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## **UNIVERSITY OF SOUTHAMPTON**

#### FACULTY OF NATURAL AND ENVIRONMENTAL SCIENCES

Centre for Biological Sciences



#### GHG Balance and Carbon Mitigation Potential of Bioenergy Crops

by

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Thesis for the degree of Doctor of Philosophy

June 2015

ii

#### UNIVERSITY OF SOUTHAMPTON

## **ABSTRACT**

#### FACULTY OF NATURAL AND ENVIRONMENTAL SCIENCES

#### <u>Biology</u>

#### Thesis for the degree of Doctor of Philosophy

#### GHG BALANCE AND CARBON MITIGATION POTENTIAL OF BIOENERGY CROPS

#### Zoe M Harris

#### ABSTRACT

SRC willow for bioenergy is a promising source of renewable energy for temperate climates such as the UK. Mass deployment of this technology will require substantial land use change, which will have consequential effects on ecosystem services. In order to avoid competition with food, bioenergy production has been proposed for cultivation on degraded or marginal land. In the UK, this land will likely come in the form of ex set-aside grasslands. The aim of this work was to quantify the greenhouse gas (GHG) balance of land use change (LUC) to 2G bioenergy, with a particular focus on SRC willow.

A systematic review and meta-analysis revealed that a conversion from arable cropping to second generation bioenergy results in an increase in soil carbon, whereas a conversion from forest results in a loss of soil carbon. Transitions from grasslands to SRC were shown to be broadly neutral, whereas a transition to perennial grasses such as *Miscanthus* there was a loss in soil carbon. This work highlighted the limited long-term empirical data available surrounding the effects of LUC to bioenergy, with particular uncertainty surrounding grasslands.

A field site was established at commercial scale in the south of England to conduct a sideby-side comparison of the net ecosystem exchange (NEE), soil GHG fluxes and soil properties of an ex-set aside grassland and SRC willow plantation. There was also the opportunity to capture the effects of a commercial harvest of SRC willow on net gas exchange.

After three years of measurements, net ecosystem exchange (NEE) indicated that SRC willow was a net sink for carbon and grassland was a net source. Soil respiration was lower in SRC willow than in grassland. Soil fluxes of nitrous oxide and methane were low at

both sites and did not contribute a significant portion of the GHG balance of these land uses. However, there was net emission of methane from grassland and a net uptake from the SRC willow over the measurement period. Soil carbon in the upper 30 cm portion was higher in grassland than in SRC willow, however for the whole 100 cm profile there was no significant difference between land uses. The effects of a commercial harvest were detected in the SRC willow where there was an increase in NEE and ecosystem respiration ( $R_{eco}$ ). Despite these increases in NEE and  $R_{eco}$ , the site became a net sink of carbon again as soon as 3 weeks post-disturbance. Soil chemistry analysis revealed that a conversion from grassland to SRC willow results in increased bulk density and a lower soil moisture.

Overall, these results suggest that a transition from grassland to SRC willow could result in GHG savings, though this is likely to be highly site and management specific. SRC willow is able to act as a sink for carbon which could have positive implications for climate change mitigation. Soil carbon differences between sites indicate that the SRC willow is still in the recovery phase for soil carbon, and these differences are likely due to the larger input of organic material in the grassland where it is mowed and left on site. Management plays a large part in determining the whole GHG balance of the grassland ecosystem and this will need to be considered for future land use change scenarios. Grasslands remain one of the most viable options for land use change to bioenergy to avoid large loss of soil carbon (such as those observed from forest transitions) and competition with food crops, though decisions to convert land will require a site by site evaluation.

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## **DECLARATION OF AUTHORSHIP**

I, Zoe Margaret Harris declare that this thesis entitled "*GHG Balance and Carbon Mitigation Potential of Bioenergy Crops*" and the work presented in it are my own and have been generated by me as the result of my own original research.

I confirm that:

- 1. This work was done wholly or mainly while in candidature for a research degree at this University;
- 2. Where any part of this thesis has previously been submitted for a degree or any other qualification at this University or any other institution, this has been clearly stated;
- 3. Where I have consulted the published work of others, this is always clearly attributed;
- 4. Where I have quoted from the work of others, the source is always given. With the exception of such quotations, this thesis is entirely my own work;
- 5. I have acknowledged all main sources of help;
- 6. Where the thesis is based on work done by myself jointly with others, I have made clear exactly what was done by others and what I have contributed myself;
- 7. Parts of this work have been published as:

**Harris ZM**, McNamara NP, Rowe RL, Dondini M, Finch J, Perks M, Morison J, Donnison I, Farrar K, Sohi S, Ineson P, Oxley JC, Smith P, Taylor G (2014) Research Spotlight: The ELUM project: Ecosystem Land-Use Modeling and Soil Carbon GHG Flux Trial. *Biofuels*, **5**(2), 111-116.

**Harris ZM**, Spake R, Taylor G Land use change to bioenergy: A meta-analysis of soil carbon and GHG emissions. *Biomass & Bioenergy*. **82**, 27–39.

Dondini M, Richard M, Pogson M, McCalmont J, Drewer J, Marshall R, Morrison R, Yamulki S, **Harris ZM**, Alberti G, Siebicke L, Taylor G, Perks M, Finch J, McNamara NP, Smith JU, Smith P (2015) Simulation of greenhouse gases following land-use change to bioenergy crops using the ECOSSE model. A comparison between site measurements and model predictions. *Global Change Biology Bioenergy*. DOI: 10.1111/gcbb.12298

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Signed:	 
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Date:	 

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## ABBREVIATIONS

Abbreviation	Definition
1G	1 <sup>st</sup> Generation Bioenergy Crops
2G	2 <sup>nd</sup> Generation bioenergy crops
ALCs	Agricultural Land Classes
atm	Atmospheres (barometric pressure)
BD	Bulk Density
С	Carbon
САР	Common Agricultural Policy
ССС	Committee on Climate Change
CE	Carbon Equivalents
СЕН	Centre for Ecology & Hydrology
$CH_4$	Methane
C:N	Carbon: Nitrogen ratio
$CO_2$	Carbon Dioxide
СТАВ	Cetyl trimethylammonium bromide
DECC	Department of Energy and Climate Change
DEFRA	Department for Environment, Food & Rural Affairs
DOC	Dissolved Organic Carbon
DNA	Deoxyribonucleic acid
EC	Eddy Covariance
ESS	Environmental Stewardship Scheme
ETI	Energy Technologies Institute

EU	European Union
FACE	Free Air CO <sub>2</sub> Enrichment
GC	Gas Chromatography
GHG	Greenhouse Gas
GLM	General Linear Models
GM	Gravimetric Moisture
GPP	Gross Primary Production
Gt	Gigatonne
GWP	Global Warming Potentials
HE	Sensible Heat Exchange
ha	Hectare
IEA	International Energy Agency
iLUC	Indirect Land Use Change
IPCC	Intergovernmental Panel on Climate Change
IRGA	Infrared Gas Analyser
IWUE	Irrigation Water Use Efficiency
LCA	Life Cycle Assessment
LE	Latent Heat Exchange
LUC	Land Use Change
MEA	Millennium Ecosystem Assessment
mg	Milligram
Modt	Million oven Dried Tonnes
N <sub>2</sub> O	Nitrous Oxide

NBP	Net
NEE	Net Ecosystem Exchange
INICL	Net Ecosystem Exchange
NEP	Net Ecosystem Productivity
$\mathrm{NH_4}^+$	Ammonium
NO <sub>3</sub> <sup>-</sup>	Nitrate
NPP	Net Primary Productivity
NUE	Nitrogen Use Efficiency
OSR	Oilseed Rape
R <sub>eco</sub>	Ecosystem Respiration
RED	Renewable Energy Directive
RH	Relative Humidity
RPM	Revolutions per minute
RTFO	Renewable Transport Fuels Obligation
SOC	Soil Organic Carbon
SOM	Soil Organic Matter
SRC	Short Rotation Coppice
SRF	Short Rotation Forestry
SSSI	Site of Special Scientific Interest
ТР	Total porosity
USA	United States of America
WFPS	Water-filled pore space
WoS	Web of Science
WUE	Water Use Efficiency

## CHAPTER 1: INTRODUCTION – THE CURRENT STATE OF KNOWLEDGE OF BIOENERGY MANAGEMENT AND LAND USE CHANGE

#### 1.1 Land-Use Change and UK Bioenergy Cropping Systems

#### 1.1.1 Global land use, climate change & increasing demands

Global greenhouse gas (GHG) emissions have increased over 80% between 1970 and 2010, from 27 to 49 Gt CO<sub>2</sub>-eq y<sup>-1</sup>, with the largest increase occurring in the past decade (2.2% increase per year; IPCC, 2014). A rise in GHG emissions has resulted in a global warming of the planet as thermal radiation becomes trapped within the atmosphere subsequently causing global climate change (IPCC, 2007). Global climate change has been acknowledged as one of the largest threats to human health, energy security, food security and biodiversity (IPCC, 2014), with serious financial implications if its effects are not mitigated (Stern, 2008).

It is now recognised that increased global demand for food, water and energy, alongside the predicted rise in global population and changes in climate, are placing natural resources under more pressure than ever before (Beddington, 2009; Godfray *et al.*, 2010). At the centre of this larger demand for food and energy is the availability and sustainable use of a finite land resource that delivers multiple Ecosystem Services (ES) and goods (Valentine *et al.*, 2011).

This need is likely to drive an increase in the area of land dedicated to agriculture, although as in the past, a large proportion of the gains in food production may be achieved through increased crop productivity per unit land area, rather than an increase in area of the landscape that is managed and farmed. Since 1970 global agricultural land area has increased by approximately 5%, whilst crop productivity has increased by more than 50% (FAOSTAT, 2012). Future increases in agricultural land vary from 5-30%, depending on the scenario considered (reported by Smith *et al.*, 2010), but all suggest increased land-use for agriculture, and in contrast to the past fifty years, this food delivery must go hand-inhand with other land-use pressures and in the face of climate change (IPCC, 2007).

Land use and land use change (LUC) both hold very large environmental implications including, but not limited to, reduced carbon stocks, soil quality, water quality and losses

in biodiversity. Sala *et al.* (2000) found that LUC is the largest driver of biodiversity loss globally, closely followed by climate change. LUC was responsible for 6-17% of total anthropogenic GHG emission during the 1990s, equalling 5.9 Gt CO<sub>2</sub>-eq y<sup>-1</sup> (IPCC, 2001).

#### 1.1.2 Energy demand, supply and pressure

The global energy supply is still dominated by fossil fuels with oil, coal and natural gas accounting for 33, 30 and 24% of the global usage, respectively (BP, 2014). Global energy consumption grew at a below-average rate of 2.3% in 2013; however the use of renewables is increasing annually and supplied 2.2% of the total global consumption (BP, 2014; Figure 1.1). Global energy demand is expected to continue rising as the global population rises and technological advances take place. Current predictions indicate that energy demand will rise by 37% by 2040 with usage in Europe, Japan, Korea and North America plateauing, and rising consumption in the rest of Asia, Africa, the Middle East and South America (IEA, 2014).



**Figure 1.1:** World primary energy consumption for 2013 (Taken from BP Statistical Review of World Energy, June 2014).

Investment in renewables continues to increase globally, with increased production, reduction in costs and supporting policies (REN21, 2015). However, investment has slowed in developed countries with only a 3% increase from 2013 to 2014, whereas investment in developing countries increased 36% compared to 2013 (REN21, 2015; Figure 1.2). The number of countries with renewable energy targets also continues to grow,

with 164 with renewable energy targets, and 145 of those with policies in place to support those targets (REN21, 2015).



**Figure 1.2:** Global investment in renewable energy technologies in developed and developing countries, 2004-2014. Source: Renewables 2015 Global Status Report, REN21 (2015).

#### 1.1.3 Role of bioenergy crops in meeting global demand

The International Energy Agency (IEA, 2010) suggests that traditional biomass burning provides approximately one third of the energy in Africa, Asia and Latin America, with this figure being as high as 80-90% in the poorest countries around the world (Chow et al., 2003; Demirbas, 2005). Currently, in a global context, bioenergy is the most significant renewable, contributing 78% of total renewables supply but remains largely under-utilised as an energy source (Somerville, 2007). The magnitude of the 'available, useable resource' as opposed to the 'technical potential' of global biomass availability has recently been questioned in several studies where quantification of the global resource has been attempted and these studies have been brought together by Slade et al. (2011). Some estimates suggest that there is potential to supply between 13-22% of the world's global energy demands by 2050 from biomass (Beringer et al., 2011), whilst the IEA (2009) suggests 50% of global energy demand is technically possible from bioenergy. Slade et al. (2011) have reviewed these studies and given a detailed account of the assumptions underlying these highly contrasting estimates of global biomass potential for bioenergy. Briefly, they include yield assumptions; technology enhancements for yield (including breeding and GM); inputs such as nitrogen fertiliser and water; land conversions that

include soil rich in biodiversity; and carbon and grazing land that may or may not become available.

