect of planting density on the distribution and dynamics of fine roots of red alder when planted in silvpastoral systems. The work reported on fine roots within the genus *Alnus* is by Rytter (1989) who reported fine root distribution of

intensively managed 2-year old stands of grey alder (*Alnus incana*) to be shallow with more than 90% of the roots being concentrated in the upper 10 cm of the soil. Dhyani and Tripathi, (2000) reported changes in fine root densities through different seasons in *Alnus nepalensis* in an agrisilvicultural practices in north-east India.

Information on variations in root distribution and density is needed in order to design best management practices of silvopastoral systems that include red alder.

The present study was, therefore, carried out to study the effect of planting density on the distribution and dynamics of fine roots in red alder.

## 4.2. Methodology

Three red alder trees from each of the forestry and agroforestry treatment plots were randomly sampled for this study. Trees in the border were unacceptable to avoid edge effects. Under each selected tree soil samples of 14 cm by 14 cm were excavated in 5 cm increments up to 15 cm soil depth in four equal directions around each tree at three distances in each direction (0.50 m, 0.75 m and 1 m) from the base of each tree. A total of 36 samples were collected under each tree species during each season (autumn, winter, spring and winter), starting from the 2002 to 2003. The samples were stored in a cold store for approximately one week before separating them into fine roots and soils in the green house as described in Section 3.2.2 (Plate 4.1). The live fine roots were taken to laboratory and were stored in 50% IMS before scanning them in the laboratory of the University of Wales Bangor, UK. Live fine roots were scanned using WIN RIZO computer software (Regent Instrument Inc., Quebec, Canada) to determine the total

root length. This was achieved by placing the roots in water in a tray and then scanning them in a HP scanjet 4c/T (Hewllet Packard, Palo Alto, CA, USA) (Plate 4.2).



Plate 4.1 Root washer



Plate 4.2 Hewllet parkard scanjet 4c/T root scanner connected to computer.

## 4.3. Analysis of data

ANOVA of Minitab 13 software package was applied to the root data to make comparison between red alder treatments, soil depth, root size categories, distance from the tree and seasons. Regression analysis was also carried out to establish the relationship between fine root length density and soil depth and between fine root length and root size.

## 4.4. Results

## 4.4.1 Fine root length density of red alder according to tree planting density

Fine root length density (RLD) in agroforestry was significantly lower (P<0.001)  $(0.36\pm0.01 \text{ cm cm}^{-3})$  than in forestry treatment  $(0.41\pm0.01 \text{ cm cm}^{-3})$  (Figure 4.1).



Figure 4.1. Root length density (cm cm<sup>-3</sup>  $\pm$ SE) in agroforestry and forestry.

### 4.4.2 Fine root length density (RLD) of red alder in relation to season

The results of ANOVA indicated that there was significant difference (P<0.05) in RLD between seasons in agroforestry but not in forestry. The highest RLD was recorded in autumn and the least in summer in agroforestry (Figure 4.2). RLD in forestry, however, remained more or less constant throughout the four seasons (Figure 4.2).



Figure 4.2 Fine root length densities over the four seasons, from autumn 2002 to summer 2003 (cm cm<sup>-3</sup>±SE) in agroforestry (Ag) and forestry (Fo).

## 4.4.3. Fine root length density of red alder in relation to soil depth

There was a significant difference (P<0.001) in RLD between soil depths (5, 10 and 15 cm) (Figure 4.3) in both agroforestry and forestry. There were also significant interactive effects of season and soil depth (P<0.01) and treatment and soil depth (P<0.001) on RLD. The highest RLD of  $0.61\pm0.03$  cm cm<sup>-3</sup> was recorded at the topsoil depth of 0 - 5 cm in autumn and the least at the soil depth of 10-15 cm in summer in agroforestry ( $0.20\pm0.01$  cm cm<sup>-3</sup>). Nearly 50% of fine roots were concentrated in the topsoil depth of 0-5 cm or more than 79% in the upper 10 cm of the soil.

RLD significantly declined with increasing soil depth as shown by the regression equations in both agroforestry (equation 4.1) and forestry (equation 4.2).

Agroforestry RLD = -0.0542(soil depth) + 0.2559 R<sup>2</sup> = 0.9418 (4.1) Forestry RLD = 0.0706(soil depth) + 0.2707 R<sup>2</sup> = 0.8979 (4.2)



Figure 4.3 Fine root length density (cm cm<sup>-3</sup>  $\pm$ SE) at different soil depths

## 4.4.4 Fine root length density of red alder in relation to distance from tree

Figure 4.4 shows that RLD declines slightly with respect to distance from tree. ANOVA showed no significant difference in RLD between different sampling distances in both agroforestry and forestry.



# Figure 4.4 Fine root length density (cm cm<sup>-3</sup> ±SE) with increasing distance from tree

## 4.4.5 Fine root length of red alder according to root diameter size classes

Fine root length differed significantly (P<0.001) between diameter size class categories of fine roots. The highest root length of  $165.07\pm5.02$  cm was recorded for size class <0.05 mm in forestry treatment. Nearly 40% of the roots in both forestry and agroforestry were less than 0.05 mm in diameter. As shown in Figure 4.5, root length declined with decrease in root size. Regression equations derived also showed very strong relationship between root length and size classes in both agroforestry (equation 4.4).
Agroforestry RL = 194 - 42.2 Size  $R^{-2} = 98.7\%$  (4.3)

Forestry RL = 208 - 43.2 Size R<sup>-2</sup> = 99.5% (4.4)



Figure 4.5: Relationship between fine root length and size classes.

#### 4.5. Discussion

#### 4.5.1 Fine root length density between treatments

The higher RLD in forestry than in forestry may be due to the higher planting density in forestry treatment (2500 tree ha<sup>-1</sup>). Fine roots of adjacent trees in closely planted trees may overlap or exploit the same soil volume. Tree spacing and spatial arrangement of trees have been reported to have effect on the distribution of fine roots in tree plantations (Gautam *et al.*, 2002). Eastham and Rose (1990) reported high fine root

densities in trees grown at high planting densities and lower densities at the low planting densities. Competition from grass roots may also result in reduced abundance of fine roots of trees in the upper most layers of soil in agroforestry systems as in the present experiment (Gautam *et al.*, 2002; Livesly *et al.*, 2000). The difference in fine roots between the two treatments in the present study may also be due to other factors such as treading by sheep in the agroforestry plots, which may result in compaction of the soil, which in turn may reduce RLD. Sheep (about 12 ewes ha<sup>-1</sup> plus their lambs) grazed the agroforestry plots from spring to autumn of every year in the present experiment.

#### 4.5.2 Horizontal distribution of fine root length density

The lack of significant difference in fine root length density with increasing distance from the tree differed from that reported in the literature (Dhyani and Tripathi, 2000; Jose *et al.*, 2001; Rytters, 1989) who reported strong declines in root length density with distance. The result of the present study is, however, similar with that reported by Bayala *et al.*, (2004) for parkland trees with extensive rooting system. Alder has been reported to form shallow, fibrous and horizontally wide spreading rooting systems (Rytter, 1989). The 1 m distance from the tree to study root distribution as in the present experiment may be too short a distance to observe any difference in root length density.

#### 4.5.3 Vertical distribution of fine root density

The decrease in fine root length density (RLD) with increasing soil depth observed in the present study is in agreement with reports in the literature (Bayala *et al.*, 2004; Lehmann and Zech 1998; Rytter 1989, and Tufekcioglu *et al.*, 1999). Report by Dhyani and Triphathi (2000) showed that the bulk of fine roots of *Alnus nepalensis* were concentrated in the upper 10 cm of the soil with a decrease to 28% between 10-20 cm layer of the soil. The results of the present study confirmed this for red alder as 79% of fine roots were concentrated in the top 10 cm of soil depth.

#### 4.5.4 Fine root length density in relation to season

The highest RLD recorded in autumn in agroforestry in the present study is consistent with results reported by Dhyani and Tripathi (2000) who recorded the highest fine root densities for *Alnus nepalensis* in the autumn season. Work published by Tufekcioglu *et al.*, (1999), also showed low counts of fine roots in the winter/spring seasons and increased from summer through to the highest count in autumn. The low RLD in summer in agroforestry in the present study may also be due to competition with pasture, which was heavily grazed by sheep in the summer. Tree roots may have resumed growth when sheep started to be removed from the plots by the end of summer and the beginning of autumn.

#### **CHAPTER 5.0**

#### WOOD QUALITY OF RED ALDER IN COMPARISON WITH SYCAMORE

#### 5.1 Introduction

Red alder (*Alnus rubra* Bong.) is one of the few temperate hardwood tree species that can be grown to useful timber in a relatively short rotation (25 to 35 years). Wood from red alder is used to make pulp, furniture, cabinets, pallets and has been reported to be satisfactory for oriented strand-boards, studs and turned products (Lei *et al.*, 1997). The wood can potentially be used as biomass energy in the production of electricity (Perem *et al.*, 1981).

A few studies have been carried out on red alder to evaluate the mechanical properties of its wood when it is grown at a spacing of less than 3 m (Gartner *et al.*, 1996; Lei, 1996; Lei *et al.*, 1997). Evans *et al.* (2000) tested the modulus of rupture (MOR) and modulus of elasticity (MOE) of juvenile and mature wood in the stems of red alder. They found that both strength properties showed significantly higher values when trees were older than ten years of age (Evans *et al.*, 2000). Evans *et al.* (2000), Lei *et al.* (1997), Gartner *et al.* (1996) and Harrington and DeBell (1980) compared the specific gravity of wood from different provenances of alder trees with different growth rates. The authors concluded that there were no significant differences in specific gravity between wood of trees growing at different growth rates and between different provenances of red alder. There are also few studies that report the fuelwood value of red alder wood. According to Harker *et al.* (1982) the energy value of red alder is 18 608 kJ kg<sup>-1</sup>. Tillman (1987) reported slightly higher value for red alder at 19 300 kJ kg<sup>-1</sup>. He also reported ash content of 0.41% in wood of red alder.

However, no work has been reported on the effect of wide tree spacing as found in certain agroforestry systems, on the physical and mechanical properties of red alder wood. Studies carried out on other tree species indicated that initial planting density has no significant effect on wood quality but most such studies were carried out on spacing less than three metres wide. Work carried out by McAllister *et al.*, (1997) showed that there was a decrease in modulus of elasticity (MOE) with increasing spacing in one category of tested wood and no significant difference in other mechanical properties. Work carried out on *Picea glauca* showed that density of wood was highest in wood from the trees with narrow spacing (Yang, 2002).

The present study was carried out to determine the effect of initial tree planting density on the physical and mechanical properties of wood from red alder in comparison with sycamore. Sycamore was chosen for comparison because it was planted at the site in the same density as red alder. Sycamore is a widely grown timber species in the UK and it is also known to have good strength properties and high stability (Aaron *et al* 1990). In the UK, sycamore is used for kitchen implements, draining boards, chopping blocks, furniture, joinery, toy making and as fencing material (Aaron *et al.*, 1990). Sycamore has a energy value of 18 585 kJ kg<sup>-1</sup> (Harker *et al.*, 1982) and is widely used as a source of biomass energy. Thus, this makes sycamore an ideal species to be used for comparison. Fuelwood under storage interacts with moisture within the storage area and will move to equilibrium depending on the moisture content of the ambient air (Sampson and McBeath, 1989). Moisture content will have a bearing on the amount of heat that will be liberated. Moist wood materials will require more heat to drive off moisture before the fuel itself begins to burn. On the other hand very dry fuels may burn readily and will yield high quantities of heat. Work carried out by Murphy and Cutter (1974) showed that wood yielded about half the quantity of energy at 100% moisture content compared to the same wood at 0% moisture content. However, no research has been undertaken to test the energy value and the fuel value index (FVI) of either red alder or sycamore wood at varying wood moisture contents.