The biomass resource is considerable and even with moderate future predications, between 10-20% of future energy demand could potentially be supplied from biomass resources (Slade *et al.*, 2011), with 10% considered more appropriate for UK supply by the recent Committee on Climate Change review (CCC, 2011). In the CCC review, four scenarios for land-use were considered to estimate global biomass availability with bioenergy crop deployment ranging from 100 Mha to 700 Mha of bioenergy cropping. The global land area was estimated at 13,000 Mha and agricultural land at 4,200 Mha. Of this approximately 1,550 Mha is currently used for crop growth and it is suggested globally that 500 Mha may be available from abandoned agricultural land.

A consensus for future energy demand suggests an increase over the next few decades of between 30-50% on current-day supply (IEA, 2010), with renewable technologies, including bioenergy, playing an increasing role. Since feedstock supply also impacts on land-use, special consideration has been given in recent years to how this resource might be deployed in future. Certainly, Somerville *et al.*, (2011) estimated that less than 1% of global agricultural land resource was in the past deployed to bioenergy however this is likely to increase in future and it is this increase, alongside that of food production and a requirement to fulfil the development goals of the Millennium Ecosystem Assessment (MEA, 2005), that provides the perfect storm described by Beddington (2009).

Energy from biomass, in addition to enhancing energy security and supply, also has global social and environmental consequences that are wide-ranging and complex. These include the contribution of bioenergy chains to ecosystem services and here, the regulating service of greenhouse gas (GHG) emission and climate regulation is considered alongside the supporting services for biogeochemical cycling of carbon and other GHGs, particularly  $N_2O$ . In the IEA (ETP, 2010) 'blue map' scenario – the scenario to achieve a GHG emissions reduction on 2005 emissions of 50% by 2050 with enhanced energy security, suggests that renewables will contribute 17% of the required reduction. Within this, biofuels meet 20% of total transport fuel demand and contribute to more than 30% of power generation from renewables by 2050. Without the 'blue map', the baseline scenario predicts that GHG emissions will double over the same timeframe, leading to a rise in temperature in excess of 2.4°C which is considered unsustainable (IPCC, 2007). Thus, in a global context, the role of bioenergy in contributing to these important regulating and supporting ES is only just beginning to be considered with limited empirical evidence on

which to base assumptions. Of particular significance is the LUC, or crop transition that is associated with the bioenergy crop. Some transitions may be detrimental (e.g. removal of tropical forest to plant sugarcane), provide no net benefits (e.g. one arable crop exchanged for another), whilst others may be positive transitions with improved GHG mitigation potential (e.g., an annual crop replaced with a perennial crop; Hillier *et al.*, 2009).

#### 1.1.4 Types of bioenergy available

Sources of biomass energy come in several forms: first generation (1G) bioenergy crops which are produced mainly from food crops such as grains, sugar beet and oil seeds; and second generation (2G) bioenergy crops which are dedicated lignocellulosic feedstocks such as short- rotation coppice (SRC), willow and poplar, and perennial grasses such as *Miscanthus* and (in the USA) switchgrass. Second generation bioenergy feedstocks can also include crop/forest residues, wood processing waste and solid municipal wastes. Third generation feedstocks often defined as those from algal growing systems, either as macro-algae or micro-algae in bioreactors and open ponds, are considered to have limited land-use implication for the UK; although their potential may be significant, these third generation (3G) feedstocks are considered beyond the scope of this study. These sources of biomass are summarised in Table 1.1.

#### 1.1.4.1 First generation feedstocks

First generation (1G) feedstocks are those from 'traditional' food crops including wheat, corn, oilseed rape, sugar beet, sugar cane. These crops are generally used to make biofuels, through alcohol fermentation, or biodiesel through transesterification of oils and fats. In 2012, there were over 110 billion litres of biofuels produced globally; the largest producers were the USA with 44 billion litres followed by Brazil with 26 billion litres (EIA, 2015). The UK produced 336 million litres of biofuel for 2012 (EIA, 2015).

#### 1.1.4.2 Second generation feedstocks

Second generation (2G) feedstocks are crops which have been bred and grown exclusively for biomass for bioenergy. There are three main types of 2G feedstocks (i) short rotation coppice (SRC) willow and poplar; (ii) perennial grasses such as *Miscanthus* and switchgrass and (iii) short rotation forestry (SRF) such as alder, birch, beech and eucalyptus. Each type is briefly discussed in the following sections.

**Table 1.1:** Main forms of biomass feedstock in the UK land system. <sup>1</sup> Indicates those crop transitions covered in this study.

	Сгор Туре	Source
Crops	First generation <sup>1</sup>	Wheat grain, oilseed rape, sugar beet
	Dedicated second generation <sup>1</sup>	SRC poplar and willow, <i>Miscanthus</i> , short rotation forestry (SRF) including eucalyptus, alder, ash, birch, sycamore, beech, conifer
	Third generation	Micro-algae and macro-algae (seaweed)
Residues	Forestry	Wood chips, sawdust, bark, brashings
	Arable crop	Straw
Wastes	Wood	Contaminated wood waste
	Organic	Animal manures and sewage sludge, food and garden waste
	Landfill gas	Gas from land-fill sites

#### 1.1.4.2.1 SRC

SRC crops undergo coppicing every 3-4 years which results in a multi-stem plant which can rapidly accumulate biomass, and on average these plantations have a life span of approximately 20 years. SRC crops are also advantageous because they require low inputs (fertiliser, pesticides, herbicides, water) and they do not have many pests and are fairly unsusceptible to disease. Short-rotation coppice and grass cultures are the most promising source of biomass at present (Rowe *et al.*, 2009) and have been shown to be the one of the most energy-efficient carbon conversion technologies to reduce GHG emissions (Styles & Jones, 2007), although there still remains limited experimental data to confirm this statement. They are also a preferred biomass crop over first generation food crops because they produce more biomass per hectare and unlike oilseed crops the entire crop is utilized as a feedstock or to produce fuel. In order for bioenergy crops to present a solution for the future, they need to be both environmentally and energetically viable and outweigh the

energy used in the harvest, transportation and production from feedstocks. For example, when compared to coal, SRC willow is able to yield 36-times more energy than coal per unit of energy input and simultaneously emit 24-times less GHG than coal (Djomo *et al.*, 2011). One review, of over 15 years of research, concluded that the benefits of SRC willow were carbon sequestration into soils, reduced erosion, phytoremediation and lower SO<sub>2</sub> and NO<sub>x</sub> emissions when biomass was co-fired with coal (Abrahamson *et al.*, 2002).

#### 1.1.4.2.2 Perennial grasses

The most commonly used perennial grass species for bioenergy are *Miscanthus*, switchgrass, reed canary grass and *Arundo* (Lewandowski, 2003). *Miscanthus* originates from Southeast Asia; switchgrass from North America; and reed canary grass and *Arundo* are native to Europe. Perennial grasses are often planted as whole rhizomes, and less commonly seeded. After establishment stems emerge in spring from the rhizome, biomass is rapidly accumulated during the summer and in autumn the crop undergoes senescence. Over winter the crop is left to translocate nutrients belowground and reduce the moisture content of the biomass. The crop is harvested annually in early spring, and then the cycle begins again. Perennial grasses are becoming increasingly popular as biomass crops due to their large yields, generally reaching between 10-30 t ha<sup>-1</sup> y<sup>-1</sup> (Lewandowski *et al.*, 2000). With such high yields attainable these crops are able to occupy less land than other bioenergy species (Heaton *et al.*, 2008). Generally, perennial grasses have been shown to have large environmental benefits including increased soil sequestration, improved nutrient cycling, reduced GHGs and little nitrate leaching when considering a transition from arable cropping (McLaughlin & Kszos, 2005; Voigt, 2015).

#### 1.1.4.2.3 SRF

Short Rotation Forestry (SRF) is used to describe forestry species which are cultivated on a 20 year rotation for bioenergy. They are called 'short rotation' as a 20 year cycle is shorter than rotational lengths usually used in conventional forestry which are around 100 years. There is less research available on implications of large scale cultivation of SRF, however given appropriate management practices SRF has the potential to have positive environmental impacts on the landscape (reviewed in Hardcastle *et al.*, 2006; Pérez-Cruzado *et al.*, 2012). Due to the wealth of different species that SRF encompasses there is no singular effect direction that applies to all species. A recent study showed there were increased carbon stocks under coniferous SRF compared to control arable land, whereas broadleaf SRF showed no effect and eucalyptus showed a decrease in C stock (Keith *et al.*, 2015). The authors attribute the loss in soil C under eucalyptus to the young age of the measured stands; other studies have shown increases in soil C with stand age in eucalyptus

compared to pasture lands (Pérez-Cruzado *et al.*, 2012). LUC impacts are also expected to be highly site specific (Hardcastle *et al.*, 2009).

#### 1.1.4.3 Third generation feedstocks

Briefly, third generation (3G) feedstocks consist primarily of microalgae and are used to produce biodiesel and bioethanol. Microalgae are able to produce significantly more oil for biodiesel than 1G crops on a per land unit area and due to the short life cycle they are able to be harvested several times a year compared to 1G or 2G feedstocks (Dragone *et al.*, 2010). 3G bioenergy will not be further discussed in this review.

#### 1.1.5 Bioenergy in the UK

In the UK context, the 'Energy Crop Scheme' provided by Natural England was a Government incentive program to encourage farmers to plant second generation, dedicated lignocellulosic (woody biomass comprised of cellulose and lignin) energy crops in the UK, in the belief that these crops represent a better GHG balance than arable crops and because they may be grown on land not suitable for high yielding arable cropping (DECC, 2012; CCC, 2011; Royal Society, 2008). A grant of up to 50% for the cost of the plantation was awarded for approved energy crops which included SRC trees and Miscanthus (Natural England, 2009), but despite this, uptake of these grants has been poor, not surprisingly, given that they aid crop planting rather than its harvest and profitability. Poor uptake reflects a complexity of concerns expressed by growers and these extend beyond financial considerations. The Energy Crops Scheme has now expired and subsidies are no longer offered; there are no signs yet if they plan to be replaced (Natural England, 2014). Sherrington et al. (2008; 2010) identified concerns over long-term contracts, long-term commitment of land to difficult crops and rooting systems and lower returns compared to annual crops, all contributing to poor uptake. However, they also noted that Miscanthus appeared to show higher gross margins than willow. A more effective Government approach could be initiated to provide guarantees for long-term security of income to enable the industry to flourish. Enabling the price of carbon and carbon credits to be used as a metric in such circumstances may provide the way forward, but for such a system to be feasible, a clear empirical evidence base of GHG benefits and costs of different land use would be required for the UK and this remains limited for SRC and Miscanthus, and is only now being addressed at the research level. Within Europe, the UK is underrepresented for natural biomass resources, ranking 19 out of 27 countries for forest resources (Global Forest Resources Assessment, 2005); although it has been highlighted that this still represents a significant and large source of biomass for the UK. This biomass

resource could be available through better management of private forests, providing up to 2 million tonnes of wood annually, for energy uses (Forestry Commission, 2009). Current estimates of the UK land area use for energy crops are 7100 ha for Miscanthus and 2700 ha for SRC willow (Digest of UK Energy Statistics, 2014). The use of SRC cultures have the dual benefit of producing abundant biomass for renewable energy production and the ability to sequester carbon both into the biomass and into the soil. It was found that in Western Europe alone SRC could annually sequester 24-29 t  $CO_2$  ha<sup>-1</sup> (Deckmyn *et al.*, 2004). On average, SRC willow is able to sequester carbon at a rate of 3.00 Mg ha<sup>-1</sup> y<sup>-1</sup>, with 1.71, 1.25 and 0.04 Mg C ha<sup>-1</sup> y<sup>-1</sup> allocated to aboveground biomass, belowground biomass and into the soil to 60 cm depth (Lemus & Lal, 2005). The most recent estimates show there is a potential 3.5 Mha of land available in the UK for 2G bioenergy which would not impact on the highest quality agricultural land (Lovett et al., 2014). Cultivation on this scale could provide over 60% of the demand for both heat and electricity for the UK (Wang et al., 2014a, Wang et al., 2014b). Cultivation on a smaller, more likely scale of 0.4 Mha could still provide more than 5% of the UK's heat and electricity demand by 2020 (Wang et al., 2014b).

#### 1.1.6 Indirect Land Use Change (iLUC)

One of the major constraints with the application of energy from biomass is the land required to cultivate energy crops. Land use in the UK is particularly strained with a population density of 256 people per km<sup>2</sup> versus the USA with only 34 people per km<sup>2</sup> (Office for National Statistics, 2011; United States Census Bureau, 2012).

In many places in the world, any land that is fertile and able to grow food crops is likely already under cultivation, with bioenergy crops directly displacing food or fodder crops. This direct displacement is now considered to lead to consequential indirect effects (Indirect Land-Use Change – iLUC) where other land is required to grow additional food and where this may be high carbon, such as that from deforestation and other pristine areas. The impact of these indirect effects (Searchinger *et al.*, 2008), is yet to be fully resolved and is beyond the scope of this review. However in developing sustainability criteria, the concept of iLUC factors, is being considered and it is likely that these factors may be twice the magnitude in some circumstances of the GHG costs through direct impacts assessed at a local level (Melillo *et al.*, 2009). These authors have suggested a global policy to protect forests and minimise the use of fertilisers (which may make the most significant contribution to overall GHG emissions), and would contribute towards the development of global sustainability criteria that take into account indirect effects most effectively.

Fritsche *et al.* (2010) review the options for taking account of iLUC in policy development, and in their review, the CCC concludes that either crop-specific iLUC factors are included for the growth of specific feedstock crops or limits are placed on the use of feedstocks with iLUC risks (CCC, 2011). For the present, much emphasis is placed on the growth of energy crops on less fertile degraded land, areas of ex-set aside or along field margins, thus avoiding both direct and indirect land- use changes associated with food production.

#### 1.1.7 Policies and obligations

The uncertainties surrounding the sustainability of biofuels (Scharlemann, 2008) has prompted a number of international initiatives to establish 'sustainability criteria' that propose frameworks and certification, to varying degrees, to ensure bioenergy feedstocks are planted only when no significant negative impacts are apparent. These standards, and the assumptions that underlie them, are important, since in Europe, GHG emissions reduction are central to the development of current and future targets for cultivation within the EU and also for import of feedstocks and fuel. The research described in this review will contribute to the development of appropriate criteria since many rely on modelled data and look-up tables from which to extract the GHG balance data for different cropping systems. This presents considerable uncertainty to policy development since few empirical data are available from which to verify figures used in LCA and other sustainability criteria, and these are prone to errors (Whitaker *et al.*, 2009; Rowe *et al.*, 2011).

The EU (as part of the Renewable Energy Directive; RED) is currently dedicated to increasing the amount of renewable energy used to 20% of total energy consumption by 2020 whilst simultaneously reducing GHG by 20% by 2020 (European Commission, 2009). Currently in the EU, around 3% (3.1 Mha) of EU croplands are used for bioenergy, supplying 8.7% of the total primary energy (IEA, 2010; European Commission, 2014). In the UK, crops occupy 71% of the total UK agricultural land area (DEFRA, 2014), and of this only 0.01 Mha is bioenergy crops under cultivation (UK Bioenergy Strategy, 2012). For first generation crops however, it is less clear how much contributes to bioenergy, for example Oilseed Rape (OSR) covers 25% of arable land in the UK and is used for both food and biofuel, but it is unclear how much is dedicated to each use (DEFRA, 2014); according to the Renewable Transport Fuels Obligation (RTFO) quarterly report, approximately 3% of all UK cereals were used to produce biofuels in 2009 (RTFO, 2010).

- 1) Policies that support bioenergy should deliver genuine carbon reductions;
- Support for bioenergy should make a cost effective contribution to UK carbon emissions objectives;
- Support for bioenergy should aim to maximise overall benefit and minimise cost across the economy;
- Policy makers should assess and respond to the impacts of increased deployment.

Through the RED, the UK should have 15% of all energy from renewable sources by 2020 and to reduce GHG emissions by at least 34% by 2020 and 80% by 2050, as enshrined in the Climate Change Act 2008 (emissions from a 1990s baseline). Currently bioenergy provides 3% of the total UK energy consumption, with 65% of this from electricity generation (UK Bioenergy Strategy, 2012). An analysis undertaken as part of the recent UK Bioenergy Strategy projects that sustainably sourced biomass will contribute 8-11% and 8-21% of the total energy by 2020 and 2050, respectively. One estimate predicts that in order to be able to meet the UK strategy, 350,000 ha of land will need to be under perennial crop cultivation, requiring an increase of over 2000% in area from only 15,000 ha grown in 2008 (Karp et al., 2009), which had risen to approximately 19,000 ha for 2009/2010 (see Don et al., 2012). The UK Bioenergy Strategy (2012) estimates the theoretical maximum area available to cultivate SRC willow and Miscanthus is estimated at between 0.93 - 3.63 Mha in England and Wales. It is clear from these and other studies, that in order to reach sustainability targets for 2020 and for 2050, the UK will need to supplement its own biomass with that from international imports (AEA, 2011; Figure 1.3). The equivalent amount of land required globally to supply the UK has been projected as 0.6-2.2 Mha for agricultural residues, 0.04-2.6 Mha oil crops and 3.7-17.2 Mha for woody biomass (UK Bioenergy Strategy, 2012). Within this requirement, it is critical that UKsourced biomass is grown in a sustainable manner. Whilst this review aims to focus explicitly on the UK system boundary, there will be global impacts on adoption of bioenergy crops in the UK. For instance, if we are able to optimise the production of UKsourced biomass feedstocks, this reduces the global impact on international imports, for example, in areas of the tropics where native tropical forest is being removed for bioenergy crop production. Brazil and Indonesia are responsible for 61% of global CO<sub>2</sub> emissions from LUC (Le Quere et al., 2009), although the contribution of bioenergy cropping to this figure is likely to be small. Presently, the largest UK import of biofuel is Argentinian

supplied soy-based diesel (DECC, 2012), although this may change with increased production of Brazilian ethanol in future.



Source: DECC analysis based on AEA Biomass resource model

**Figure 1.3:** Amount of energy provided from biomass supplied from domestic and international supplies (from UK Bioenergy Strategy, DECC (2012)).

'Set-aside' is land which is prevented from being cultivated on farms across Europe, and was introduced in 1992 as part of the Common Agricultural Policy (CAP). Previously this was obligatory and was a percentage of the total land a farmer had in cultivation; however, as of 2007 it became voluntary to participate and it was completely removed from the CAP in 2008. The purpose of set-aside was to prevent over-production on farms and to help avoid negative environmental impacts on the soil and on the landscape. After the set-aside initiative came to an end, as much as 20% of land in the EU was immediately re-entered into cultivation (Don *et al.*, 2012). In the UK some of the land was also redistributed into the Environmental Stewardship Scheme (ESS), another governmental incentive to protect the landscape, where famers are paid not to cultivate land. It is these lands which have the potential to be converted into bioenergy crops in the future to avoid cultivation on fertile lands, and therefore direct competition with food production. However, currently they are excluded from the ESS: if payment is received for ESS it cannot be received from the Energy Crops Scheme.

It is important that there is a reliable and rigorous means of measuring LUC when considering land conversions to bioenergy crops. At present, the IPCC (2006) present a standard method for estimating soil organic carbon (SOC) stock based on land-use and management, measured at three tiers, depending on the data collected for that area. However, there are fundamental flaws in the system, requiring further development and
implementation so that LUC decisions can be better informed for conversions like that to bioenergy crops (Smith *et al.*, 2012). An additional policy issue highlighted by Gallardo and Bond (2011) is that in the UK there is no legal mandate for conducting assessment for LUC to bioenergy crops (except for rare cases where protected lands are involved), therefore highlighting that more could be done at a governmental level to help protect the environment.

# 1.2 The Effects of Land Use Change on Soil Carbon and GHG Emissions

## 1.2.1 The importance of the soil for GHG mitigation in land use transitions to bioenergy

The soil is extremely important in the global carbon cycle as it holds 1500 Pg C (equivalent to 1500 billion tonnes), which is more than the vegetation and atmosphere are able to hold together (Fig 1.4), representing the largest terrestrial stock of carbon. Lal (2004) highlights the importance of SOC for its on-site and off-site values to our landscape and to human well-being (Table 1.2). SOC is considered as any organic input from plant, animal or microbial matter which is at any stage of decomposition. The amount of carbon fixed into the soil is the balance between the rate of inputs, in this case from litter for example, and the mineralization of the existing soil carbon stores (Jenkinson, 1988; Post & Kwon, 2000). The global carbon pool is made up of 5 main pools as shown in Figure 1.5; these all play a part in CO<sub>2</sub> efflux from the soil but only 'SOM-derived CO<sub>2</sub>' significantly contributes to changes in atmospheric CO<sub>2</sub> concentration (Kuzyakov, 2006). It is important to be able to measure the different sources of CO<sub>2</sub> efflux from each of the different pools, as this allows us to evaluate whether the soil is acting as source or a sink for CO<sub>2</sub>; this can be found in a comprehensive review of partitioning methods by Kuzyakov (2006). The soil is not an unlimited sink, and has a limited carbon storage capacity which is determined by vegetation type, climate, nutrient content, hydrology and topography (Gupta & Rao, 1994; Nair et al., 2009). Anthropogenic activities such as LUC and land management have extremely large impacts on the soil carbon pool, resulting in increased emission of CO<sub>2</sub> due to decomposition of SOM or increased soil respiration (Schlesinger, 2000).

Soil functioning underpins ecosystem services and is important to consider when discussing the effects of LUC, although few studies have considered processes such as predation in bioenergy systems. In a comprehensive review by Baum *et al.* (2009), it was found that land conversions to SRC willow and poplar can have both positive effects (such as increased abundance of earthworms and positive effects on nutrient cycling), as well as negative effects (such as dominance of arbuscular mycorrhizal host plants). Rowe *et al.* (2013) have also considered ecosystem functioning alongside biodiversity and report significant increases in predator abundance and altered decomposition rates in SRC willow compared to arable crops.

Table 1.2: Onsite and offsite benefits of SOC on the landscape (From Lal, 2004)

On-site benefits of SOC	Off-site benefits of SOC		
Source and sink of principle plant nutrients	Reduced sediment loads in streams and rivers		
Source of charge density and responsible for ion exchange	Filters pollutants from agricultural chemicals		
Able to absorb water at low moisture potential thereby increasing plant available water capacity	Aids biodegradation of contaminants		
Promotes soil aggregation which improves soil tilth	Buffers GHG emissions from soils into the atmosphere		
Caused high water infiltration capacity & low losses due to surface runoff			
Substrate for soil microbial communities therefore increase biodiversity			
Provides strength to soil aggregates leading to a reduction in erosion susceptibility			
Encourages high nutrient and water use due to reduced losses by drainage, evaporation and volatilization			
Buffers against pH fluctuations due to addition of agricultural chemicals			
Moderates soil temperature through effect on soil colour and albedo			



**Figure 1.4:** The global carbon cycle showing where carbon can be stored in pools or where it is released as fluxes (Adapted from <a href="http://globecarboncycle.unh.edu/diagram.shtml">http://globecarboncycle.unh.edu/diagram.shtml</a>)

## 1.2.2 Initial conversion considerations

The initial conversion process from one land-use to another usually results in a release of emissions due to the removal of the current crop cover (manually, with fire, or by chemicals), preparation of the land for planting (chemical and mechanical), and then the crop establishment phase (chemical). *Miscanthus* propagation in particular is known to be energy and GHG-intensive during the first stage of crop establishment (Styles & Jones, 2007). In a conversion from arable to SRC poplar, Arevalo *et al.* (2011) found initially a release of carbon occurred, but the soil had become a net sink by year two. The point at which the newly established land-use is equal to that of the previous land-use is sometimes referred to as the 'break-even point' or 'carbon neutrality' – in one study for arable to SRC



**Figure 1.5:** Soruces of biogenic CO<sub>2</sub> efflux from soil in order of turnover rates and mean residence times in the soil (Adapted from Kuzyakov, 2006).

poplar it was found to be 4 years, similar to other studies for this type of conversion (Price *et al.*, 2009). The first year of cultivation has been highlighted as a particularly sensitive year with respect to carbon balance. Data from a clustered eddy covariance approach shows how large carbon imbalances can be invoked, but also how a conversion to bioenergy can help stabilize and negate emissions in the long term (Zenone *et al.*, 2011). Abbasi and Abbasi (2010) are careful to point out that while bioenergy crops are considered 'carbon neutral', they are not necessarily 'nutrient neutral' as each crop will exert varying amounts of pressure on the nutrients of the soil on which they are cultivated.