Wood density was also reported to influence energy values of fuel wood. Wood obtained from trees with high density was reported to have high energy values and compared with low-density wood (Bhat and Todaria, 1992). Since widely spaced trees are known to have high proportions of juvenile wood (MacDonald and Hubert, 2002), studies are required to determine the energy values of wood at varying planting density of trees.

The objectives of the present study were:

- 1. To study the effect of initial planting density of trees on the physical and mechanical properties of wood of red alder in comparison with sycamore.
- To study biomass energy properties of wood of red alder, at varying wood moisture contents and tree planting density, in comparison with sycamore.

#### 5.2 Materials and methods

#### 5.2.1 Determination of mechanical properties of wood

#### 5.2.1.1 Harvesting of wood and processing

Six trees each of red alder and sycamore (in total 24 trees) were randomly sampled and harvested in September 2003 from two different planting density treatments at the Henfaes Research Centre of the University of Wales Bangor, UK (see Chapter 1). The planting density treatments were 400 trees per hectare which was an agroforestry treatment grazed by sheep and 2500 trees per hectare which was an ungrazed forestry treatment. Two sections of 40 cm long were obtained from two tree height positions, one from the base and the second at breast height of each randomly sampled tree to extract samples used in this test. The sections of the stem were then marked on their cross-section to obtain samples of 30\*30\*500 mm from four cardinal positions of north, south, east and west on each bolt (lavers 1983) (Plate 5.1). Each sample was then marked appropriately to show the height and the cardinal position of its origin. The samples were then kept in a conditioning room at room temperature at  $20\pm3^{\circ}$ Cand  $65\pm2\%$  moisture content until they were dry.

#### 5.2.1.2 Preparation of samples

Air-dried samples were then planed to 20\*20 mm. From this sample, test pieces 300mm long for were obtained for the bending test, 60 mm long for the compression test, whereas samples for testing density were cut to a thickness of 5 mm. The position of each size test piece was determined the length of clear wood within each piece. All

samples were placed in the conditioning room at  $20\pm3^{\circ}$ C and  $65\pm2\%$  to attain moisture content of 12-17% with no further weight change before being subjected to bending and compression tests and density measurements according BS 373 (1957). All tests and measurements were made within the conditioning room to ensure accuracy.



# Plate 5.1. Stems of wood from sycamore (white in colour) and red alder showing distinct red colour stain.

#### 5.2.1.3 Density

After reaching constant weight in the conditioning room, a digital calliper was used to measure the dimensions of the wood samples to calculate their volumes. The samples were then weighed using an electronic balance. The calculated volume was divided by the weight to obtain the density using the formula below:

$$\rho = \frac{m}{v}$$

Where  $\rho$  - density in kg  $m^3$ 

m - mass in kg $v - volume in m^3$ .

#### 5.2.1.4 Compression stress

Compression stress test was carried out using Instron 5500R series (Instron limited, High Wycombe) on wood samples measuring 20x20x60 mm. The Instron was attached to and operated from a computer. Pressure was applied at the speed of 6 mm min<sup>-1</sup>. The force was applied parallel to the grain of the samples and the maximum load was calculated and obtained as an output from the computer.



Plate 5.2 Instron 5500R series during compression test.

#### 5.2.1.5 Bending test

The bending test was carried out using Instron 5500R series (Instron limited, High Wycombe, Bucks) (Plate 5.2). Three point bending test was carried out using wood samples measuring 20x20x300 mm, supported by trunions at the ends and the force applied on the radial section by applying pressure at speed of 6.6 mm min<sup>-1</sup> to the radial side of the wood sample. The Instron was attached to and operated from a computer

using Merlin software. The data output from the computer included maximum load, maximum stress and flex modulus. Modulus of rupture and elasticity were calculated using formulae 1 and 2, respectively as shown below:

$$MOR = \frac{3PL}{2bd^2}$$
(5.2)

Where MOR - modulus of rupture is in N  $\mathrm{mm}^{-2}$ 

- P load in N.
- L the span in mm.
- b width in mm.
- d depth in mm.

$$E = \frac{P'L^3}{4\Delta'bd^3}$$
(5.3)

Where  $E - the modulus of elasticity in bending, N mm^{-2}$ .

- P' the load in N, at the limit of proportionality.
- L-is the span in mm.
- $\Delta$ ' the deflection in mm at the limit of proportionality
- b the width in mm
- d the depth in mm.

#### 5.2.2 Determination of biomass energy values of wood

#### 5.2.2.1 Wood harvesting and sample preparation

Wood left over from the harvested trees of red alder and sycamore for the above study of mechanical properties of wood (see Section 5.2.1.1) was collected at Henfaes Research Centre. Care was taken to avoid decaying wood. The wood pieces were then allowed to air dry in storage. Using a chisel and a hammer, the wood pieces were cut to very small pieces, which were then ground to pass through a 2 mm sieve. The ground wood materials were conditioned to different wood moisture contents following the procedure described by Lide (1999) until there was no more weight change in the samples. For conditioning wood at 0% moisture content, samples were placed in the oven and dried for two days at 80°C. For conditioning at 12% samples were placed in a dessicator with super concentrated solution of lithium chloride. For wood moisture conditioning at 32% and 52% super concentrations of magnesium chloride and sodium dichromate were used, respectively. The moisture content of the conditioned samples was tested at the end of conditioning as described in Section 5.2.2.2 below.

#### 5.2.2.2 Checking the moisture content of the conditioned wood

To check the moisture contents of the conditioned wood samples, one gram of ground wood sample from each dessicator was weighed into a crucible. The crucible was placed in a preheated oven (at 105°C) with the lid off. The crucible was removed from the oven after 24 hours, weighed and the lid replaced. The moisture content was calculated using the formula below:

$$MC = \frac{(m_w - m_o)}{m_w} *100$$
(5.4)

Where  $m_w - mass$  of sample used, grams.

mo – mass of oven dry sample, grams.

#### 5.2.2.3 Energy values

Energy values of red alder and sycamore were tested using a ballistic bomb calorimeter and with deflections read from a galvanometer (Cam Metric, Cambridge, England) (Figure 5.3). Tests were carried out on wood samples of both species at moisture contents of 52, 32, 12 and 0% with 5 samples. The ballistic bomb calorimeter was calibrated following the method described in the instrument manual. Cotton wool thread, 5 cm in length, was used to generate heat and 0.7 g of benzoic acid as a standard material. The energy values of both the cotton wool and the benzoic acid were measured. The values of energy values of cotton wool thread and benzoic acid were then used to derive the calibration constant as follows, and the values are converted to kilojoules:

Calculation of the calibration constant,

1.	Mass of benzoic acid		= W <sub>1</sub> (grams)	
2.	Energy value of benzoic acid	= 6.32	kcal g <sup>-1</sup> .	
3.	Therefore the heat release from the benzoic acid		$= 6.32 W_1 kca$	ıl.
4.	Galvanometer deflection without sample		$= 0_1$ divs.	
5.	Galvanometer deflection without benzoic acid		$= 0_2$ divs.	
6.	Therefore galvanometer deflections with benzoic ac	id	$= 0_2 - 0_1$ divs.	

-----

#### 7. And calibration constant

$$= 6.32 \text{ W1 kcal g}^{-1}$$
  
-----  
 $0_2 - 0_1 \text{ divs}$   
= Y<sub>1</sub>

1.0 g of ground wood sample was weighed into cup, which was then placed inside the bomb. The cup was then carefully enclosed inside using the cover. The cover was screwed in until it was tightly fixed. Oxygen was then pumped slowly and steadily into the bomb to 25 atmospheres. The bomb was then fired to combust the load sample. The amount of heat liberated was read as the maximum deflection on the scale. The energy value of the sample was then calculated according to the formula below:

$$CV = (X_1 - X_2) \frac{Y}{z}$$
 (5.5)

Where,

CV – energy value, kcal g<sup>-1</sup>

 $X_1$  – galvanometer deflection with the sample.

 $X_2$  – galvanometer deflection without sample.

Y - calibration constant.

Z – mass of the sample.



# Plate 5.3 Assembled ballastic bomb calorimeter, showing a galvanometer in the background.

#### 5.2.2.4. Ash content

Four samples of ground wood, each weighing 4 grams were obtained from the samples prepared in section 5.2.2.1, each of red alder and sycamore were weighed into four clean crucibles. The crucibles were weighed with their lids on. The samples were placed in a muffle furnace where they were heated at 450 °C overnight. Upon removal from the furnace, samples were placed in a dessicator to cool. After twenty minutes in the dessicator, the samples and the crucibles were weighed in a sattories electronic balance. The formulae below was then used to calculate ash weight and finally, percent ash content within the sample.

$$A_{\rm w} = \frac{m_3 - m_1}{m_2 - m_1} *100 \tag{5.6}$$

Where  $m_1 = mass$  of a crucible with lid.

 $m_2 = mass of crucible with lid + sample.$ 

 $m_3 = mass$  of crucible with lid + residue after incineration.

#### 5.2.2.5. Fuel value index

Fuel value index (FVI) is an index that ranks fuel according to its energy value, density of the material, ash content and moisture content. High energy value and density tend to increase FVI whereas high ash and moisture contents will reduce this value. There are several modifications of this formula where energy value, moisture content and ash content may each be omitted (Abbot, *et al.*, 1997; Jain and Singh, 1999; Kataki and Konwer, 2002). In the present study, fuel value index was calculated for sycamore and red alder using the formula below (Jain and Singh, 1999).

$$FVI = \frac{CV * density}{AC}$$
(5.7)

CV – energy value, kJ g<sup>-1</sup>.

AC – ash content,  $g g^{-1}$ .

Density, g cm<sup>-3</sup>

#### 5.3 Analysis of results

Analysis of variance was carried out to compare wood density, compression and bending strengths, energy value, ash content and fuel value index between red alder and sycamore and between planting density treatments. Regression analysis was also applied to the data to establish relationships between compression stress and density of wood or between compression stress and wood grain angle and between energy value and wood moisture content. All analyses were done using Minitab release 13 software package.

#### 5.4. Results

#### 5.4.1 Mechanical properties of red alder and sycamore

#### 5.4.1.1. Wood Density

The results of ANOVA indicated that the density of red alder wood was significantly (P<0.001) less than sycamore wood (Table 5.1). Wood samples taken from breast height of red alder trees grown at 2500 stems per hectare gave significantly lower (P<0.05) wood density. There was, however, no significant difference between the two planting densities of sycamore. There was also no significant effect of cardinal position on the wood on density in either species.

Species	Planting	Height of woo	od sample	Mean	Significance
	density				between
	(trees ha <sup>-1</sup> )				treatments
		Base	Breast		
			height		
Red Alder	2500	$0.48 \pm 0.006$	$0.44 \pm 0.007$	0.46±0.005	
	400	$0.52 \pm 0.006$	$0.51 \pm 0.006$	$0.51 \pm 0.004$	
	Mean	$0.50{\pm}0.005$	$0.48 \pm 0.007$	$0.49 \pm 0.004$	*
Sycamore	2500	$0.66 \pm 0.008$	$0.64 \pm 0.008$	$0.65 \pm 0.006$	
	400	$0.66 \pm 0.012$	$0.62 \pm 0.012$	$0.64 \pm 0.009$	
	Mean	$0.66 \pm 0.007$	$0.63 \pm 0.007$	$0.64 \pm 0.005$	ns
Significance				***	
between					
species					

# Table 5.1Wood density (g cm<sup>-3</sup> ± SE) in red alder and sycamore planted at<br/>different densities

Data in the same row or column are not significantly different at p>0.05 = ns, significant at p<0.05 = \* and in the same column p<0.001 = \*\*\*

#### 5.4.1.2 Modulus of rupture and elasticity

Modulus of rupture of red alder wood was significantly lower (P<0.001) than that of sycamore wood (Table 5.2). There was, however, no significant difference in modulus of elasticity between the two species.