A second study looking at the effects of the first year after establishment found that a conversion from arable land to SRC willow and poplar incurred a GHG debt of 7.4 and 11.6 Mg ha<sup>-1</sup> y<sup>-1</sup>, showing that while CO<sub>2</sub> emissions were 29-42% less than arable, the amount of N<sub>2</sub>O emissions and nitrate leaching increased in the SRC plots (Nikiema *et al.*, 2012). This highlights the importance of taking into account all the effects of a conversion, showing how critical the first year can be in determining GHG savings; in the long term however these debts should be repaid and the overall environmental gain should be greater than if no conversion was to occur. Styles and Jones (2007) found that while the conversion from de-stocked grassland to bioenergy crops would create a small increase in

GHG emissions during cultivation, these would be greatly offset by their displacement of traditional fossil fuel use, a full LCA showing almost a 90% decrease in GHG emissions.

The importance of initial site conditions have also been highlighted when considering LUC to bioenergy. Palmer *et al.* (2014) showed that a conversion from grassland to SRC willow/poplar resulted in larger emissions at establishment when grassland had a higher initial C stock. The site with higher C stocks had been grassland for over 20 years, whereas the other grassland had been under row crop cultivation 5 years prior to SRC establishment. The resultant GHG emissions were 43.2 and 33.0 t CO<sub>2</sub>-eq ha<sup>-1</sup> in the long-idol site compared to 4.8 and -1.5 t CO<sub>2</sub>-eq ha<sup>-1</sup> in the recently disturbed site, for SRC poplar and willow respectively.

The initial land-use, management and fertiliser regime are the main factors in determining whether a conversion to bioenergy crops will be beneficial or detrimental, and also the conversion crop type (e.g. Tolbert et al., 2002; Morris et al., 2010). For liquid transport fuels, 1G crops OSR and wheat are primarily cultivated (Gallardo & Bond, 2011), and which are annual row crops. Most annual cropping systems are associated with lower SOC contents than perennial crops and therefore these losses incurred (mainly by harvesting, ground preparation practices and removal of residues) need to be compensated by other management practices such as fertiliser of winter cover crops (discussed later in section 3.2). In a comparison between the effects of growing OSR versus Miscanthus and SRC willow, it was shown that OSR not only has detrimental effects on soil quality with decreased amounts of SOC during occupation but also had the highest acidification and eutrophication potentials (Brandao et al., 2011). This study highlights the need to understand the full array of consequences of land-use and also how differing management strategies impact on the life cycle of various crops. Figure 1.6 shows that fertiliser is the main contributor to the GHG emissions of the life cycle for OSR due to associated field CO<sub>2</sub> and N<sub>2</sub>O emission, as well as the emissions associated with manufacturing the fertiliser. Miscanthus and willow SRC do not have fertiliser-associated emissions as they do not undergo this management type, and the sequestration via the soil offsets any other life cycle emissions. There are no associated life cycle emissions for the collection of forest residues for use as bioenergy.



**Figure 1.6:** GHG emissions of different land uses broken down into contributing factors from different parts of the life cycle, expressed per reference unit (ha<sup>-1</sup> yr<sup>-1</sup>) (from Brandao *et al.*, 2011).

When discussing the effect of LUC on soil carbon stocks, it seems most appropriate to start with the classic review by Gou and Gifford (2002). Gou and Gifford (2002) conducted a meta-analysis to quantify the effect of LUCs on soil carbon stocks and their results are summarised in Table 1.3. It is clear from this analysis that a conversion to croplands is detrimental and any conversion out of a cropland system causes an increase in soil carbon stocks – this is likely due to vast differences in management practices. The main takehome message from this paper, and a wealth of others in the published literature, is generally that a conversion away from the native land-use tends to have a negative effect on carbon stocks (e.g., Fargione *et al.*, 2008). It is not to say, however, that these changes are permanent, as these time-series are limited. One benefit of this review is that it considered soil depth in some detail and given the literature on tillage (see Section 1.2.3.1), it is likely that this may impact the potential for bioenergy cropping systems to lead to improved soil carbon.

**Table 1.3:** Summary of the findings from Gou and Gifford (2002) who conducted a meta-analysis to assess the effects of LUC on soil carbon stocks across all regions of the world. 'Native forest' is forest before clearing for other land use; 'Pasture' is for grazing (including natural grasslands); 'Crop' is land cultivated for food and fibre; 'Secondary forest' is forest that has regenerated on abandoned land after being used for other purposes; and 'Plantation' is forests established through human activity.

Original Land use	Transition to:	Effect on soil carbon stocks
Pasture	Plantation	-10%
Native forest	Plantation	-13%
Native forest	Crop	-42%
Pasture	Сгор	-59%
Native forest	Pasture	+8%
Сгор	Plantation	+18%
Сгор	Pasture	+19%
Сгор	Secondary forest	+53%

The 'carbon debt' is a measure of the extent to which the use of bioenergy is able to reduce carbon emissions relative to a fossil fuel reference system. This is most often reported as an amount of years which will be required for the land conversion to be able to 'pay back' the carbon debt to the land. For example, in a study by Fargione *et al.* (2008), it was shown that a conversion from US grassland to corn for bioethanol would incur a carbon debt of 93 years, and from abandoned cropland to corn, a 48 year carbon debt. This again presents another example where a conversion from a native ecosystem leads to more negative environmental impacts than a conversion from an already anthropogenically altered ecosystem. A more worrying estimate was one of a conversion to corn, again in the USA, presenting a 167 year payback time when indirect effects on land-use were also considered (Searchinger *et al.*, 2008). Recently, Mello *et al.* (2014) showed the payback time for soil C was only 8 years for a conversion from native vegetation to sugarcane ethanol, though

these estimates do not consider the GHG and ecosystem C changes. Failure of studies to take into account the effects of LUCs (both direct and indirect) will give an inaccurate picture of the effects of a conversion to bioenergy crops and needs to be incorporated into all studies considering land conversions (Searchinger *et al.*, 2008; Fargione *et al.*, 2008).

Soil methane (CH<sub>4</sub>) fluxes are often not discussed in many papers due to the fluxes being very small in relation to CO<sub>2</sub>, but they are still an important component in the GHG balance of bioenergy crops. The soil acts as an important biological sink for CH<sub>4</sub>, fixed by oxidation by methanotrophic bacteria in aerobic soils; however in anaerobic environments methanogenic bacteria dominate, resulting in an emission of CH<sub>4</sub> (see refs within Kern *et al.*, 2012). In a comparison between annual and perennial bioenergy crops, it was found that annual crops consumed more CH<sub>4</sub>, 6.1  $\mu$ g CH<sub>4</sub> m<sup>-2</sup> ha<sup>-1</sup> versus 4.3  $\mu$ g CH<sub>4</sub> m<sup>-2</sup> ha<sup>-1</sup> for perennial bioenergy crops; it appears that soil water content is the main reason this difference is seen (Kern *et al.*, 2012). Thus far it has been found that perennial crops have a far greater environmental advantage over first generation annual crops, however in the case of CH<sub>4</sub> emissions annual crops seem to come up ahead in this case. The overall GHG balance can be supplemented by the fact that CH<sub>4</sub> is taken up by the soil; Kern *et al.* (2012) predicted that 3-4% of CO<sub>2</sub>-eq from soil borne N<sub>2</sub>O emissions can be compensated by the consumption of methane in this experiment.

#### 1.2.3 Specific crop transitions of relevance to the UK

## 1.2.3.1 Transition from arable to bioenergy crops

Several studies have investigated the effects of a conversion from traditional annual, arable to perennial bioenergy crops. The general consensus is that this conversion to SRC and *Miscanthus* results in increased SOC and soil quality (e.g. Tolbert *et al.*, 2002; Anderson-Teixeira *et al.*, 2009). However, care should be taken in assessing the results since many rely on modelled and not measured data, and when measured studies are undertaken, often only the top 30 cm soil profile is investigated. In an analysis of the literature, Harrison *et al.* (2011) have concluded that this can lead to erroneous conclusions, and caution against shallow soil sampling in studies to quantify soil C pools and changes in soil C over time. New data are now emerging such as Gauder *et al.* (2012), who have measured GHG flux across willow SRC, *Miscanthus* and corn at two levels of fertiliser, and show fluxes of these gases to be greatest from fertilised corn, but no data as yet are available for SOC. It's likely that these studies over the long-term will provide more conclusive data to address this question.

Future research should be focused on long-term measurement campaigns with field-grown trees and grasses, in controlled replicated studies to ensure the evidence base to assess changes in soil carbon is firm and UK-specific.

Future research should target resources for long-term soil-based studies that include the whole soil profile down to 1 m.

The UK Bioenergy Strategy (2012) found that the energy balance of dedicated biomass crops can have lower direct carbon impacts between 0.5 - 6.1 t CO<sub>2</sub>-eq ha<sup>-1</sup> y<sup>-1</sup>, than food production 3.4 - 11 t CO<sub>2</sub>-eq ha<sup>-1</sup> y<sup>-1</sup>. The detrimental impacts of arable lands have been highlighted in several studies; one study found that if 50% of the area in the EU which is currently planted with silage corn is replaced by permanent grass or temporary grass, GHG emissions would be reduced by 1.3 Mt CO<sub>2</sub>-eq ha<sup>-1</sup> y<sup>-1</sup> and 0.9 Mt CO<sub>2</sub>-eq ha<sup>-1</sup> y<sup>-1</sup> (Mt =  $10^6$  tonnes; Henriksen *et al.*, 2011). The complete conversion of arable land to permanent grass is estimated to increase soil carbon by 0.5 t ha<sup>-1</sup> y<sup>-1</sup> (IPCC, 2001; Conant *et al.*, 2001). In terms of SOM, annual crops to SRC results in an average SOM increase of 1 t C ha<sup>-1</sup> y<sup>-1</sup>; yield increases due to every additional tonne in SOM are approximately 5% (see refs within Nijsen *et al.*, 2012). These carbon gains from conversion are likely mainly due to the change in management practice, particularly no-tillage, thereby highlighting the vast impact management can have on carbon balance (See section 3.2).

A comparison between fields under two different land uses (space-for-time comparisons) is one method to investigate the experimental effects. In one study where arable OSR and wheat were compared to SRC willow and *Miscanthus*, it was shown that the main difference was the N<sub>2</sub>O fluxes were significantly smaller for bioenergy crops than arable crops (Drewer *et al.*, 2012), but this effect was reversed when fertiliser was added to the perennial bioenergy crops in both *Miscanthus* and SRC; this suggested that the GHG mitigation potential of crop transitions from arable to perennial crops is highly dependent on fertiliser regime. In a review by Anderson-Teixeira *et al.* (2009), it was shown that upon conversion to perennial species the average SOC accumulation rate was <1 Mg C ha<sup>-1</sup> y<sup>-1</sup> in the top 30 cm of soil. Similar data were reported in a review by Don *et al.*, (2012), with 0.44 Mg soil C ha<sup>-1</sup> y<sup>-1</sup> for poplar and willow and 0.66 Mg soil C ha<sup>-1</sup> y<sup>-1</sup> for *Miscanthus*. For switchgrass, up to 1.1 Mg soil C ha<sup>-1</sup> y<sup>-1</sup> was reported (Monti *et al.*, 2012). These changes are likely attributable to a change in management practice and changes to soil properties by the crop cultivated; for example a switch to a "no-tillage regime" results in less exposure of SOM and therefore decreased decomposition, but this may be complicated at depth in the profile and this is often not fully investigated in approximately 50% of the studies reported by Anderson-Teixeira *et al.* (2009).

The consensus of evidence available suggests that transitions from annual arable to perennial grass and SRC crops leads to improved SOC, but the overall GHG balance to farm-gate may be positive or negative and largely driven by fertiliser input and consequent  $N_2O$  emissions. Accumulation of SOC is in the range 0.44-1.1 Mg C ha<sup>-1</sup> y<sup>-1</sup>.

The cultivation of perennial crops helps to stabilise the soil after a conversion by allowing the soil to accumulate into macro-aggregates, thereby allowing the sequestration of more organic carbon (Grandy & Robertson, 2007). Perennial crops are also able to provide benefits through their deeper and more extensive rooting system, providing both physical stability and chemical stability through the presence of mycorrhizal fungi in symbiosis with roots. Godbold *et al.* (2006) illustrated in a Free Air CO<sub>2</sub> Enrichment (FACE) carbon labelling experiment using poplar SRC that movement of carbon into the SOM pool was predominantly via the mycorrhizal external mycelium, exceeding the input from leaf litter and fine root turnover.

The benefits of a conversion to SRC cultivars for energy have been quantified in other studies as an economic value which represents the savings that will be made as a result of the LUC; for example Updegradd *et al.* (2004) found a saving due to carbon sequestration of \$13-15 ha<sup>-1</sup> over a 5-year rotation period for SRC poplar when used as a bioenergy crop. More recently Valentine *et al.* (2011) have extended this and placed a value of \$56-218 bn per annum for the carbon emissions savings globally, given the price of carbon at \$40 per tonne.

## 1.2.3.2 Transition from degraded, marginal and abandoned land to bioenergy crops

It has been suggested that the indirect impacts of increased bioenergy crop deployment globally, could be largely mitigated if energy crops are planted on degraded and abandoned land that does not provide any provisioning ecosystem services (Gallagher, 2008). The problem with this approach is two-fold. Firstly, such areas may provide significant 'other' ecosystem services related to biodiversity and amenity that may be enhanced or lost with transition to bioenergy crops. Secondly, considerably lower crop yields are likely from such land. The total global area of degraded land has been estimated in several recent studies, with varying results. Nijsen *et al.* (2012) gave a figure of 1836 Mha, with less than 6% of this in the EU (Nijsen *et al.*, 2012). A study based on satellite and historic data

suggested an abandoned agricultural land resource between 385-472 million hectares (Campbell *et al.*, 2008), with a mean bioenergy crop yield of 4.3 t ha<sup>-1</sup> y<sup>-1</sup>. Cai *et al.* (2010) estimated marginal agricultural land at 320-702 Mha (increasing to 1411 Mha if grassland savannah and shrubland with marginal productivity were included), with a suggestion that Africa and Brazil together have more than half of the total marginal land available for bioenergy crop production. This further emphasises the likely requirement of Europe to seek imported feedstock resources in future, where sustainability standards are difficult to control. Globally, the main causes of soil degradation are deforestation (29.4%), overgrazing (34.5%), and intensive agriculture (28.1%) (Oldeman, 1994; Lemus & Lal, 2005). Growing any crop on marginal, degraded or poor quality lands will result in lower yields due to lower levels of water and nutrients. Simulations performed by Nijsen et al. (2012) showed that woody crops (SRC willow and poplar) and grass species (switch grass and *Miscanthus*) yielded 8.9 and 6.8 odt  $ha^{-1} y^{-1}$  on degraded lands respectively; Campbell et al. (2008) suggest 4.3 tonnes ha<sup>-1</sup> y<sup>-1</sup>. These projected yields are lower when compared to the latest available varieties in the UK at 6.71 - 12.3 odt ha<sup>-1</sup> y<sup>-1</sup> and 12-16 odt ha<sup>-1</sup> y<sup>-1</sup> for SRC willow and Miscanthus respectively (Macalpine et al., 2011; Natural England, 2007). This suggests that breeding targets in future should focus on breeding for optimum, rather than maximum, yields with limited inputs of fertiliser and water (Sims, et al., 2006; Karp & Shield, 2008).

The type and the severity of the degradation will determine the amount of yield losses, varying between 4.6 - 88% yield reductions (Nijsen *et al.*, 2012). Such losses however may not be permanent due to the positive effects of planting SRC and *Miscanthus* on the land. These crops are able to significantly increase the productivity of the landscape by increasing soil stability through rooting structures, increased SOM through residue/litter fall and increased biodiversity (e.g. Rowe *et al.*, 2010). SRC willow and poplar are known for their ability to grow on poor quality lands and for their phytoremediation capacity, making them well suited to cultivation on marginal and degraded lands (e.g. Doty, 2008; Baum *et al.*, 2009).

Several different estimations have been given for the potential of growing energy crops on degraded lands (Table 1.4); on average, together they predict a potential between  $4.2 - 24.2 \text{ EJ Mha}^{-1} \text{ y}^{-1}$ , irrespective of yield and therefore variable depending on crop and level of degradation.

Area of degraded land (Mha)	Bioenergy Potential (EJ y <sup>-1</sup> )	Yield (Mg ha <sup>-1</sup> y <sup>-1</sup> )	Reference
430 - 580	8 - 110	1 – 10	Hoogwijk <i>et al.</i> (2003)
500	45	4.5	Tilman <i>et al.</i> (2006)
550	43	-	Van Vuuren <i>et al.</i> (2009)
1836	151 - 193	6.8 – 8.9	Nijsen <i>et al.</i> (2012)

**Table 1.4:** Global energy potential for the production of bioenergy on degraded lands.

In the context of this review with focus on the UK, agricultural land classes (ALCs) may be used to identify areas of low productivity and these have been linked to other land constraints including national parks, pristine high-carbon soils and land with high biodiversity value in Sites of Special Scientific Interest (SSSI). Using this constraint mapping approach, estimates of biomass supply have been made for both SRC and *Miscanthus*. Lovett *et al.* (2014) reported that between 4-28% low quality agricultural land would be required to supply 3.5 Mha Miscanthus, with a total production of 4.56 Modt from England that would enable 2.4% of total energy demand to be met - just from plantings in very poor agricultural land. Similarly for SRC in England, Aylott et al. (2010) showed that 7.5 Modt was realistically available for England, requiring growth on 800,000 ha of poor-quality land, supplying 4% of current electricity demand. Research is currently in progress to identify how these two crop types will act together, since in general, SRC yields better in the westerly areas of the UK, whilst Miscanthus shows preference for the south and east (Bauen et al., 2010); this has also been confirmed by more recent work (Tallis et al., 2013) and within the ETI (BVCM research project; Richter et al., data unpublished; Taylor et al., data unpublished). More recent analyses show planting of Miscanthus and SRC in the south-west and north-west England, respectively, have multiple yield and ES benefits (Milner et al., 2015)

Others have also investigated mass scale afforestation on degraded or poor-quality land with SRC cultures; for example in a modelling study in Canada, afforestation with SRC willow over 2.12 Mha of marginal land in Saskatchewan showed sequestration rates of 5.7-7.5 Mg C ha<sup>-1</sup> y<sup>-1</sup> over a 44 year simulation (Amichev *et al.*, 2012). The importance of taking into account the quality of the land can be seen when comparing a grassland to degraded grassland; for example, conversion from a grassland to corn caused an emission of 79 gCO<sub>2</sub>/MJ whereas a conversion from degraded grassland sequestered 11 gCO<sub>2</sub>/MJ (Lange, 2011). Beringer *et al.* (2011) warns that if biomass cultivation is not restricted to abandoned or marginal lands, the spatial expansion will put already vulnerable ecosystems at further risk.

### 1.2.3.3 Transition from grassland to bioenergy crops

Improved grasslands are important sources of terrestrial carbon storage, holding the second largest store after bogs, with approximately 274 million tonnes of carbon (Ostle et al., 2009). It has been shown that a conversion of arable to permanent grass will increase soil carbon by 0.5 t ha<sup>-1</sup> y<sup>-1</sup> (IPCC, 2001; Conant *et al.*, 2001). Ostle *et al.* (2009) found that the single largest contributor to soil carbon losses due to LUC was the conversion from improved grassland to arable crops, between 1990 and 2000 in the UK. In the UK context, conversion of semi-permanent, permanent or managed grassland to bioenergy cropping systems probably represents one of the most controversial land-use transitions as grassland is a significant part of the UK landscape (4-5 Mha, DEFRA, 2007) and because management of different grasslands can vary widely in the UK, particularly with respect to fertiliser input and grazing. This can have a dramatic impact on the GHG benefit or cost of transition to either first, or second generation bioenergy cropping systems. St Clair et al. (2008) and later refined by Hillier et al. (2009) provides the most comprehensive UKcentric data set, used in the recent CCC review (2009). Here the results are clear (Figure 1.7): transition from grassland to 1G crops results in a net loss of carbon from the system whilst transition to 2G crop provides a net benefit. However, these data represent modelled outputs only, with limited validation. Consideration of limited, but increasing, field data sets from long-term studies provides no clear picture on the likely consequences of grassland conversion.



**Figure 1.7:** Taken from Hillier *et al.* (2009). Net annual gas (t CE ha<sup>-1</sup>, CE = carbon equivalents) balance for all replacement scenarios, arable, grassland and forest/semi-natural, with bioenergy in the UK. Black – soil emissions, grey – incorporating before and after management emissions; light grey – incorporating fossil fuel substitutions. Error bars represent  $\pm 2$  SD.

In one report, long-term belowground storage of carbon by bioenergy crops has been shown to be equal to or greater than that of grasslands due to the long rotation and extensive fine roots of SRC crops and the rhizome mass of *Miscanthus* (see refs within Style & Jones, 2007). However, recent work by Zimmermann *et al.* (2012) in a comparison of *Miscanthus* and tilled grassland at 16 sites following conversion in 2006 showed no significant improvement in SOC when sampled after 2-3 years post-conversion. However, for a switchgrass modelling study that considered 12 simulation scenarios, a net C sequestration was observed in 11 out of 12 simulations, as determined by amount of nitrogen fertilisation and initial soil carbon stocks (Garten, 2012); this makes generalisations difficult. Again, these are modelled data with few empirical studies available. Monti *et al.* (2011) confirmed both positive and negative changes in soil C for switchgrass, but in a modelling exercise by Anderson-Teixeira *et al.* (2009), grassland conversion to either *Miscanthus* or switchgrass resulted in a net increase in SOC, with the largest effects seen in switchgrass.

The GHG benefit of conversion from grassland to second generation cropping systems remains uncertain due to limited empirical data and mis-match between modelling and measurement reports.

Conversion of grassland to 1G crops appears to provide a more robust dataset. Conversion to soybean from unmanaged grassland and arable lands have been compared and it was shown that there are greater benefits from converting from arable lands as there is a lower soil quality and lower initial carbon stocks (Bhardwaj *et al.*, 2011). In Europe, the conversion from set-aside grassland and improved grassland to OSR has been shown to sequester 0.6–3.3, and 2.2–10.6 t CO<sub>2</sub>-eq ha<sup>-1</sup> y<sup>-1</sup>, respectively (Flynn *et al.*, 2012). Smeets *et al.* (2009) in a modelling study reported reduced GHG balance of sugar beet, OSR and wheat relative to a grassland control; although most of the study was considering N<sub>2</sub>O fluxes rather than soil sequestration. Lange (2011), considered transitions from both grassland and degraded grassland and for wheat found that emissions savings associated with LUC were both positive and negative depending on grassland type, with more productive grassland conversion found to have a smaller change in soil carbon. Hillier *et al.* (2009) show that all emissions were increased following grassland conversion to either OSR or wheat.

There is likely to be a negative impact on GHG balance of transition from grassland to first generation bioenergy cropping systems.

Grasslands could also be considered as a source of energy themselves; for example Tilman *et al.* (2006) suggested that low-input, high-diversity prairie systems involving mixtures of native grassland perennials can provide more usable energy, greater environmental benefits, and less agrichemical pollution per hectare than corn-ethanol or soybean biodiesel. However, in recent experimental work, this notion has been questioned, since the low biomass yields in such biodiverse systems (~4 t ha<sup>-1</sup> y<sup>-1</sup>) do not compare favourably with those of switchgrass and *Miscanthus* (Tilman *et al.*, 2006).

Biodiverse grasslands are unlikely to provide significant yields enabling them to compete commercially with bred varieties of perennial bioenergy grasses and should not be considered further as sources of biomass feedstock.

## 1.2.4 Management practices and their relevance to bioenergy

Management practices are important when assessing GHG and soil carbon impacts on the land regardless of the land-use type. The way the land is managed is one of the key contributors to the GHG balance and soil carbon; this can be done in such a way to reduce emissions, but more frequently is referred to in the literature as a means by which we are causing an excess of emissions, such as modern agriculture.