There were significant interactive effects of planting density and height of wood sample in red alder on modulus of rupture (P<0.05) and modulus of elasticity (P<0.01). At 2500 trees ha<sup>-1</sup> planting density wood taken from breast height showed lower modulus of rupture (75.28±1.54 MPa) and a higher modulus of elasticity (8550.39±209.41 MPa) whereas at 400 trees ha<sup>-1</sup> planting density wood from the base gave higher modulus of rupture (76.36±1.66 MPa) and a lower modulus of elasticity (7649.47±341.11 MPa). There was also no significant difference in either modulus of rupture or elasticity between wood samples taken from different height, cardinal position and planting density of trees in sycamore.

# Table 5.2Modulus of rupture and elasticity in wood of red alder and<br/>sycamore planted at different densities

	Planting	Modulus	of rupture	Modulus of elasticity (MPa± SE)	
	density	(MPa± SE)			
	(trees ha <sup>-1</sup> )				
Species			Significance		Significance
			between		between
			treatments		treatments
Red alder	2500	73.02±1.19		7843.77±191.56	
	400	74.16±1.23		$7270.95 \pm 209.31$	
	Mean	73.48±0.86	ns	7614.64±145.69	*
Sycamore	2500	90.94±1.31		7601.12±232.21	
	400	88.44±1.62		6987.29±284.26	
	Mean	90.24±1.05	ns	7430.05±187.46	ns
Significance		***		Ns	
between					
species					

Data in the same row or column are not significantly different at p>0.05 = ns, significant at p<0.05 = \* and in the same column p<0.001 = \*\*\*

#### 5.4.1.3. Compression stress

There was no significant difference in compression stress between wood samples taken from either different tree height or cardinal positions. There was, however, significant difference (P<0.001) in compression stress between red alder and sycamore wood (Table 5.3). The highest wood compression strength was observed in wood of sycamore grown at 2500 stems ha<sup>-1</sup> according to the results of ANOVA's interaction analysis. According to the results of regression analysis, no relationships were found between compression stress and density of wood.

# Table 5.3. Compression stress of red alder and sycamore wood planted at different densities

Species	Spacing	Compression strength	Significance between
-		$(MPa \pm SE \text{ of means})$	treatments
Red alder	2500	32.56±0.42	
	400	31.70±0.39	
	Mean	32.13±0.29	ns
Sycamore	2500	38.59±0.61	
	400	34.21±1.27	
	Mean	36.49±0.72	**
Significance between species		***	

Data in the same row or column are not significantly different at p>0.05 = ns, significant at p<0.05 = \*, p<0.01 = \*\*, p<0.001 = \*\*\*

#### 5.4.2. Biomass energy properties

#### 5.4.2.1. Energy value of wood at different wood moisture contents

Tables 5.4 and 5.5 show energy values of sycamore and red alder, respectively. Comparisons between energy values at differing wood moisture contents showed a significant difference between red alder and sycamore at 52% moisture content only (P< 0.01). However at wood moisture contents of 32%, 12% and 0%, there were no significant differences observed in energy values of wood between the two species.

It was observed that there was a significant decline in the energy value as wood moisture content increased in sycamore (Figure 5.1). Regression analysis also showed a strong linear relationship ( $R^2$ =68.7%) between moisture content and energy value in sycamore wood (Figure 5.2) but a very weak relationship ( $R^2$ =8.9%) in red alder (Figure 5.3).

Sample No	ple No Moisture content				
	0%	12%	32%	52%	
1	22876.4	21445.4	19885.8	19885.8	
2	21315.5	20665.5	20275.7	19365.9	
3	22095.3	20795.6	20015.7	19365.9	
4	22095.3	23369.0	19079.9	19339.9	
Mean±SE	22095.3±318.4	21568.9±.623.8	19814.3±257.9	19489.4±132.3	

Table 5.4:Energy values (kJ kg<sup>-1</sup>) of firewood of sycamore at differentmoisture contents (n=4).

Table 5.5:Energy values (kJ kg-1) of firewood of red alder at different<br/>moisture contents (n=4).

Sample No	Moisture content			
	0%	12%	32%	52%
1	20145.7	19885.8	19885.8	20925.6
2	20665.6	20535.6	19755.8	20535.6
3	21315.5	19235.9	20015.7	20015.7
4	23759.0	21029.5	21289.5	20795.6
Mean±SE	21471.4±7991.2	20171.7±390.1	20236.7±354.9	20568.1±201.2



Figure 5.1 Relationship between moisture content and energy value in sycamore and red alder.







Figure 5.3: Regression of energy value against moisture content in red alder

#### 5.4.2.2. Energy value of wood at different tree planting densities

Energy values of wood samples of red alder and sycamore from the two planting densities and conditioned to 0% wood moisture content were tested to find out the effect of planting density on wood energy value. Table 5.6 below shows energy values of the two species at different planting densities. The results of ANOVA showed that there were no significant differences in caloric values between the two species and the wood from the different planting densities.

#### Table 5.6Energy values (kJ kg<sup>-1</sup>) of wood at different planting densities (n=3)

	Acer pseudoplatanus	Alnus rubra
Forestry (2500 stems ha <sup>-1</sup> )	21218.7±848.4	23481.8±1528.0
Agroforestry (400 stems ha <sup>-1</sup> )	22914.1±938.2	21379.8±1780.3

#### 5.4.2.3. Ash content

There was significant difference (P<0.05) in wood ash content between red alder and sycamore. The wood of sycamore gave a higher ash content of 0.97% than that of red alder with ash content of 0.59% (Table 5.7).

#### Table 5.7.Ash content (%) of sycamore and red alder wood (n=4).

Sample	Sycamore	Red alder
1	0.77	0.49
2	1.19	0.51
3	0.83	0.74
4	1.07	0.62
Average and SD	0.97±0.09	0.59±0.06

#### 5.4.2.4. Fuel value index

Tables 5.8 and 5.9 show the fuel value indices at different moisture contents of sycamore and red alder wood respectively. The fuel value index of red alder was significantly higher (P<0.05) than that of Sycamore. There was, however, no significant difference in fuel value index between wood with different moisture contents in either red alder or sycamore.

 Table 5.8.
 Fuel value index of sycamore at different moisture contents (n=4).

Species	Acer pseudoplatanus				
Moisture content	0%	12%	32%	52%	
Energy value	5280.90±76.10	5155.00±149.00	4735.70±61.6	4658.1±31.6	
Density	0.65±0.01	$0.78 \pm 0.01$	0.85±0.01	0.98±0.01	
Ash content	0.97±0.099	0.97±0.09	0.97±0.09	0.97±0.09	
Fuel value index	1480.60	1445.31	1327.75	1398.70	

Species	Alnus rubra			
Moisture content	0%	12%	32%	52%
Energy value	5132.00±191.0	4821.20±93.2	4836.70±84.8	4915.90±48.1
Density	0.45±0.01	$0.50 \pm 0.01$	0.60±0.01	0.68±0.01
Ash content	0.59±0.1	0.59±0.1	0.59±0.1	0.59±0.1
Fuel value index	1637.8	1538.5	1597.6	1568.8

Table 5.9. Fuel value index of red alder at different moisture contents (n=4).

#### **5.5 Discussion**

#### 5.5.1 Wood density in red alder and sycamore

The results of the present study indicated that red alder had lower wood density than that of sycamore. The wood density value of red alder was similar to that reported by Gartner, *et al.*, (1997), Harrington and DeBell (1980) and Lowell and Krahmer (1993). The density value obtained for sycamore was, however, much higher than the value recorded by Lavers (1983). When the value for red alder was compared with most broadleaf and coniferous tree species, it could be ranked as medium density tree species (US Forest Service, 2000).

Initial tree planting density did not have effect on wood density in sycamore. In red alder, however, there was a significant effect and the highest wood density was observed in the agroforestry treatment. Similar contradictory results on the effect of planting density on wood density are also reported in the literature. For example, Lei *et al* (1997) studied the effect of 12 treatment combinations including tree spacing on wood properties of 7-year-old red alder. They found that there was no significant effect of spacing on specific gravity of red alder. But the lack of significant difference in Lei *et al's* (1997) study may be due to the very close spacing between trees, the maximum being 2 m only. On the other hand, Yang (2002) found tree spacing to have significant effect on relative density of wood in *Picea glauca*. Thus, the effect of spacing on wood density may be species-specific in nature.

The results of the present study also indicated that the position of wood on the tree had no effect on wood density. This finding is in agreement with that reported by Gartner *et al* (1997). They studied the effect of height position of wood on wood properties of 40year-old red alder and found that wood height position had no effect on specific gravity of red alder.

#### 5.5.2 Modulus of rupture and modulus of elasticity

The results of the present study showed that red alder wood gave lower value of modulus of rupture (MOR) than that of sycamore. However, although there was no significant difference, red alder wood had higher value of modulus of elasticity (MOE) than that of sycamore. The values of MOE and MOR for red alder were very similar to the values reported by Lie *et al* (1997) and listed in the Wood Handbook of USA forestry service (USA Forestry Service, 1987). The average MOR and MOE values for sycamore were, however, much lower than those measured and reported by Reinprecht *et al.*, (1999) at 139 MPa for MOR and 13360 MPa for MOE. This may be due to the wood of sycamore in the present study having more juvenile material than the wood

tested by Reinprecht *et al.*, (1999). In red alder, however, juvenility has been reported to end at much earlier age. Evans *et al.*, (2000) studied red alder wood properties by taking wood samples from 50-year-old trees from pith to bark to determine the effect of age on red alder wood properties. They found that the value of MOE started to plateau from age 10 and MOR from age 16.

The results obtained in the present experiment also showed that planting density had significant effect on MOE in red alder but not in sycamore. This may be due to the effect of spacing on wood properties being species-specific. Wood in agroforestry treatment of red alder had lower MOE than in forestry treatment but not MOR. Results reported in the literature on the effect of planting density on MOR and MOE are, however, contradictory. Lie *et al* (1997) reported no significant effect of spacing on MOR and MOE in 7-year-old red alder. McAlister et al. (1997), on the other hand, reported that MOR decreased with increased tree spacing but not MOE in *Pinus elliottii*.

Wood obtained from the base of widely spaced trees of red alder had higher MOR and MOE while in closely spaced trees MOR and MOE were higher at breast height. This may be due to the effect of spacing on the relative compositions of compression and tension wood in different parts of trees (Desch and Dinwoodie, 1996). Isolated trees may tend to allocate relatively more compression and tension wood at the base than at breast height.

#### 5.5.3 Compression strength parallel to the grain

In the present study sycamore was found to have higher compression strength than red alder and the highest compression strength of 38.59 MPa was measured in sycamore forestry treatment. The above value for sycamore is, however, very much less than the compression strength of 48.2 MPa compiled by Lavers (1983). The value for red alder is also lower than the value of 40 MPa listed in the Wood Handbook of USA forestry service (USA Forestry Service, 2000). This may be due to difference in the age of trees tested.

#### 5.5.4. Energy values of red alder and sycamore

According to the results of the present study wood moisture content had significant effect on energy value of sycamore but not on red alder. As a result, sycamore wood at 52% wood moisture content gave the least energy value of 19493.26 kJ kg<sup>-1</sup>. At 0% wood moisture content (oven dried wood), there was no significant difference in energy value between sycamore and red alder. Initial tree planting density did not also have effect on energy value in either sycamore or red alder.