Several strategies are now being employed to encourage carbon sequestration and to minimise disturbance that may cause a large release of emissions into the atmosphere. These include, but are not limited to, retention of residues on the soil, decreased/optimised fertiliser application, reduced or no-tillage and reduced/optimized irrigation. It should be kept in mind that current and past breeding for yield may have mitigated against soil stability and improved GHG balance. Future breeding and management are much more likely to be focussed on 'efficient crops' that are managed for optimum rather than maximum yields (Kell, 2011)

Work undertaken by the IPCC investigated the potential GHG mitigation strategies available to us and how we can manipulate current agricultural practises to aid carbon mitigation. Table 1.5 shows the GHG savings that could be made if certain land management strategies were improved from their current state, including the use of bioenergy crops as a whole. Davis *et al.* (2013) recently coined the term 'management swing potential' which describes how management can be used to alter the GHG mitigation potential of LUC to bioenergy. She outlines a number of case studies which highlight the ways in which bioenergy crops can be managed to ensure minimal environmental impacts as a result of LUC to bioenergy, such as by rotation length, use of fertiliser to increase C fixation and timing of harvesting (Davis *et al.*, 2013). Data on all management practices employed in any land use system must be collected therefore to properly evaluate the potential GHG benefits that can be derived from LUC to bioenergy.

To enable the soil to be used as a sink for anthropogenic sources of excess  $CO_2$  from the atmosphere, the amount of SOC needs to be increased. This can be done by increasing the amount of SOM which enters the soil and this can be achieved by changing the way crops are managed; Lal *et al.* (1999) suggested these need be as simple as conservation tillage, irrigation, reducing/eliminating fallow and retention of crop residues.

The above mentioned management strategies will be discussed in this report in the context of bioenergy crops, whilst other land-use and management strategies for increasing carbon sequestration exist, they will not be discussed due to lack of relevance to bioenergy cropping systems. The use of these management strategies will vary largely based on the crop being assessed and may not be relevant to all crop types. 

 Table 1.5: IPCC mean estimate of GHG-mitigation potential of management strategies (From Smith *et al.*, 2007).

Mitigation Strategy	Mitigation potential	Climatic Zone
	(t CO <sub>2</sub> -eq ha <sup>-1</sup> y <sup>-1</sup> )	
Improved water management	1.14	All climatic zones
Improved crop management	0.39 – 0.98	Dry and moist
Improved tillage and residue management	0.53 – 0.72	Cool-moist & warm moist
Bioenergy crops	0.53 – 0.72	Cool-moist & warm moist
Improved nutrient management	0.33 – 0.62	Dry and moist
Improved tillage and residue management	0.17 – 0.35	Cool-dry & warm-dry
Bioenergy crops	0.17 – 0.35	Cool-dry & warm-dry

## 1.2.4.1 Tillage as a management strategy

Tillage is defined as the practice where soil is prepared for planting by mechanised disturbance using digging and overturning. Several types of tillage exist, namely conventional tillage, conservation or reduced tillage and no-tillage, and these categories refer to the amount of soil disturbance and amount of residue that is buried. Once a crop has been harvested there will be residue left on the surface. Conventional tillage will cause almost all residues to be turned and mixed in with the soil, with less than 10-15% residue left on the soil surface. Reduced or conservation tillage will leave between 15-30% of residues on the soil surface and in a no-tillage system, the ground is not tilled but will only be disturbed during planting.

Within the literature there is general agreement that reduced tillage provides less disturbance and therefore will be a more suitable means of sequestering carbon within the soil compared with conventional tillage (Paustian *et al.*, 1997; van Groenigen *et al.*, 2011; Chen *et al.*, 2009). Decreased disturbance results in decreased aeration, decreased soil erosion, water and heat/thaw cycles, minimised oxidation of organic matter and encourages better aggregation, all contributing to the stabilization of soil organic matter (SOM;

References within Lennon & Nater, 2006). The IPPC guidelines for GHG accounting inventories suggest a multiplication factor of 1.0 for a conversion from conventional tillage to reduced tillage (IPCC, 1997), which translates to an approximate SOC increase of 10% (West & Post, 2002). Paustian et al. (1997) showed in a comparison of 39 paired sites (conventional tillage vs no-tillage) that soil carbon was 8% (285 g m<sup>-2</sup>) higher when subjected to a no-tillage management regime. It should be noted that the compared studies were not necessarily looking at the GHG balance and soil sequestration potential of the two management regimes, so are likely an under-estimate of the effect of reduced tillage on carbon storage. In a global analysis of the effect of tillage on soil C sequestration, West and Post (2002) found that a switch from conventional tillage to no tillage can sequester 57  $\pm$  14 g C m<sup>-2</sup> yr<sup>-1</sup> and that the majority of the SOC change seen occurs in the first 10-15 years following the switch over. Reduced tillage encourages SOM accumulation by reducing disturbance of the soil and limiting soil and residue contact (Carter, 1992). Reduced tillage shows an increase in bulk density in the upper soil layers (~0-30 cm; Van Groenigen et al., 2011; Dolan et al., 2006). A recent meta-analysis has shown that no tillage reduced surface runoff by 21.9% and 27.2% compared to reduced and conventional tillage, respectively (Sun et al., 2015).

Whilst many have found reduced or no-tillage treatments do sequester more carbon than their conventional tillage counter parts (Van Groenigen et al., 2011; West & Post, 2002; Ogle et al., 2005), there remain inconsistencies. It appears that the amount of sequestration may often be equivalent, but the distribution of stored carbon may differ along the entire soil profile (Dolan et al., 2006; Blanco-Canqui & Lal, 2008; Angers et al., 1997; Vanden Bygaart et al., 2002). Dolan et al. (2006) and others showed that the amount of SOC and soil nitrogen was significantly higher in the no-tillage treatments compared to conventional tillage for the top 0-15 cm of soil. They found 15-20 cm to be a transition zone where there was no significant difference in SOC or soil N, but then for the deep parts of the profile, 20-45 cm, conventional tillage showed a higher amount of SOC and soil N. When summating for the entire profile (0-45 cm), there was no significant difference in SOC between tillage treatments, but soil N was significantly higher in no-tillage treatment (Dolan et al., 2006). This shows that while reduced tillage is often favoured for its environmental impacts, it may be less effective than thought as a management tool for soil carbon sequestration, with effectiveness determined by soil type, nitrogen treatments and other soil characteristics such as fungal community (Six et al., 2004). In a review of our current knowledge on tillage and carbon sequestration, Baker et al. (2007) reported that the majority of studies have only sampled soil to a depth of 30 cm, this perhaps being the reason why there is widespread preference for reduced/no-tillage systems. However, studies which sampled to lower depths found no significant difference in carbon storage between conventional and reduced/no-tillage systems and in many cases found that more C was stored beneath conventional systems (Baker *et al.*, 2007; Blanco-Canqui & Lal, 2008). It should also be highlighted that many studies on tillage are taken on small experimental plots which minimise interference of other variables which is important for establish effects, but is not necessarily how these management strategies will be put into practice on a commercial scale (Blanco-Canqui & Lal, 2007).

Dolan *et al.* (2006) found that the profile effect documented for SOC and soil N was the same for bulk density (higher in conventional tillage surface soils but lower below 30 cm compared to no-tillage) and for the  $\delta$ 13C signature (less negative in surface soils for conventional tillage and then more negative below 30 cm compared to no-tillage). This also appears consistent with other findings (Blanco-Canqui & Lal, 2008), and it is recommended that future research on bioenergy LUCs should consider the whole soil profile in some detail. In a meta-analysis by Angers and Eriksen-Hamel (2007), it was concluded that effects of no-tillage on soil organic C content above and below 35 cm differed, and this was an extensive study using more than 25 pieces of original research, varying from 5 to 30 years duration. The authors were unable to identify why they found a significant stock change in SOC between no-tillage and tillage with increased SOC above 35 cm, with a relative accumulation of SOC at depth in the tillage regime. This was a general conclusion not limited by soil type. It is important to understand this management change in order to achieve effective soil carbon sequestration.

In addition to soil profile depth, several studies have highlighted the importance of sampling strategy to ensure a full picture of what is occurring is captured (Dolan *et al.*, 2006; Blanco-Canqui & Lal, 2008). This includes one of the largest and most highly cited reviews on the effects of tillage on C sequestration, which drew many of its conclusions from studies which only sampled the top 30 cm of soil (West & Post, 2002). The effect of tillage on SOC and soil N are site- and soil-specific, leading many studies to have highly variable results (Blanco-Canqui & Lal, 2008; Chen *et al.*, 2009; Lennon & Nater, 2006; Dolan *et al.*, 2006). Kaiser *et al.* (2014) recently showed a depth-specific response to tillage regimes, with a larger labile C and N stocks under reduced and no-tillage compared to conventional tillage; which showed to have a higher labile C and N pool for sub-surface soils.

Bioenergy cropping systems encompass both annual and perennial crops, with the assumption that no-tillage operations associated with perennial crops are likely to lead to enhanced SOM and soil carbon. In general, in the context of bioenergy crops this type of land preparation would be expected to occur more often for 1G crops (annual crops such as wheat and sugar beet) than for 2G crops (lignocellulosic such as willow and *Miscanthus*). However, the long term effects of the tillage may be offset because 2G crops will be in the ground for at least a 10 year cycle. From the above literature, it can be concluded that there is still a lively debate occurring since the effect of a change from conventional tillage to reduced/no-tillage may merely redistribute the carbon in the soil profile rather than affect the amount of carbon stored.

The magnitude and direction of change in soil carbon in relation to no-tillage treatments in bioenergy cropping systems remains uncertain and future work should focus on longterm experiments where soil profiles to 1 m are sampled with replicated tillage and notillage plots under different land use regimes in side-by-side comparisons.

## 1.2.4.2 Residues as a management strategy

It seems the most appropriate topic to follow tillage is the effects of residue management on the soil C and GHG balance of the soil, due to the close link these two management practices hold. Residues, also known as stover, may be defined in agricultural use as the parts of the crop that are not harvested and as a result are left on the soil. In bioenergy chains, residues have another meaning in that they can be the 'remains', 'wastes' or more commonly 'co-products' following primary energy or chemical extraction from the feedstock, and their end-use may have a large impact on the whole life cycle carbon cost of the bioenergy chain (Whitaker *et al.*, 2009).

Whether the residues are left on the soil or are removed will depend (i) on the management regime of that crop, (ii) whether the residues can be used as biomass, and (iii) economic limitations of the plantation. Residues as co-products of some crops can themselves be used as a renewable source of energy by conversion to bioethanol, which holds some great potential according to several authors; for example for the US alone, 244 million Mg of stover could be used to produce bioethanol every year (Tally, 2000). Use of forest residues left over from timber harvesting is an alternative proposed use of residues for bioenergy production. However the reduction of carbon stocks as a result of decreased litter input is large compared to the amount of energy produced, so would only be viable as a long term source of energy, for a minimum of 60-80 years (Repo *et al.*, 2015). Other options currently being considered for the use of crop residues is the CROPS idea: Crop Residue

Oceanic Permanent Sequestration. This is where crop residues are transported deep into the ocean floor to help sequester carbon dioxide, a technique boasting to be the most permanent and rapid solution to removing  $CO_2$  from the atmosphere (Stand & Benford, 2009). Whilst both of these ideas are interesting, one must consider the effect this removal will have on the land and the cost and benefits associated with these techniques. It appears from the literature that residue removal is generally considered detrimental to the management of crops, but this can vary depending on the soil and crop type (Andrews, 2006).

Residue retention can have various positive effects on the soil including decreased soil erosion and runoff, increased SOM, increased soil function, decreased disease-producing organisms, increased crop yields, enhanced soil biodiversity from habitat and substrate and increased soil sequestration (Andrews, 2006; Lal 2008; Franzleuebbers 2002). Many of these positive effects are interdependent and highly interactive with one another, and this has been summarised by Lal (2008) and can be seen in Figure 1.8. Much of the above discussed benefits are very direct effects on the soil but removal of residues also has indirect effects such as compaction from the increased use of machinery during removal which can in turn affect root growth and increase soil erosion (Wilhelm *et al.*, 2004). Here we will briefly discuss some of these benefits in more detail providing examples from experimental trials.



Figure 1.8: The interacting benefits of returning residues to the surface. (Adapted from Lal, 2008).

The surface cover provided by crop residues is extremely important and it is this loss of cover which results in many of the detrimental effects we see when it is removed. For example, residues largely influence the radiation balance and energy fluxes of the soil thereby reducing the evaporation rate (Wilhelm *et al.*, 2004). This is also linked to the change in yield seen when crops are removed. The effect of residues on yield, like all other effects, varies depending on soil type, crop and climate. Several authors have shown removal of residues results in reduction in yields (e.g., Wilhelm *et al.*, 1986; Linden *et al.*, 2000; Lal, 2008). Wilhelm *et al.* (1986) found reduced grain and biomass yield for corn and soybean crop attributable to reduced water availability and increased soil temperature. In certain conditions yields can be decreased by 10-20% due to residue removal (Lal, 2008). Ismayilova (2007) showed that the removal of two thirds of forest residues results in increased surface run off, increased sediment yield and increased transport of nitrogen and phosphorus. But it did show a decrease in the level of nitrate in the ground water of that area.

Residue retention is considered a major management strategy for sequestering carbon into the soil sink; calculations have estimated that global retention of residues on croplands can sequester 1 billion t C y<sup>-1</sup> which is equivalent to 30% of the annual increase in atmospheric  $CO_2$  (Karlen *et al.*, 2009). There have been many examples in the literature to support this contention: Bushford & Stokes (2000) estimated a 60% increase in soil C storage when residual slash is incorporated into SRC poplar plantations; Dolan (2006) found that retained stover residue stored significantly more SOC and soil N across the whole of the soil profile (0-45 cm).

It is clear that the ability to increase sequestration into the soil from the retention of residues is due to the increased C inputs. This was nicely shown by Paustian *et al.* (1992) using the CENTURY model, where there was a positive linear relationship between C inputs and SOC change; these findings have been confirmed by many field observations (see Refs within Wilhelm *et al.*, 2004). In a recent review, Lemke *et al.* (2010) reviewed 35 studies, finding 27 of these reported increase soil C for residue retention but only 7 of which were significant.

Quantitative data to compare the removal of residues for energy purposes or the remainder in the soil for sustainability and GHG balance are limited, but are likely to be crop specific. In the UK context, with future emphasis on SRC and Miscanthus which have minimum residues, this is likely to not be an issue of significant concern.

In summary, it is extremely important for soil health that residues remain, due to the various benefits as described above; this also has the benefit of saving money for the land managers as work is not required to remove and dispose of residues. In a comparison of the use of residues for ethanol production versus improving soil quality, Lal (2008) concluded that residues should be retained to improve soil quality, despite the large potential for producing bioethanol from residues.

### 1.2.4.3 Fertiliser as a management strategy

Several reports in this review suggest that the largest component of GHG balance in bioenergy cropping systems is that determined by fertiliser usage and consequent N<sub>2</sub>O emission, with associated increased nitrate in soil and water, run-off leading to eutrophication and long-term application leading to decreased soil health. Over 30 million tonnes of fertiliser was utilised in the EU in 2008, with 7.1 million tonnes of this being nitrogen surplus – equivalent to 55 kg N ha<sup>-1</sup> (Henriksen *et al.*, 2011). Fertiliser production also has a CO<sub>2</sub> cost, with the production of inorganic fertiliser using 1.2% of the world's energy and being responsible for 1.2% of global GHG emissions (Kongshaug, 1998).

The use of fertilisers is largely associated with 1G energy crops, in the UK context, but can also be applied to 2G energy crops such as SRC willow and *Miscanthus*, particularly when yields may begin to decline after third rotations; however, limited experimental evidence or commercial practice is available on which to make generalisations. The application strategy will vary dependant on the individual needs of the sites, but generally 1G bioenergy crops are fertilised every year. For SRC and *Miscanthus* which grow very rapidly, it is difficult to fertilise in the years after establishment, so all fertilisation is usually completed in year one.

There are two broad categories of fertiliser, namely organic and inorganic. Organic fertilisers are a more traditional means of fertilising crops and can include manure - the faeces of animals such as cows and horses - and sewage sludge which is produced from an array of organic and sewage wastes. Inorganic, or chemical fertilisation, is the most widely applied type of fertiliser in UK agricultural practices; most commonly this is made up of phosphorus (P), potassium (K) and nitrogen (N) in varying ratios depending on the user needs. The rise in atmospheric nitrous oxide  $(N_2O)$  is the main consequence of fertiliser application and animal production (IPCC, 2007), and is of particular concern as a GHG because it is 298 times more potent than  $CO_2$  (IPCC, 2007). An understanding of the point at which fertiliser application will no longer benefit yield and also limits the amount of nitrous oxide flux coming from the soil is important to maximise economic benefits and minimise environmental impacts. A small decrease in the amount of fertiliser can show large changes in the amount N<sub>2</sub>O flux; for example, a study of a corn-wheat rotation showed that reducing fertiliser by only 25 kg N ha<sup>-1</sup> (to 125 kg N ha<sup>-1</sup>) caused a 34% reduction in N<sub>2</sub>O flux without significantly changing yield (Sehy et al., 2003). One estimate of this equilibrium amount of N-application has been suggested for corn-soybean rotations in the Midwestern US at 135 kg N ha<sup>-1</sup> a value which restricts  $N_2O$  emissions and does not significantly affect yield (Sawyer et al., 2006).

It is also important to understand the way in which these emissions arise and the time frames over which they occur after fertiliser application. In a comprehensive study by Hoben *et al.* (2011), it was shown that the increase in soil inorganic N occurred within 11 days of application and the majority of the N<sub>2</sub>O emissions occur in weeks 4-8 after application. They concluded that the way in which these fluxes occur are non-linear, and often exponential with increasing N concentration after fertiliser application, with 180 and 225 kg N ha<sup>-1</sup> causing a 44% and 115% increase in N<sub>2</sub>O flux over the baseline 135 kg N ha<sup>-1</sup>. Other studies have highlighted the long duration over which N<sub>2</sub>O fluxes continue to

be seen after application: for SRC willow and poplar, enhanced  $N_2O$  emissions were seen for up to 4 weeks (Hellebrand *et al.*, 2008).

As well as chemical fertilisation, sewage sludge can be used as an agricultural fertiliser as it contains essential crop nutrients, such as nitrogen and phosphorus. The UK produces approximately 1.35 million tonnes annually, a proportion of which is used as an agricultural fertiliser (UK Water Report, 2009), so there is great potential to use this as an alternative to chemical fertilisers. Potential advantages of using sewage sludge is that 40-60% of the nitrogen is inorganic (DEFRA, 2003), therefore readily available for the plant to use, and the main attribution of emission from N application is due to the organic fraction of the nitrogen occurring though processes of nitrification and denitrification in the soil. Gilbert et al. (2011) compared the effect of inorganic fertiliser and sewage sludge to a no fertiliser reference scenario LCA, and found that inorganic fertiliser increased the Global Warming Potential (GWP) by 2% and sewage sludge increased it by a lower extent when applied to SRC willow and *Miscanthus*. This translates to a need for a <0.2 t ha<sup>-1</sup> yield increase to offset the emissions associated with producing these fertilisers. Applications of sewage sludge and waste water as a means of fertilising bioenergy crops has also proven to be economically advantageous in Europe due to increased yields and reduced fertilisation costs (Dimitriou & Rosenquist, 2011; and references within).

Several studies have shown that addition of organic fertilisers can increase SOC (Iazurralde *et al.*, 2001; Kaur *et al.*, 2008; Fronning *et al.*, 2008; Hellebrand *et al.*, 2008). In a comparison between organic and inorganic fertilisers on SOC under a corn-wheat cropping system it was shown that in both cases SOC increased, and those active fractions of SOC increased significantly, specifically for the integrated use of both organic and inorganic fertiliser (Kaur *et al.*, 2008). In a perennial cropping system of SRC willow and poplar, fertiliser blocks showed increased SOC, perhaps due to increased crop residues, whereas non-fertilised blocks experience significant decreases in SOC (Hellebrand *et al.*, 2008). This study also showed that annual crops had higher N<sub>2</sub>O fluxes than perennial SRC willow and poplar.

Different bioenergy crops are able to utilise different amounts of N-fertiliser; for example, in a comparison between annual and perennial crops it was shown that annual cropping combined with fertiliser application doubles the amount of N<sub>2</sub>O emissions (4.3 kg ha<sup>-1</sup> vs. 1.9 kg ha<sup>-1</sup>), indicating that the use efficiency of nitrogen was greater for perennial plants (Kavdir *et al.*, 2008). Corn, a principle feedstock in the USA, has the highest application rates of both fertiliser and pesticides per hectare (FAO, 2008) whereas an SRC willow

plantation will often be unfertilised and has very few pests. Trials with SRC willow have shown that yield increases can be obtained from modest application rates of 60 kg N ha<sup>-1</sup>, with declines in productivity observed with higher application rates (Sevel *et al.*, 2014). *Miscanthus* is a commonly fertilised 2G bioenergy crop, and recent analyses have shown that the yield increases achieved are more than sufficient to offset associated N<sub>2</sub>O emissions (Roth *et al.*, 2015).

Large scale cultivation of bioenergy crops requires that all species grown are consistent and reliable in terms of yield and response to management treatments such as fertiliser. Work conducted with SRC poplar, to be used as an energy crop, showed that while landfill leachate fertilisation was able to increase aboveground biomass the trait response of these trees varied depending on the clonal variety treated (Zalesny Jr. *et al.*, 2009). Whilst it is important to plant mixed varieties to increase resistance of the crop as a whole, such clonal variation can be problematic in terms of economic returns for fertiliser applied versus yield out, which may be off putting to certain farmers. Therefore in order for the cultivation of bioenergy crops to remain an attractive investment such variation needs to be restrained to within reasonable limits. Work on SRC and *Miscanthus* suggests that nitrogen fertiliser application may be the most significant management practice determining GHG balance (Drewer *et al.*, 2012).

Fertiliser application in bioenergy cropping systems may lead to large emission of  $N_2O$ , contributing the most significant part of the GHG balance for these cropping systems. Future efforts to improve NUE (nitrogen use efficiency) are urgently required, as are management strategies to reduce unnecessary fertiliser use.

### 1.2.4.4 Water use and irrigation as a management strategy

The water footprint of bioenergy cropping systems has proved to be controversial in recent years. In the USA, reports suggest detrimental effects on water supply following largescale cultivation of perennial energy grasses such as *Miscanthus* (VanLoocke et al., 2010; Phong et al., 2011), with water-use increased more than 50% compared with corn. The water-use footprint of 13 biofuel/energy crops was estimated by Gerbens-Leenes et al. (2009) and showed that Jatropha (a tropical 2G crop used for biodiesel) used more water than all 1G crops studied, including five times the water used for ethanol corn. However, all of these reports rely on modelled data or inventories: these are blunt tools with which to determine future policy, since there is very little experimental evidence on which to base model assumptions. These models also assume uniform cultivation across landscapes, but plantations can be managed and sited to more effectively use limited water resources. Indeed, when spatial water use and variation in crop cover were incorporated into hydrodynamic models, VanLoocke et al., (2010) were able to identify less sensitive areas for Miscanthus cultivation and reduce predicted hydrological impacts. Such areas should be targets for experimental verification, enabling the development of prescriptions for hydrologically and environmentally sustainable Miscanthus cultivation. Water use in SRC and Miscanthus has been quantified and work by Finch and Richte (2008), suggests lower transpiration rates when compared to grass, winter wheat and corn; however interception losses due to an extensive canopy may be higher in *Miscanthus* (Finch and Riche, 2010). Vanloocke et al. (2010) also showed that water use in Miscanthus could out-pace supply in many areas of the mid-west USA, so there is cause for concern. In a similar modelling exercise, Bonsch *et al.* (2014) showed that in order to output 200 EJ y<sup>-1</sup> from bioenergy by 2095 would result in double the current agricultural water withdrawals. For SRC, it has been suggested that water use on a seasonal basis is greater than grass or arable crops and more similar to tall forest (Finch et al., 2004); although recent work on a ForestGrowth-SRC, a process-based model has shown that water use efficiency in poplar may be twice that of willow (Tallis et al., 2013), suggesting that there may be room for improvement in SRC genotypes if this high Water Use Efficiency (WUE) trait can be captured in future breeding programmes. It also highlights the limitations of process-based models parameterised for single genotypes, or from data sets in the literature, again representing blunt tools from which to make generalisations.

There is adequate data to suggest cause for concern for crop water use in SRC and Miscanthus, since water use may be greater than other crop types and may outstrip ecosystem water supply. The impacts of water-use, and interaction with droughted environments for soil GHG balance remain to be elucidated.

Irrigation is the practice of applying water to crops to aid their growth; plants which are not subject to irrigation are often referred to as 'rain-fed'. Irrigation is sometimes necessary to ensure adequate yields and encourage desirable traits but is associated with additional costs to the farmer as well as potential environmental problems. Negative impacts of irrigation include water pollution from run-off, increased soil erosion, salinisation and over-abstraction, though it does have some positive impacts on certain landscapes, such as increased biodiversity through the creation of new habitats (Baldock *et al.*, 2000). Approximately 70% of all freshwater withdrawn globally is used for agricultural purposes (Comprehensive Assessment of Water Management in Agriculture, 2007), so a potential increase in agricultural production, including bioenergy crops, could put global freshwater supplies under pressure through competition. Europe is expected to see increased winter rainfall and reduced summer rainfall leading to increased drought (IPCC, 2007). This may increase the need to irrigate in future climates.