The decline in energy value with increasing moisture content has not been reported for either red alder or sycamore in the literature. However, similar results on other tree species have been reported by Murphey and Cutter (1974). Their report showed a steep decline in energy value from 0% to half the energy value at 100% moisture content in seven hardwood species of North Eastern USA. Moisture contained in wood material reduces energy value of wood since a significant amount of energy is initially used to drive off the moisture. The steep decline in moisture content of sycamore may be due to the fact that higher density wood tends to absorb more moisture compared with the same size of wood material of lower density.

Energy value of 21313 kJ kg<sup>-1</sup> for red alder measured in the present study is slightly higher than values reported by Harker *et al.*, (1982), and Tillman, (1987) of 18608 and 19300 kJ kg<sup>-1</sup>, respectively. Energy value of sycamore of 22095 kJ kg<sup>-1</sup> was also slightly higher when compared with that reported by Harker *et al.*, (1977) and Nunez-Regueira *et al.*, (2001) of 18585 kJ kg<sup>-1</sup> and 17978 kJ kg<sup>-1</sup>, respectively. The difference may be due to slight differences in chemical composition of wood from tree to tree and from one place to another and from season to season (Harker *et al.*, 1982; Nunez-Regueira *et al.*, 1997; Nunez-Regueira *et al.*, 2001). Another possibility may be that the values in the literature were measured using old methods and the equipment for measuring energy value has advanced since then. Seasonal differences of 3.5% in energy values were reported in wood of red alder through seasons (Nunez-Regueira *et al.*, 1997; Nunez-Regueira *et al.*, 2001).

#### 5.5.5 Ash content and fuel value index

Sycamore yielded more ash (0.97%) than red alder (0.57%). Tillman (1977) reported a lower ash content of 0.40% in red alder. Ash content of sycamore has not been reported in the literature and as such comparison could not be made.

Red alder wood ranks higher than sycamore wood when the fuel value index (FVI) is considered. The difference in FVI between the two species is due to the ash content. Sycamore wood has a high ash content, which has lowered its FVI considerably. Based on fuel value indices estimated in the present study, red alder and sycamore can be ranked as medium bio-energy producing tree species when compared with other tree species that have been studied elsewhere (Goel and Behl, 1996; Jain and Singh, 1999; Kataki and Konwer, 2002). The highest fuel value index of 4620 was reported in *Tyrus pashia* in India (Jain and Singh, 1999). The lowest value of 370 was reported in *Acacia excelsa* by Kataki and Konwer (2002).

Overall, wood of sycamore yielded superior mechanical properties when compared with red alder, except in modulus of elasticity where red alder wood was higher. Different plant densities did not affect mechanical properties of wood of both tree species. Wood from red alder yielded lower energy values than those of sycamore but had a higher fuel value index. Planting densities did not affect the energy values of wood of both tree species.

#### **CHAPTER 6.0**

### GENERAL DISCUSSION CONCLUSIONS AND RECOMMENDATIONS

#### 6.1 General discussion

The results of the present study showed that red alder, a nitrogen-fixing tree, has potentials to improve soil nitrogen and organic matter balance in silvopasture. Red alder has been planted in a mixture with other trees to improve soil fertility in the Pacific Northwest of the US and other areas (Binkley *et al.*, 1994). The capacity of red alder to colonise a site after disturbance and to subsequently fix nitrogen are also well known facts (Binkley *et al.*, 1985). The present study has confirmed that red alder has also a great potential to be used in silvopasture.

Red alder grown in silvopasture can mitigate organic matter and nitrogen deficiencies in the soil through nitrogen fixation, deposition of leaf litter and root and nodule death and decomposition. Red alder was estimated to contribute 4.0 t ha<sup>-1</sup> yr<sup>-1</sup> organic matter and 27 kg ha<sup>-1</sup> yr<sup>-1</sup> of fixed nitrogen in silvopastoral systems. The inclusion of red alder in silvopasture can, therefore, reduce input and consequently mitigate the negative environmental impact of nitrogen fertiliser.

Nitrogen-15 natural abundance method has been carried out once previously in red alder but without success (Binkley *et al.*, 1994). In the present study, however, nitrogen-15 natural abundance method proved to be a useful method in estimating

nitrogen fixation in red alder. Using this method, it was shown that about 85% of the nitrogen in red alder was due to nitrogen fixation and the rate of fixation was estimated at 30.95 and 117.84 kg ha<sup>-1</sup> yr<sup>-1</sup> in agroforestry and forestry, respectively. There were slight variations in the proportions of fixed nitrogen in leaves and roots through the different seasons. Summer and autumn seasons showed high percentages of fixed nitrogen and the reverse was observed in winter and spring seasons. This was also true in many nitrogen-fixing trees and shrubs studied elsewhere (Sprent and Sprent, 1990). This seasonal difference may due to the conducive environmental conditions in the summer and autumn such as high temperature and rainfall and high photosynthetic activity of host plants, which have been reported to enhance nitrogenase activities of nitrogen-fixing symbiotic bacteria in root nodules (Sprent and Sprent, 1990; Teklehaimanot and Martin, 1999).

The present study showed that fine roots and root nodules of red alder can make substantial contribution to organic matter and nitrogen balance of soil. The change in fine root and nodule weight density observed in the present study through the different seasons is evidence of root and nodule turnover that may take place at least once a year in soil under red alder and consequently their contribution to soil organic matter and nitrogen. It was for the first time that such studies on fine root and nodule dynamics in red alder have been made. In particular, information on nodule weight density and its changes with season is the most important contribution of the present study to the understanding of the nature and performance red alder root nodules. The study showed that there were more live and dead root nodules in silvopasture (880 and 520 kg ha<sup>-1</sup>, respectively) than in forestry (800 and 310 kg ha<sup>-1</sup>, respectively) but with very high standard errors. This large variation in nodule data may be due to the non-random

distribution of root nodules in the soil. Nodules excavated and weighed ranged from small ones, under one gram, to very large ones, which were the size of a tennis ball. Root nodules in red alder have been reported to be indeterminate (Binkley *et al.*, 1994) and as such dead coral masses of nodules were often found together with live nodule masses. Small nodules tended to have a few dead parts and dead nodules increased with the increase in size of the nodule mass.

Fine root length density distribution and dynamics were also for the first time that they were studied in the present experiment. According to Teklehaimanot *et al.* (2002) and also as shown in Table 1.1 (Chapter 1), there was no significant effect of planting density on tree growth of red alder at the study site.

Red alder has a lower C:N ratio. The results showed that the lowest C:N ratio occurred in the summer and the highest in the winter period. The C:N ratio is observed to be to be positively correlated with nitrogen content in the leaves. The C:N ratio in the leaves of red alder could not be compared with the *Alnus glutinosa*, a local *Alnus* tree species since such information was not available in the literature. However, the decay rates from literature show that the two tree species have similar decay rates and variations may be experienced due to variations in other factors.

Although red alder wood showed lower wood density, modulus of rupture and compression strength when compared with that of sycamore, the values derived for red alder were much higher than the values derived for most conifers such as Sitka spruce, hemlock, cedar and several pines and some hardwoods such as ash, aspen, chestnut and elm (US Forest Service, 2000).

127

Conifers have traditionally been the species of choice for use in furniture manufacturing, panelling and studs because of their fast growth. Because of the adverse effect of acid rain on conifers and the consequent acidification of rivers and streams, environmental pressure has forced consideration of alternative sources in the timber marketplace. Red alder, being a fast-growing species, can yield two to three rotations in the time needed for one rotation of some conifers (Lowell and Krahmer, 1993).

Furniture is one of main items manufactured from red alder (Lowell and Krahmer, 1993; Lei *et al.*, 1997). Modulus of elasticity is an accepted criterion of strength of wood used for furniture manufacturing. The results of the present study confirm this for red alder. Although there was no significant difference, modulus of elasticity in red alder was even higher than sycamore at this age.

Because of little or no variation in energy value of oven dried wood (0% wood moisture content) between tree species as also observed between red alder and sycamore in the present experiment, fuel value index (FVI) has been used as a criterion in ranking firewood species (Abbot *et al.*, 1997). Thus, despite the lower energy value of red alder when compared with sycamore, the fuel value index of red alder was much higher than sycamore.

The energy value of sycamore was significantly affected by wood moisture content whereas the energy value of red alder wood remained very high even in wood with moisture content of 52%. This means wood of red alder can be stored with moisture contents of up to 50% without change in energy value.
# 6.2 CONCLUSIONS

The following conclusions have been drawn from the results of the present study:

- a) Red alder grown at Henfaes, North Wales was actively fixing nitrogen from the atmosphere as shown by the delta values, which were negative or very close to zero. As a result, it was concluded that almost 85% of nitrogen in red alder was fixed from the atmosphere and the rate of fixation was comparable to values reported in the literature for red alder and other nitrogen-fixing tree species.
- b) A large biomass of fine roots and nodules were found in the soil of red alder, which were assumed to decompose and contribute to soil organic matter and ultimately to the nitrogen balance of the soil.
- c) Red alder wood has average mechanical properties, when compared with sycamore, which are good for furniture and ordinary non-structural uses such as panelling and studs.
- d) The fuel value index of red alder wood was high when compared with sycamore and therefore wood from red alder can be used for heating homes and generating electricity.

It was, therefore, concluded that the nitrogen-fixing capability, the contribution to soil organic matter, the average wood mechanical properties and the high fuel value index of red alder all make the species a good candidate for incorporation in silvopasture.

# 6.3 RECOMMENDATIONS

# i. Refinement of methods

1. Nodules were not included in the assessment of nitrogen fixation because reference plants did not have nodules. If this was included the rate of fixation estimated could be much higher as the amount of nodule turnover was very high. This may be regarded as one of the drawbacks of the <sup>15</sup>N natural abundance method. It is, therefore, recommended that methods of assessing fixed nitrogen in nodules be developed in future studies.

2. In the present study it was difficult to assess adequately the distribution and dynamics of root nodules. A lot of samples collected did not have root nodules. It is, therefore, recommended that a different approach be applied when sampling for root nodules in future studies.

3. It is possible that the mechanical properties of wood measured in the present study could have been influenced by processing of the wood samples, which is likely to have a bearing on the measurements of dimensions of the specimens. Several specimens were found to be uneven in shape, which may introduce large error in measurement. It is, therefore, recommended that care must be taken in the preparation of specimens in future studies. 4. In the present study, the number of samples used for studying mechanical properties was small due to a few numbers of trees available for harvesting. The natural variability of wood is well known. Not only should a greater number of trees be tested, but the number of specimens from within the tree should be increased in future studies.

#### ii. Further work at the experimental site

1. In the present experiment fine root dynamics was studied at quarterly intervals. However, the rate of turnover of fine roots may take place at shorter intervals. Evaluation of root dynamics at shorter intervals e.g. monthly bases is recommended as it may give a better picture of the dynamics and distribution of fine roots and consequently the contribution to the soil organic matter.

2. Nitrogen fixing trees have been reported to have the ability to alter the <sup>15</sup>N isotopic signature of soil and other plants within the same environment. This aspect was not studied here due to limited financial budget. It is, therefore, recommended for future studies.

3. The quarterly sampling intervals of nitrogen fixation applied in the present study may not adequately show the variations that are likely to occur within one season. Evaluation of nitrogen fixation at shorter intervals e.g. monthly bases using leaves to evaluate the nitrogen fixation is recommended. Such a study will yield a better average value of FNdfa.

131

4. Sheep grazing in the agroforestry plots of the present experimental site may have contributed to the net nitrogen balance of the soil through deposition of organic manure and urine. Such deposition may contribute significantly not only to the net nitrogen balance, but may also have an effect on the net nitrogen fixation by red alder. It is, therefore, recommended the contribution of livestock to the soil nitrogen be studied.

5. Leaf litter dynamics and their potential contribution to soil nitrogen were not explored in the present study. It was only assumed. Leaf litter is subject to various physical and chemical processes, which determine their contribution to the soil organic matter and nitrogen. It is, therefore, recommended that this aspect should be further explored including the percent carbon, nitrogen and the C:N ratio of leaves, the leaf litter and their rate of turnover. Comparison of the C:N ratio of red alder with other alders, their effects and the annual cycle of N contribution to the sward should also be explored.

6. Clover is well known to tolerate flooded and semi-flooded areas. Previously nitrogen fixation in clover was reported to be higher than in red alder when using acetylene reduction assay method. Flooding is reported to affect nitrogen fixation in red alder with fixation levels returning to normal after a few days. Evaluation of nitrogen fixation using natural abundance in flooded and semi-flooded areas has not been carried out to compare red alder and clover. Comparison red alder fixation with clover in flooded and semi-flooded conditions should be carried out so to assess their potential contribution to the soil N in UK soils.

7. Spacing and tree canopy influences the primary production with grass through competition for resources and deposition of organic matter. The root turnover may influence may lead to varying organic matter under the canopy where at the same time the fine roots may compete for moisture, space and other resources differently from the tree bole to the centre of the line. Primary production in grass may also be affected by varying microclimate under the canopy during to reduced solar radiation. It is recommended that such variations in sward productivity should be studied in the future to evaluate the effect of spacing and canopy cover from the tree bole up to 2 metres.

8. Phenological variations are likely to occur in the nitrogen content through the season in the soil, also affecting the overall nitrogen economy of soils under the tree canopy and within the root zones of the tree component where tree spacing allows. This type of study is recommended for the future.

9. It is recommended that overall productivity of the system be evaluated through measurements of sward production and live weight gains in sheep grazed in the plots.

## **REFERENCES:**

- Aaron, J. R. and Richards, E. G. (1990). British Woodland Produce. Stobart Davies, London.
- Abbot, P., Lowere, J., Khofi, C. and Werren M. (1997). Defining firewood quality: a comparison of quantitative and rapid appraisal techniques to evaluate firewood species from a Southern African savanna. *Biomass and energy*, **12(6)**, 429-437.
- Akinnifesi, F. K., Kang, B.T. and Lipido, D. O. (1999). Structural root form and fine root distribution of some wood species evaluated for agroforestry systems. *Agroforestry Systems*, 42, 121-138.
- Arbon, I. M. (2002). World-wide use of biomass in power generation and combined beat and power schemes: Proceedings of the Institution of Mechanical Engineers Part A. *Journal of Power and Energy*, **216**(A1), 41-57.
- Baddeley, J. A. and Watson, C. A. (2004). Seasonal patterns of fine-root production and mortality in *Prunus avium* in Scotland. *Canadian Journal of Forest Research*, 34, 1534-1537.
- Batzli, J. M. and Dawson, J. O. (1997). Physiology and morphological responses of red alder and sitka alder to flooding. *Physiologia Plantarum*, **99**, 653-663.

- Bayala, J. (2002). Tree crown pruning as a management tool to enhance the productivity of parklands in West Africa. A thesis submitted in fulfilment for the Philosophiae Doctor at the University of Wales, Bangor, UK.
- Bayala, J., Teklehaimanot, Z. and Ouedraogo, S. J. (2004). Fine root distribution of pruned trees and associated crops in a parkland system in Burkina Faso. *Agroforestry Systems*, 60, 13-26.
- Bengtsson, C. (2000). Stiffness of spruce wood influence of moisture conditions. Holz Alz Roh-Und Werkstoff, 58(5), 344-352.
- Bengtsson, C. (2001). Variation of moisture induced movements in Norway spruce (*Picea abies*). Annals Forest Science, **58**, 569-581.
- Berg, A. and Dorksen, A. (1975). Natural fertilization of heavily thinned Douglas-fir stand by understory red alder. Forest Research Laboratory, Oregon State University, Corvallis, Research note 56.
- Bergersen, F. J. (1980). Measurement of nitrogen fixation by direct means. In F. J. Bergersen (ed.), *Methods of Evaluating Biological Nitrogen Fixation*, pp 65-110.
- Bezkorowajnyj, P. G., Gordon, A. M. and McBride, R. A. (1993). The effect of cattle traffic on soil compaction in a silvopastoral system. *Agroforestry Systems*, 21, 1-10.

- Binkley, D., Cromack, Jr. K. and Baker, D. D. (1994). Nitrogen fixation by red alder: biology, rates and controls. In D. Hibbs, D. DeBell, and R. Tarrant (eds). *The Biology and Management of Red Alder*. Oregon State University Press, Corvallis, pp 57-72.
- Binkley, D., Sollins, P. and McGill, W. B. (1985). Natural abundance of nitrogen-15 as a tool for tracing alder-fixed nitrogen. *Soil Science Society of America Journal*, 49, 444-447.
- Binkley, D., Senock, R. and Cromack Jr. K. (2003). Phosphorus limitation on nitrogen fixation by *Facaltaria* seedlings. *Forest Ecology and Management*, **186**, 171-176.
- Bhat, B. P. and Todaria, N. P. (1992). Fuelwood characteristics of some Indian mountain species. *Forest Ecology and Management*, **47**, **(1-4)**, 363-366.
- Black, K. E., Harbron, C. G., Franklin, M., Atkinson, D. and Hooker, J. E. (1998). Differences in root longevity of some tree species. *Tree Physiology*, 18, 259-264.
- Boddey, R. M., Peoples, M. B., Palmer, B. and Dart, P. J. (2000). Use of the <sup>15</sup>N natural abundance technique to quantify biological nitrogen fixation by woody perennials. *Nutrient Cycling in Agroecosystems*, **57**, 235-270.

- Böhm, W. (1979). Methods of studying root systems. Ecological studies 33. Springer Berlin.
- British Standard Institution (1957). Methods of Testing Small Clear Specimens of Timber. 2<sup>nd</sup> Revision.
- Cadisch, G., Sylvester-Bradley, R. and Nosberger, J. (1989). <sup>15</sup>N-based estimation of nitrogen fixation by eight tropical forage-legumes at two levels P:K supply. *Field Crops Research*, 22, 181-194.
- Campbell, L. C. (1998). Managing Soil Fertility Decline. In Z. Rengel (ed.), Nutrient Use in Crop Production. Food Products press, New York, pp29-52
- Canhoto, C. and Graca, M. A. S. (1996). Decomposition of *Ecalyptus globulus* leaves and three native species (*Alnus glutinosa, Castenea sativa and Quercus faginea*) in Portugeese low order stream. *Hydrobiologia*, 333 (2): 79-85.
- Chesney, P. and Nygren, P. (2002). Fine root and nodule dynamics of *Erythrina* poeppigiana in alley cropping system in Costa Rica. Agroforestry Systems, 56, 259-269.
- Cole, E. C. and Newton, M. (1986). Nutrient, moisture and light relations in 5-year-old Douglas-fir plantations under variable competition. *Canadian Journal of Forest Research*, 16, 727-732.

- Compton, J. E. and Cole, D. W. (2001). Fate and effects of phosphorus additions in soils under N<sub>2</sub>-fixing red alder. *Biogeochemistry*, 53, 225-247.
- Cote, B. and Camire, C. (1984). Growth, nitrogen accumulation, and symbiotic nitrogen fixation in pure and mixed plantings of hybrid poplar and black alder. *Plant Soil*, 78, 209-220.
- Curt, T., Lucot, E. and Mouchand, M. (2001). Douglas-fir rooting biomass and rooting profile in relation to soil in a mid-elevation area. *Plant and Soil*, **233**, 109-125.
- Danso, S. K. A. (1995). Assessment of biological nitrogen fixation. Fertilizer Research, 42(1-3): 33-41.
- Danso, S. K. A., Bowen, G. D. and Sanginga, N. (1992). Biological nitrogen fixation in trees in agro-ecosystems. *Plant and Soil*, 141, 177-196.
- Desch, H. E. and Dinwoodie, J. M. (1996). *Timber, Structure, Properties, Conversion* and Use. Macmillan, London.
- DEFRA (2002). Consultation exercise on the bioenergy infrastructure scheme. http://www.defra.gov.uk
- De Moraes, P. D., Rogaume, Y. and Triboulot, P. (2004). Influence of temperature on the modulus of elasticity (MOE) of *Pinus sylvestris* L.. *Holzforschung*, 58, 143-147.

- Dhyani, S. K. and Tripathi, R. S. (2000). Biomass production of fine and coarse roots of trees under agrisilvicultural practices in north-east India. *Agroforestry Systems* 8, 107-121.
- Diouf, D., Diop, T. A. and Ndoye, I. (2003). Actinorhizal, mycorhizal and rhizobial symbiosis: how much do we know? *African Journal of Biotechnology*, 2 (1), 1-7.
- Dommergues, Y. R. (1997). Contribution of actinorhizal plants to the tropical soil productivity and rehabilitation. Soil Biology and Biochemistry, 29, (5/6); 931-941.
- Domench, A. M., Kurdali, F. and Bardin, R. (1989). Estimation of symbiotic dinitrogen fixation in alder forest by the method based on natural <sup>15</sup>N abundance. *Plant and Soil*, **118**, 51-59.
- Domench, A. M., Kurdali, F., Daniere, C. and Bardin, R. (1989). Determination of isotopic identity of nitrogen fixed by Frankia associated with Genus Alnus. Canadian Journal of Botany, 66(7), 1241-1247
- Dulormne, M. Sierra, J., Nygren, P. and Cruz, P. (2003). Nitrogen-fixation dynamics in a cut-and-carry silvopastoral system in the subhumid conditions of the Guadeloupe, French Antilles. *Agroforestry Systems*, **59**, 121-129.

- Dunham, R. A. (1996). The influence of growth rate on the wood and stem properties of silver birch (*Betula pendula* Roth.). A thesis submitted in fulfilment for the Philosophiae Doctor at the University of Aberdeen, UK.
- Eastham, J and Rose, C. W. (1990). Tree/pasture interactions at a range of tree densities in an Agroforestry Experiment. I. Rooting patterns. *Australian Journal of Agricultural Research*, 41, 683-695.
- Eissenstat, D. M. and Yani, R. D. (1997). The ecology of root lifespan. Advanced Ecological Research, 27, 1-60.
- Elkan, G. H. (1992). Biological nitrogen fixation systems in tropical ecosystems: An overview. In K. Mulongoy, M. Gueye and D. S.C. Spencer (eds), *Biological Nitrogen Fixation and Sustainability of Tropical Agriculture*, pp.27-40.
- Emmett., B. A., Brittain, S. A., Hughes, S. Gorres, J., Kennedy, V., Norris, D., Rafarel,
  R. Reynolds, B. and Stevens, P. A. (1995). Nitrogen additions (NaNO<sub>3</sub> and NH<sub>4</sub>NO<sub>3</sub>) at Aber forest, Wales: I. Response of throughfall and soil water chemistry. *Forest Ecology and Management*, 71, 45-59.
- Evans, J.W., Senft, J. F., and Green, D. W. (2000). Juvenile wood effect in red alder: analysis of physical and mechanical data to delineate juvenile and mature wood zones. *Forest Products Journal*, **50(7/8)**, 75-87.

- Fawupe, J. A. (1993). Charcoal and fuel value of agroforestry tree crops. Agroforestry Systems, 22, 175-179.
- Forgel, R. (1983). Root turnover and productivity of coniferous forests. *Plant Soil*, **71**, 75-85.
- Forsberg, D. and Warensjo, M. (2001). Grain angle variation: a major determinant of twist in sawn *Picea Abies* (L.) Karst. *Scandanavian Journal of Forest Research*, 16, 269-277.
- Fownes, J. H. and Anderson, D. G. (1991). Changes in nodule and root biomass of Sesbania sesban and Leucaena leucocephala following coppicing. Plant and Soil, 138, 9-16.
- Garcia-Montiel, D. C. and Binkley, D. (1998). Effect of *Eucalyptus saligna* and *Albizia falcataria* on soil processes and nitrogen supply in Hawaii. *Oecologia*, **113**, 547-556.
- Gathumbi, S. M., Cadisch. And Giller, K. E. (2002). <sup>15</sup>N natural abundance as a tool for assessing N<sub>2</sub>-fixation of herbaceous, shrub and tree legumes in improved fallows. *Soil Biology and Biochemistry*, **34**, 1059-1071.
- Gartner, B. L., Lei, H. and Milota, M. R. (1996). Variation in specific gravity of wood within and between trees of red alder (*Alnus rubra* Bong.). Wood and Fibre Science, 29(1), 10-20.

- Gautam, M. K., Mead, D. J., Chang, S. X. and Clinton, P. W. (2002). Spatial variation and understory competition effect of *Pinus radiata* fine roots in a silvopastoral system in New Zealand. *Agroforestry Systems*, 55, 89-98.
- Giller, K. E. (2003). Biological nitrogen fixation. In G. Scroth and F. L. Sinclair (eds), Trees, Crops and Soil Fertility: Concepts and Research Methods. CABI publishing. Wallingford, pp259-270
- Giller, K. E. (2001). Nitrogen fixation in tropical cropping systems. 2<sup>nd</sup> Edition. CABI publishing.
- Goel, V. L. and Behl, H. M. (1996). Fuel quality of promising tree species for alkaline soil sites in relation to tree age. *Biomass and Energy*, 10, (1), 57 – 61.
- Govindarajan, M., Rao, M. R., Mathuva, M.N. and Nair, R. (1996). Soil water and root dynamics under hedgerow intercropping in semi-arid Kenya. Agronomy Journal, 88, 513 – 520.
- Graboski, M. S. and Bains, R. L. (1979). Properties of biomass relevant to gasification; A survey of biomass gasification, vol II – Principles of Gasification. Solar Energy Research Institute, SERI/TR 33-239. Golden, CO.
- Greggory, P. J. (1994). Resource capture by root networks . In J. L. Montieth, R. K. Scott and M. H. Unsworth (eds.), *Resource Capture by Crops*, pp.77-89.

- Griffths, A. P. and McCormick, L. H. (1984). Effects of soil acidity of *Alnus glutinosa* and viability of *Frankia*. *Plant and Soil*, **79**, 429-434.
- Groves, K. W. and Chivuya, A. M. (1989). Fuelwood evaluation of four Australian grown species. In D. J. Boland (ed), *Trees of the Tropics: Growing Australian Multipurpose Trees and Shrubs in Developing Countries*. Australian Centre for International Agricultural Research. Canberra, pp159-170.
- Gualttieri, G. and Bisseling, T. (2000). The evolution of nodulation. *Plant Molecular Biology*, **42**, 181-194.
- Gwozdz, R. (2003). Comparisons of red alder and reed canagrass litter inputs to a Pacific Northwest stream ecosystem. Department of Biology, Western Washington University, Bellingham, WA 98225. pp28.
- Haeussler, S. (1991). Autecology of Common Plants in British Columbia: A Literature Review. FRDA report number 158.
- Haeussler, S. and Tappeiner II, J. C. (1993). Effect of the light environment on seed germination of red alder (*Alnus rubra*). *Canadian Journal of Forest Research*, 23, 487-1491.
- Hansen, J. P. and Vinther, F. P. (2001). Spatial variability of symbiotic N<sub>2</sub> fixation in grass-white glover pastures estimated by <sup>15</sup>N isotope dilution method and the natural <sup>15</sup>N abundance method. *Plant and Soil*, **230**, 257-266.

- Handley, L. L. and Raven, J. A. (1992). The use of natural abundance of nitrogen isotopes in plant physiology and ecology. *Plant Cell and Environment*, 15, 965-985.
- Hardy, R. W. F., Burns, R. C. and Holsten (1973). Applications of the acetyleneethylene assay for measurement of nitrogen fixation. *Soil Biology and Biochemistry*, **5**, 47-81.
- Harker, A. P., Sandels and Burley, J. (1982). Calorific values for wood and bark and a bibliography for fuelwood. Tropical Products Institute. Commonwealth Forestry Institute.
- Harrington, C. A. (1990). Alnus rubra Bong. Agriculture Handbook654, Volume 2;
  Hardwoods. United States department of agriculture. Http/www.na.fs.fed.us/spfo/pus/silvics.
- Harrington, C. A. and DeBell, D.A (1980). Variation in specific gravity of red alder (Alnus rubra Bong.). Canadian Journal of Forest Research, 10, 293-299.
- Hawkins, B. J. and McDonald, S. (1993). The influence of temperature and soil water on growth, photosynthesis, and nitrogen fixation of red alder (*Alnus rubra*) seedlings. *Canadian Journal of Forest Research*, 24, 1029-1032.

- Heilman, P. and Ekuan, G. (1982). Nodulation and nitrogen fixation by red alder and Sitka alder on coal mine spoils. *Canadian Journal of Forest Research*, **12**, 922-97.
- Hendrick, R. L. and Pregitzer, K.S. (1993). The dynamics of fine root length, biomass, and nitrogen content in two northern hardwood ecosystems. *Canadian Journal* of Forest Research, 23, 2507-2520.
- Hogberg, P. (1986). Nitrogen-fixation and nitrogen relations in Savanna woodland trees (Tanzania). *Journal of Applied Sciences*, **23**, 675-688.
- Hogberg, P. (1997). <sup>15</sup>N natural abundance in soil-plant systems. *New Phytologist*, **137**, 179-203.
- Hosie, R. C. (1990). Native Trees of Canada. Fritzherry and Whiteside, Markham.
- Hurd, T. M., Raynal, D. J. and Schwintzer, C. R. (2001). Symbiotic N<sub>2</sub> fixation of *Alnus incana* ssp. *Rugosa* in shrub wetlands of the Adirondack Mountains, New York, USA. *Oecologia*, **126**, 94-103.
- Itagaki, N. Mihashi, H, Ninomiya, S., Yoshida, N. and Esashi, T. (1999). Influence of knots on tensile strength of sugi lamina. *Mokuzai Gakkaishi*, 45(5), 367-374.
- Jain, R. K. and Singh, B. (1999). Fuelwood characteristics of selected indigenous tree species from central India. *Bioresources Technology*, 68, 305-308.

- Johnson, F. D. (1967). Taxonomy and Distribution of Northwestern Alders. In J. M.Trappe, J. F. Franklin, R. F. Tarrant and G. M. Hansen (eds), *Biology of Red Alder*. Proceedings of a Symposium held at Northwest Scientific Association. Fortieth Annual Meeting. Pullman, Washington. April 14-15. Portland, pp 9-22.
- Jones, M., Sinclair, F. L. and Grime, V. L. (1998). Effect of trees and crown pruning on root length and soil water content in semi-arid agroforestry. *Plant and Soil*, 201, 197-207.
- Jose, S., Gillespie, A. R., Seifert, J. R. and Pope, P. E. (2001). Comparison of minirhizotron and soil core methods for quantifying root biomass in a temperate alley cropping system. *Agroforestry Systems*, 52, 161-158.
- Joslin, J. D. and Henderson, G. S. (1987). Organic matter and nutrients associated with fine root turnover in white oak stand. *Forest Science*, **33**, 330-346.
- Jozsa, L. A. and Middleton, G. R. (1994). A Discussion of Wood Quality Attributes and Their Practical Implications. Special publication no. SP-34.
- Kaelke, C. M. and Dawson, J. O. (2003). Seasonal flooding regimes influence survival, nitrogen fixation, and the partitioning of nitrogen and biomass in *Alnus incana* ssp. *rugosa*. *Plant and Soil*, **254**: 167-177.

- Kakei, M. and Clifford, P. E. (2000). Lon-term effects of lime application on <sup>15</sup>N availability to Sitka spruce seedlings grown in pots containing peat soils. *Forestry*, 4, 392-401.
- Kataki, R. and Konwer, D. (2002). Fuelwood characteristics of indigenous tree species of north-east India. *Biomass and Energy*, **22**, 433-437.
- Kern, C. C., Friend, A. L., Johnson, J. M. F. and Coleman, M. D. (2004). Fine root dynamics in developing *Populus deltoids* plantation. *Tree Physiology*, 24, 651-660.
- Killham, K. (1994). Soil ecology. Cambridge press.
- Kliger, I. R. (2001). Sprial grain on logs under bark reveals twist-prone raw material. Forest Products Journal, **51(6)**, 67-73.
- Kurdali, F. (2000). Seasonal nitrogen changes in Alnus orientalis and Populus nigra and N<sub>2</sub> fixation by exotic alder species in Syria. Communication in Soil Science and Plant Analysis, 31(15&16), 2509-2522.
- Kreibich, H. and Kern, J. (2000). Studies on the role of nitrogen fixation in the Varzea floodplain forest preliminary results using the <sup>15</sup>N natural abundance method.
  In R. Lieberei, H. K. Bianchi, V. Boehm and C. Reisdorff (Eds)., German-Brazilian Workshop on Neotropical Ecosystems Achievements and Prospects of Cooperative Research, Hamburg, Germany. Pp 485 488.

- Ladha, J. K. and Reddy, P. M. (2003). Nitrogen fixation in rice systems: state of knowledge and future prospects. *Plant and Soil*, **252(2)**, 151-167.
- Lavers, G. M. (1983). The Strength Properties of Timber. Building research establishment report.
- Laegreid, M., Bockman, O. C. and Kaarstad, O. (1999) Agriculture, Fertilizers and the Environment. CABI International. Wallinford.
- Lei, H. (1995). The effects of growth rate and cambial age on wood properties of red alder (*Alnus rubra*, Bong.), and Oregon white oak (*Quercus garryana* Dougl.).
  PhD dissertation, Departments of Forest Products, Oregon State University, Corvallis.
- Lei, H., Gartner, B. L. and Milota, M. R. (1997). Effect of growth rate on the anatomy, specific gravity, and bending properties of wood from 7-year-old red alder (Alnus rubra). Canadian Journal of Forest of Forest Research, 27, 80-85.
- Lehmann, J. and Zech, W. (1998). Fine root turnover of irrigated hedgerow intercropping in Northern Kenya. *Plant and Soil*, **198**, 19-31.

Lide, D. R. (1999). Handbook of Chemistry and Physics. CRC Press, Boca Raton.

- Livesley, S. J., Gregory, P. J. and Buresh, R. J. (2000). Competition in tree row agroforestry systems. 1. Distribution and Dynamics of fine root length and biomass. *Plant and Soil*, 227, 149-161.
- Lowell, E. C. and Krahmer, R. L. (1993). Effects of lean in red alder trees on wood shrinkage and density. *Wood and Fiber Science*, **25(1)**, 2-7.
- Lopez, B., Sabate, S and Gracia, C. (1998). Fine root dynamics in a Mediterranean forest: effects of drought and stem density. *Tree Physiology*, **18**, 601-606.
- Lopez, E. S., Pardo, I. and Felpeto, N. (2001). Seasonal differences in green leaf breakdown and nutrient content of deciduous and evergreen tree species and grass in a granatic headwater stream. *Hydrobiologia*, **464 (1-3)**: 51-61.
- Lukac, M. and Godbold, D. L. (2001). Short Communication; A modification of the ingrowth-core method to determine root production in fast growing tree species. *Journal of Plant Nutrition and Soil Science*, **164**, 613-614.
- Luken and Fonda (1983). Nitrogen accumulation in a chronosequence of red alder communities along the Hoh river, Olympia National Park, Washington. *Canadian Journal of Forest Research*, 13, 1223-1237.
- Macdonald, E. and Hubert, J. (2002). A review of the effects of silviculture on timber quality of Sitka spruce. *Forestry*, **75(2)**, 107-138.

- Mafongoya, P. L., Giller, K. E. and Palm C. M. (1998). Decomposition and nitrogen release patterns of trees prunings and litter. *Agroforestry Systems*, **38**, 77-97.
- Makkonen, K. and Helmisaari, H. (1999). Assessing fine-root biomass production in Scots pine – comparison of soil core and root in-growth core methods. *Plant* and Soil, 210, 43-50.
- Mariotti, A. Sougoufara, B. and Dommergues, Y. R. (1992). Estimation of nitrogenfixation using natural abundance method in plantation of *Casuarina equistifolia* (Forst). *Soil Biology and Biochemistry*, 24(7), 647-653.
- Martin, K. J., Posavatz, N. J. and Myrold, D. D., (2003). Nodulation potential of soils from red alder stands covering a wide age range. *Plant and Soil*, **254**; 187-192.
- McAlister, R. H., Clark III, A. and Saucier, J. R (1997) Effect of initial spacing on mechanical properties of lumber sawn from unthinned slash pine at age 40. *Forest Products Journal*, 47 (7/8), 107-109.

Merschner, H. (1995). Mineral Nutrition of Higher Plants. Academic press.

Minchin, F. R., Sheehy, J. E. and Witty, J. F. (1986). Further errors in the acetylenereduction assay – effects of plant disturbance. *Journal of Experimental Botany*, 37 (183): 1581 – 1591.

- Mulvaney, R. L. (1993). Mass Spectrometry. In R. Knowles and T. H. Blackburn (eds). Nitrogen Isotopes Techniques. Academic press limited. London, pp.11-54.
- Munoz, F. and Beer, J. (2001). Fine root dynamics of shaded cacao plantations in Costa Rica. Agroforestry Systems, 51, 119-130.
- Murphey and Cutter (1974). Gross heat of combustion of five hardwood species at differing moisture contents. *Forest Products Journal*, **24 (2)**, 44-45.
- Myrold, D. D. and Huss-Danell, K. (2003). Alder and lupine enhance nitrogen cycling in degraded forest soil in Northern Sweden. *Plant and Soil*, **254**, 47-56.
- Newton, M., El Hassan, B. A., Zavitkovski (1968). Role of red alder in western Oregon forest succession. In J. M. Trappe, J. F. Franklin, R. F. Tarrant, and G. M. Hansen (eds.), *Biology of Red Alder*. USDA Forest Services, PNW For. Range Experimental Station., Portland Oregon, pp 73-83.
- Niemiec, S. S., Ahrens, G. R., Willits, S. and Hibbs, D. E. (1995). Hardwoods of the Pacific northwest. Research contribution 8. College of Forestry, Forest Research Laboratory, Oregon State University.
- Nunez-Regueira, L., Rodriguez-Anon, J. and Proupin-Castineiras, J. (1997). Calorific values and flammability of forest species in glacia, continental high mountainous and humid Atlantic zones. *Bioresources Technology*, **61**, 111-119.

- Nunez-Regueira, L., Rodriguez-Anon, J., Proupin-Castineiras, J., and Romero-Garcia, A. (2001). Energetic evaluation of biomass originating from forest waste by bomb calorimetry. *Journal of Thermal Analysis and Calorimetry*, 66, 281-292.
- Nygren, P. and Ramirez, C. (1995). Production and turnover of N<sub>2</sub> fixing nodules in relation to foliage development in periodically pruned *Erythrina poeppigiana* (Leguminosae) trees. *Forestry Ecology and Management*, **73**, 59-73.
- Obertello, M., Sy, M. Laplaze, L., Santi, C., Svistoonoff, S., Auguy, F., Bogusz, D. and Franche, C. (2003). Actinorhizal nitrogen fixing nodules: infection process, molecular biology and genomics. *African Journal of Biotechnology*, **2(12)**, 528-538.
- O'Hara, G. W. (1998). The Role of Nitrogen Fixation in Crop Production. In Z. Rengel (ed.) Nutrient Use in Crop Production. Food Products press, New York, pp115-138.
- Oliveira, M. R., van Noordwijk, M., Gaze, S. R., Brower, G., Bona, S., Mosca, G. and Hairiah, K. (2000). Auger sampling, ingrowth cores and pinboard methods. In A.L. Smit, A. G. Bengough, C. Engels, M. van Noordwijk, S. Pellerin and S. C. van de Geijn (eds), *Root Methods; a handbook*. Springer, Berlin pp.175-210.
- Pandey, C. B., Singh, A. K. and Sharma, D.K. (2000). Soil properties under Acacia nilotica trees in traditional agroforestry systems in Central India. Agroforestry Systems, 49 (1), 53-61.

- Parrotta, J. A., Baker, D.D. and Fried, M. (1994). Application of N-15 enrichment methodologies to estimate nitrogen-fixation in *Causuarina equisetifolia*. *Canadian Journal of Forest Research*, 24(2), 201-207.
- Parsons, R. and Sunley, R. J. (2001). Nitrogen nutrition and the role of root-shoot nitrogen signalling particularly in symbiotic systems. *Journal of Experimental Botany*, 52, 435-443.
- Paynel, F. Murray, P. J. and Cliquet, J. B. (2001). Root exudates: a pathway for shortterm N transfer from clover and rye grass. *Plant and Soil*, 229, 235-243.
- Peoples, M. B., Palmer, B., Lilley, D. M., Duc, L. M. and Herridge, D. F. (1996). Application of <sup>15</sup>N and xylem ureide methods for assessing N<sub>2</sub> fixation of three shrub legumes periodically pruned for forage. *Plant and Soil*, **182**, 257-266.
- Pereira, A. P. Graca, M. A. S. and Molles, M. (1998). Litter decomposition in relation to litter physio-chemical properties, fungal biomass, arthropod colonisation and geographical origin of the plant species. *Pedobiologia*, 42(4), 316-327.
- Perem, E., McBride, C. F. and Keith, C. T. (1981). Commercial woods. In E. J. Mullins and T. S. McNight (Eds). *Candian Woods, Their Properties and Uses*. University of Toronto Press, Toronto. Pp9-44.

- Persson, B., Persson, A., Stahl, E. G. and Karlmats, U. (1995). Wood quality of *Pinus sylvestris* progenies at various spacings. *Forest Ecology and Mangement*, 76, 127-138.
- Piccolo, M. C., Neill, C., Melillo, J. M., Cerri, C. C. and Steudler, P. A. (1996). <sup>15</sup>N natural abundance in forest and pasture soils of the Brazilian Amazon. *Plant and Soil*, 182, 249-258.
- Pitcher, K., Hilton, B. and Lundberg, H. (1998). The ARBRE project: progress achieved. *Biomass and Energy*, **15**, 213-218.

Postgate, J. (1998). Nitrogen Fixation. Cambridge University Press.

- Porter, A.W. (1981). Strength and physical properties of wood. In E. J. Mullins and T.S. McKnight (eds). *Canadian Woods; their properties and uses*. University of Toronto press. Toronto. Pp71-96.
- Pregitzer, K. S., King, J. S., Burton, A. J. and Brown, S. E. (2000). Research review: Responses of tree fine roots to temperature. *New Phytologist*, **147**, 105-115.
- Proe, M. F., Craig, J., Griffiths, J., Wilson, A. and Reid, E. (1999). Comparison of biomass production in coppice and single stem woodland management systems on an imperfectly drained gley soil in central Scotland. *Biomass and Bioenergy*, 17, 141-151.

- Reinprecht, L., Kacik, F. and Solar, R. (1999). Relationship between the molecular structure and bending properties of chemically and thermally degraded Maplewood; I. Decrease of bending properties compared with changes in the basic chemical composition of wood. *Cellulose Chemistry and Technology*, 33, 67-79.
- Resch, H. (1980). Utilization of red alder in the Pacific Northwest. Forest Products Journal, **30 (4)**, 21-25.
- Robinson, D. (2001). <sup>15</sup>N as an integrator of nitrogen cycle. *Trends in Ecology and Evolution*, **16(3)**, 153-162.
- Rojas, N.S., Li, C.Y., Perry, D.A and Ganio, L.M. (2001). Frankia and nodulation of red alder and snowbrush grown on soils from Douglas-fir forests in the H. J. Andrews experimental forest of Oregon. *Applied Soil Ecology*, **17**, 141-149.
- Rytter, L. (1989). Distribution of roots and root nodules and biomass allocation in young intensively managed grey stands on peat bog. *Plant and Soil*, **119**, 71-79.
- Roy, R. N., Misra, R. V. and Montanez, A. (2003). Decreasing reliance on mineral nitrogen – yet more food. *Ambio*, **31(2)**, 177-183.
- Rowe, E. C., van Noordwijk, M., Suprayog D., Hairiah, K., Giller, K. E and Cadish, G (2001). Root distributions partially explain <sup>15</sup>N uptake patterns in *Gliridicia* and *Peltophorum* hedgerow intercropping system. *Plant and Soil*, 235, 167-179.

- Ruess, R. W., Van Cleve, K., Yarie, J. and Viereck, L. A. (1996). Contribution of fine root production and turnover to the carbon and nitrogen cycling in taiga forests of the Alaskan interior. *Canadian Journal of Forest Research*, 26, 1326-1336.
- Ruess, R. W., Hendrick, R. L. and Bryant, J. P. (1998). Regulation of fine root dynamics by mammalian browsers in early successional Alaskan taiga forest. *Ecology*, 79, 2706–2720.
- Sampson, G. R. and McBeath, J. H. (1989). Storing whole wood tree chips in interior Alaska. *Forest Products Journal*, **39**, (2), 53-57.
- Sanborn, P., Preston, C. and Brockley, R. (2002). N2-fixation by Sitka alder in a young lodgepole pine stand in central interior British Columbia, Canada. Forest Ecology and Management, 167, 223-231.
- Sayed, W. F., Wheeler, C. T., Zahran, H. H. and Shoreit, A. A. M. (1997). Effect of temperature and soil moisture on the survival and symbiotic effectiveness of *Frankia* spp. *Biology and Fertility of Soils*, 25, 349-353.
- Sanchesz, P. A and Logan, T. J. (1995) Myth and Science about the chemistry and fertility of soils in the tropics. In: R. Lal and P. A. Sanchez (eds), *Myths and Science of Soils of the Tropics*, pp.34-46. SSSA spec. pub. No. 29, ASA. CSSA, Madison, WI, USA.

- Schroth, G. (2001). Root systems. In G. Schroth and F. L. Sinclair (eds), Trees, Crops and Soil Fertility: Concepts and Research Methods. CABI publishing, pp235-257.
- Scroth G. and Sinclair, F. L. (2003). Impacts of trees on soil fertility of Agricultural soils. In G. Schroth and F. L. Sinclair (eds), *Trees, Crops and Soil Fertility: Concepts and Research Methods*. CABI publishing, pp1-11.
- Schwencke, J. and Caru, M. (2001). Advances in actinorhizal symbiosis: host plant Frankia interactions, biology, and Applications in Arid Land Reclamation: A Review. Arid Land Research and Management, 15, 285-327.
- Schwintzer, C. R. and Tjepkema, J. D. (2001). Effect of elevated carbon dioxide in the root atmosphere on nitrogenase activity in the three actinorhizal plant species. *Canadian Journal of Botany*, **79**, 1010 – 1018.
- Seiter, S., Ingham, E. R., Horwath, W. R. and William, R. D. (1995). Increase in soil microbial biomass and transfer of nitrogen from alder to sweet corn in an alley cropping system. In: J. H. Ehrenreich, D. L. Ehrenreich, and H. W. Lee, (eds), *Growing a Sustainable Future*, Proceedings of 4<sup>th</sup> Conference on Agroforestry in North America, Boise, ID.
- Seiter, S. William, R. D. and Hibbs, D. E. (1999). Crop yield and leaf production in three planting patterns of the temperate-zone alley cropping in Oregon, USA. *Agroforestry Systems*, 46, 273-288.

- Shear, G., Feldman, L., Bryan, B. A., Skeeters, J. L., Kohl, D. H., Amarger, N., Marrioti, F. and Mariotti, A. (1982). <sup>15</sup>N abundance of nodules as an indicator of N metabolism in N<sub>2</sub>-fixing plants. *Plant Physiology*, **70**, 465-468.
- Shear, G. and Kohl, D. H. (1986). N<sub>2</sub>-fixation in field settings: estimations based on natural <sup>15</sup>N abundance. *Australian Journal of Plant Physiology*, **13**, 699-756.
- Shear, G. and Kohl, D. H. (1993). Natural abundance of <sup>15</sup>N: fractional contribution of two sources to a common sink and use of isotope discrimination. In R. Knowles and T. H. Blackburn, (eds), *Nitrogen Isotope Techniques*. Academic press. San Diego, pp.89-122.
- Singh, G. (1998). Effects of herbicides on nodulation, biological nitrogen fixation and growth of peas (*Pisum sativum* L.). A thesis submitted in fulfilment for the philosophaiae doctor at the University of Wales, Bangor.
- Singh, G. and Wright, D. (2003). Faults of determining nitrogenase activity. *Journal of* Agronomy and Crop Science, **189**, 162-168.
- Smith, D. M. (2001). Estimation of tree lengths using fractal branching rules: a comparison with soil coring for *Grevillea robusta*. *Plant and Soil*, **229**, 295-304.
- Smit, A. (2000). Preface in; In A.L. Smit, A. G. Bengough, C. Engels, M. van Noordwijk, S. Pellerin and S. C. van de Geijn (eds) (Ed.) Root Methods; a handbook. Springer Berlin. Pp.iii-vii.

- Sougoufara, B, Danso, S. K. A., Diem H. G., Dommergues, Y. R. (1990). Estimating N<sub>2</sub> fixation and N derived from the soil by *Casuarina equisetifolia* using labelled <sup>15</sup>N fertilizer: some problems and solutions. *Soil Biology and Biochemistry*, **12**, 695-701.
- Spek, L. Y. and Van Noordwijk, M. (1994). Proximal root diameter as predictor of total root size for fractal branching models II. Numeral model. *Plant Soil*, 164, 119-127.
- Sprent, J. I. and Sprent P. (1990). Nitrogen Fixing Organisms; Pure and Applied Aspects. Chapman and Hall.
- Sylla, S. N., Ndoye, I., Gueye, M., Ba, A. T. and Dreyfus, B. (2002). Estimates of biological nitrogen fixation by *Pterocarpus lucens* in a semi-arid natural forest park in Senegal using <sup>15</sup>N natural abundance method. *African Journal of Biotecnology*, 1(2), 50-56.
- Tarrant, R. F. and Trappe, J. M. (1971). The role of *Alnus* in improving the forest environment. *Plant and Soil*, **1971**, 335-348.
- Teklehaimanot, Z. (1997). United Kingdom National Silvopastoral Agroforestry Network Experiment; Henfaes. First five-year report. November 1992 to December 1997. School of Agriculture and Forest Sciences. University of Wales, Bangor.

- Teklehaimanot, Z. and Anim-Kwapong, G. (1996). The potentials of *Albizzia zygia* (D.C.) Macbride for soil amelioration. *Applied Soil Ecology*, **3**, 59-68.
- Teklehaimanot, Z. and Martin, R. (1999). Diurnal and season patterns of nitrogenase activity of red alder in comparison with white clover in silvopastoral agroforestry systems. *Biology and Fertility of Soils*, **28**, 267-270.
- Teklehaimanot, Z. Jones, M. and Sinclair, F. L. (2002). Tree and livestock productivity in relation to tree planting configuration in a silvopastoral system in North Wales, UK. *Agroforestry Systems*, **56**, 47-55.
- Teklehaimanot, Z. and Sinclair, F. L. (1993). Establishment of the silvopastoral network experimental site, Henfaes, Bangor. *Agroforestry Forum*, **4**, 18-21.
- Teskey, R. O. and Hinkley, T. M. (1981). Influence of temperature and water potential on root growth of white oak. *Physiologia Plantarum*, **52**, 363-369.
- Tierney, G. L. and Fahey, T. J. (2001). Evaluating minirhizotron estimates of fine root longevity and production in the forest floor of temperate broadleaf forest. *Plant* and Soil, 229, 167-176.
- Tierney, G. L., Fahey, T. J., Groffman, P. M., Hardy, J. P., Fitzhugh, R. D., Driscoll, C. T and Yavitt, J. B. (2003). Environmental control of fine root dynamics in northern hardwood forest. *Global Change Biology*, 9, 670-679.

- Tillman, D. A. (1987). Biomass combustion. In D. O. Hall and R. P. Overend (eds), Biomass; Renewable Energy. John Wiley and Sons. Chichester, pp.203-219.
- Tillman, D., Rossi, A. J. and Kitto, W. D. (1981). Wood Combustion; principles, processes and economics. Academic press. Washington.
- Tisdale, S. L., Nelson, W. L., Beaton, J. D. and Havlin, J. L (1993). Soil Fertility and *Fertilizers*. MacMillan Publishing, New York.
- Tjepkema, J. (1978). The role of oxygen diffusion from the shoots and nodule roots in nitrogen fixation by root nodules of *Myrica gale*. *Canadian Journal of Botany*, 56, 1365-1371.
- Tjepkema, J. D., Schwintzer, C. R., Burris, R. H., Johnson, G. V. and Silvester, W. B. (2000). Natural abundance of <sup>15</sup>N in actinorhizal plants and nodules. *Plant and Soil*, **219**, 285-289.
- Tjepkema, J. D. and Schwintzer, C. R. (1992). Factors affecting acetylene induced decline during nitrogenase assays in root nodules of *Myrica gale L. Plant Physiology*, 98, 1451-1459.
- Tomlinson, H., Teklehaimanot, Z., Traore, A. and Olapade, E. (1995). Soil amelioration and root symbiosis of *Parkia biglobosa* (Jacq.) Benth. In West Africa. *Agroforestry Systems*, **30**, 145-159.

- Tomlinson, H., Traore, A. and Teklehaimanot, Z. (1998). An investigation of the root distribution of Parkia biglobosa in Burkina Faso, West Africa, using a logarithmic spiral trench. *Forest Ecology and Management*, **107**, 173-182.
- Tripp, L. N., Bezdicek, D. F. and Heilman, P. E. (1979). Seasonal and diurnal patterns and rates of nitrogen fixation by young red alder. *Forest Science*, 25(2), 371-380.

Trosesso, M. A. (2002). Wood energy: the way ahead. Unasylva, 211(53), 3-12.

- Tufekcioglu, A., Raich, J. W., Isenhart, T. M. and Schultz, R. C. (1999). Fine root dynamics, coarse root biomass, root distribution and soil respiration in a multispecies riparian buffer in Central Iowa, USA. *Agroforestry Systems*, 44, 163-174.
- Turner, G. L and Gibson, H (1980). Measurement of nitrogen fixation by indirect means. . In F. J. Bergersen (ed.), *Methods for Evaluating Biological Nitrogen Fixation*, Wiley-Interscience, Chichester, pp111 – 138.
- Upreti, B. R. and van der Horst, D. (2004). National renewable energy policy and local opposition in the UK: the failed development of a biomass electricity plant. *Biomass and Energy*, **26**, 61-69.
- USA Forest Service (2000). Wood Handbook: Wood as an Engineering Material. Handbook 72. Forest Products Laboratory, Madison, Wisconsin.

- Valverde, C. and Wall, L. G. (2002). Nodule distribution on the roots of actinorhizal Discaria trinervis (Rhamnaceae) growing in pots. Environmental and Experimental Botany 47, 95-100.
- Van Noordwijk, M., Brower, G., Meijboom, M., Oliveira, M. R., and Bengough, A. G. (2000) Trench profile techniques and core break methods. In A.L. Smit, A. G. Bengough, C. Engels, M. van Noordwijk, S. Pellerin and S. C. van de Geijn (eds) (Ed.) *Root Methods; a handbook*. Springer, pp.211-234.
- Van Noordwijk, M., Spek, L. Y. and De Willigen, P. (1994). Proximal root diameter as predictor of total root size for fractal branching models I. Theory. *Plant Soil*, 164, 107-117.
- Vinther, F. P., and Jensen, E. S. (2000). Estimating N<sub>2</sub> fixation in grass-glover mixtures of a grazed organic cropping systems using two <sup>15</sup>N methods. *Agriculture*, *Ecosystems and the Environment*, **78**, 139-147.
- Vitousek, P. M., Shear, G. and Kohl, D. (1989). Foliar 15N natural abundance in Hawaiian rainforest: patterns and possible mechanisms. *Oecologia*, 78, 383-388.
- Wall, L. G. (2000). The Actinorhizal symbiosis. Journal of Plant Growth Regulation, 19; 167-182.

- Watt, M. S., Clinton, P. W., Whitehead, D., Richardson, B., Mason, E. G. and Leckie, A. C. (2003). Above-ground biomass accumulation and nitrogen fixation of broom (*Cytisus scooparius* L.) growing with juvenile *Pinus radiata* on dryland site. *Forest Ecology and Management*, **184**, 93-104.
- Winship, L. J. and Tjepkema, J. D. (1985). Nitrogen fixation and respiration by root nodules of *Alnus rubra*, Bong,: effects of temperature and oxygen concentration. *Plant Soil*, 87, 91-107.
- Winship, L. J. and Tjepkema, J. D. (1982). Simultaneous measurements of acetylene reduction and respiration gas exchange of attached root nodules. *Plant Physiology*, **70**, 361-365.
- Wheeler, C. T., Hooker, J. E., Crowe, A. and Berrie, A. M. M. (1986). The improvement and utilization in forestry of nitrogen fixation by actinorhizal plants with special reference to *Alnus* in Scotland. *Plant and Soil*, **90**, 393-406.
- Wheeler, C. T., McLaughlin, M. E. and Steele, P. (1981). A comparison of symbiotic nitrogen fixation in Scotland in *Alnus glutinosa* and *Alnus rubra*. *Plant and Soil*, 61, 169-188.
- Yang, K. C. (2002). Impact of spacing on wood properties. *Taiwan Journal of Forest Science*, 17(1), 13-29.
Yavitt, J. B. and Wright, J. S. (2001). Drought and Irrigation effects on the fine root dynamics in tropical moist forest, Panama. *Biotropica*, 33(3), 421-434.

Young, A. (1997). Agroforestry for Soil Management. CAB International. Wallingford.

- Zitzer, S. F. and Dawson, J. O. (1989). Seasonal changes in nodular nitrogenase activity of *Alnus glutinosa* and *Elaeagnus angustifolia*. *Tree Physiology*, **5**, 185-194.
- Zobel, B. J. and van Buitjitenen, J. P. (1989). Wood variation; its causes and control. Springer-Verlag Berlin.