Additionally, increased temperatures will result in higher evapotranspiration thereby increasing the need for irrigation, even if rainfall is not dramatically different in the future (IPCC, 2007). Land-use and water quality have bidirectional effects on one another; with land management having direct effects on water quality, but also the water quality of the land often dictating its use (Bhardwaj *et al.*, 2010).

However, current levels of irrigation in European bioenergy cropping are by no means excessive compared to the total amount of irrigation applied (crops food and fibre), with bioenergy crops using only 2.3% of the total irrigation water consumed in Europe (Dworak *et al.*, 2009). Only 3.2% of the total cropping area in Europe is taken up by bioenergy crops and of this total area only 1.9% is subject to irrigation treatments (See Fig 1.9; Dworak *et al.*, 2009). In a study where three scenarios were examined ('business as usual', 'increased irrigation water demand' and 'water saving scenario') it was shown that even with future climates and a 4.5-fold larger bioenergy cropping area by 2020, that water availability will not present a problem for consumption by bioenergy crops (Dworak *et al.*, 2009). Even the scenario where water use is more restricted will not affect the ability to produce large amounts of biomass, and in general the increased area will not require an increase in irrigation (Dworak *et al.*, 2009). More recent analyses suggest that if dedicated

bioenergy crops are prohibited from irrigation a substantial increase in the amount of land would be needed to produce the same level of output (Bonsch *et al.*, 2014).



**Figure 1.9:** Relative irrigation water consumption presented as a % of total irrigation water consumption for bioenergy crops (From Dworak *et al.,* 2009).

Presently, it is uncommon in the UK to irrigate plantations of 2G crops as the amount of rainfall is sufficient to support their growth to satisfactory yields, though irrigation is common in Mediterranean climates where summers are drier (Sevigne *et al.*, 2011). For example, ranges of applied irrigation for SRC poplar in different regions of Spain range from 2000-6500 m<sup>3</sup> ha<sup>-1</sup> y<sup>-1</sup> (Sevigne *et al.*, 2011; Sixto *et al.*, 2007), in some cases representing up to 48% of the total water consumption in high-density plantings (Sevigne *et al.*, 2011). *Miscanthus* generally has a much higher WUE (due to C4 photosynthetic pathway and a larger/deeper rooting system) and the amount of biomass used to contribute to the production of bioenergy crops is generally larger, with 1G crops only having about 50% of their aboveground biomass directly contributing to the production of biofuel (Wirsenius, 2000; Zeri *et al.*, 2013). 1G bioenergy crops therefore tend to continue to be

treated as if they were cultivated for traditional purpose, be it food or fibre, by being subjected to a level of irrigation scheduling.

Proper applied use of irrigation as a management strategy to reduce GHG could be effective; as an increase in biomass (as a result of irrigation) will lead to increased carbon sequestration as C is fixed into above- and belowground biomass (Henriksen *et al.*, 2011). Partial root-zone irrigation is one of the latest methods which could be effective at reducing the environmental impacts of irrigation (Henriksen *et al.*, 2011). This method works by irrigating half the root zone and allowing the other half to dry out, with the side which is irrigated being alternated periodically to prevent permanent damage being done. It has been shown to have little effect on the yield and physiology of the plant compared to full irrigation and conventional deficit irrigation, and confers a significant increase in Irrigation Water Use Efficiency (IWUE) across many crop types (Sadras, 2009; Kirda *et al.*, 2007)

To conclude, irrigation has the potential to increase carbon sequestration due to increased plant biomass but remains an environmental threat from the perspective of water availability, particularly in the face of climate change. Though at present very few bioenergy crops are irrigated, the need for irrigation may increase with future predicted climates. More effective irrigation strategies have a role to play also in GHG savings through reduced use of automated farm machinery and better use of irrigation water.

## **1.3 Conclusions**

The review has highlighted a number of trends and gaps:

- The review illustrates gaps in the literature. These are particularly apparent for forest transitions into first generation crops and uncertainties surrounding grassland transitions. Forests transitions may not be relevant in a UK context given the limited scope for enhanced planting of first generation crops for future feedstock supply and because in the UK context de-forestation goes against current policy and is unlikely to be an important LUC. This transition is, and will be, of great importance in tropical regions where deforestation for 1G feedstock supply is currently taking place. More detailed consideration needs to be given to the grassland to bioenergy transitions, since it is here where there is the largest paucity of data and because this represents an important transition for the UK. GHG benefits of this transition remain uncertain and may depend strongly on management regime, fertiliser use and grazing, as well as the age and soil conditions. Given these caveats, generalisations are difficult.
- Analysis of the literature reveals limited soils data that assess the whole soil profile down to 1m, and yet conflicting results on soil carbon sequestration are apparent, when only top soil layers are considered.
- The consensus for transition from annual arable to perennial grass and SRC crops suggests improved SOC, but the overall GHG balance to farm gate may be positive or negative and largely driven by fertiliser input and consequent  $N_2O$  emissions. Accumulation of SOC is in the range 0.44-1.1 tonnes C ha<sup>-1</sup> y<sup>-1</sup>.
- There is likely to be a negative impact on GHG balance of transition from grassland to first generation bioenergy cropping systems.
- The magnitude and direction in soil carbon in relation to no-tillage treatments remains uncertain for bioenergy cropping systems but for second generation crops, with long rotation times, this may not be significant, although long-term experiments are warranted with soil profile sampling to 1 m.
- Quantitative data to compare the removal of residues for energy purposes or the remainder in the soil for sustainability and GHG balance are limited, but are likely to

be crop specific. In the UK context, with future emphasis on SRC and *Miscanthus*, with minimum residues, this is likely to not be an issue of significant concern.

- Fertiliser application in bioenergy cropping systems may lead to large emissions of N<sub>2</sub>O, contributing the most significant part of the GHG balance for these cropping systems. Future efforts to improve NUE (nitrogen use efficiency) are urgently required, as are management strategies to reduce unnecessary fertiliser use. There is a strong case for improved real-time instrumentation in the network of sites to capture this and other trace GHGs.
- There is adequate data to suggest cause for concern for crop water use in SRC and *Miscanthus*, since water use may be greater than other crop types and may outstrip ecosystem water supply. The impacts of water-use, and interaction with droughted environments for soil GHG balance remain to be elucidated.

This review has revealed some major knowledge gaps and highlighted areas of uncertainty where further data are required. This evidence has informed the design of the PhD thesis in such a way that I will be assessing the impacts of a transition from grassland, assessing soil carbon to 1 m; and assessing the impacts of a commercial harvest on ecosystem carbon balance.

## 1.4 Systematic Review Methodology

Systematic search methodology (Pullin & Stewart, 2006) was used to collect publications for use in literature review and subsequent meta-analysis (Chapter 3) to understand the effects of land use change to bioenergy cropping systems. The initial search was conducted in 2011 using a structured search string (Figure 1.10) using Web of Scholar, Science Direct and Google Scholar to capture grey literature. The search was subsequently repeated in summer 2013 and early 2014 to capture the most up to date publications, but only using Web of Science and Science Direct. Google Scholar was not used in subsequent searches as it produced an extremely large number of hits with very few being of relevance to the literature review or meta-analysis. The search string used can be seen in Figure 1.10. For full systematic search methodology see Appendix A.



Figure 1.10 - Search terms used for systematic review and meta-analysis literature assimilation.

## 1.5 Aims of Thesis and Chapter outlines

### 1.5.1 Overall study aims

Biomass for bioenergy is a proposed means to provide both renewable energy and to sequester  $CO_2$  to help mitigate the effects of climate change. Utilisation of bioenergy in the UK would result in large landscape changes in the form of land use change. As learned from this literature review, little is currently understood about the long term effects of LUC to bioenergy in the UK, especially from grasslands. This thesis, therefore, aims to assess the current state of knowledge regarding the effects of land use change to bioenergy in temperate regions using a systematic review and meta-analysis. Through a field study, this thesis also aims to detect any difference in GHG balance between an established grassland and SRC willow plantation. Additionally, a commercial harvest took place in SRC willow during the experiment. This thesis aims to capture the effect of the harvest on the soil and ecosystem GHG balance of the SRC willow.

## 1.5.2 Chapter outline

## Chapter 2: Research Spotlight: The ELUM project: Ecosystem Land-Use Modeling and Soil Carbon GHG Flux Trial

Chapter 2 is a published paper from the journal *Biofuels* which outlines the Ecosystem Land-Use Modeling and Soil Carbon GHG Flux Trial (ELUM) project. The ELUM project was a multi-million pound, seven-member consortium project which looked to elucidate the effects of land use change to bioenergy in the UK. The final output of the project was a meta-model with a user-friendly interface which would allow predictions of the effects of LUC in the UK on soil carbon and GHGs for specified transitions. This PhD work contributed to the delivery of this project.

## Chapter 3: Land use change to bioenergy: A Meta-Analysis of Soil Carbon and GHG Emissions

This Chapter is a published paper which featured in a special edition of Biomass and Bioenergy from the 1<sup>st</sup> International Bioenergy Conference, which took place in Manchester, UK, March 2014. Chapter 3 is a quantitative analysis of the effects of LUC to bioenergy in temperate regions of the current published literature. This Chapter aims to quantify the effects of LUC on soil carbon and GHG emissions using meta-analysis.

## Chapter 4: The effect of land use change from grassland to SRC willow for bioenergy on GHG emissions and soil carbon: a paired site approach

Work conducted in previous Chapters highlights knowledge gaps in the literature of our understanding of the effects of a conversion from grassland to bioenergy species. This Chapter uses a paired site approach on a commercial scale bioenergy plantation of SRC willow and an ex set-aside grassland to help close these gaps. Whole ecosystem carbon balance and soil GHGs are monitored in the two land uses to allow conclusions to be made about the possible effects of LUC from grassland to SRC willow in Southern England.

### Chapter 5: Influence of land use, litter fall and litter decomposition on soil chemistry

This chapter aims to assess the influence of land use change on soil chemical and physical properties in both grassland and SRC willow, as well as the impacts of litter fall and litter decomposition in SRC willow only.

#### **Chapter 6: General Discussion**

The final Chapter discusses the findings of the thesis in relation to the viability of bioenergy in the UK. This Chapter discusses bioenergy in the context of climate change and current policy initiatives. It highlights the importance of the influence of management practices in determining the LUC impacts of 2G bioenergy cultivation. Study limitations are discussed and suggestions for further research needed to help close current knowledge gaps surrounding the effects of LUC to 2G bioenergy on soil carbon and GHG emissions.
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# CHAPTER 2: RESEARCH SPOTLIGHT: THE ELUM PROJECT: ECOSYSTEM LAND-USE MODELING AND SOIL CARBON GHG FLUX TRIAL

Harris, ZM, McNamara NP, Rowe RL, Dondini M, Finch J, Perks M, Morison J, Donnison I, Farrar K, Sohi S, Ineson P, Oxley JC, Smith P, Taylor G (2014) Research Spotlight: The ELUM project: Ecosystem Land-Use Modeling and Soil Carbon GHG Flux Trial. *Biofuels*, **5**(2), 111-116.

**Statement of contribution:** I coordinated the writing of this paper, writing the majority of the text and adding flow to the manuscript where other authors contributed writing.

# 2.1 Abstract

There is increasing interest in the use of non-food second-generation (2G) lignocellulosic feedstocks and a move away from food crops for bioenergy applications, but questions still remain on sustainability. Empirical data are needed to quantify the greenhouse gas (GHG) balance of land-use transition to lignocellulosic bioenergy cropping systems, to inform life cycle analyses and aid model validation.

The aim of this project 'Ecosystem Land Use Modelling & Soil Carbon GHG Flux Trial' is to produce a framework for predicting the sustainability of bioenergy deployment across the UK. This £4m consortium project is commissioned and funded by the Energy Technologies Institute, UK.

## 2.2 Introduction

Although bioenergy is considered to be one of the key renewable energy technologies set for future expansion [1], concern still remains over its sustainability. In particular, the greenhouse gas savings that are possible in many bioenergy and biofuel chains have recently been questioned [2]. Although bioenergy may help meet national and international targets for emissions reductions, significant work is still required to establish robust sustainability criteria [3, 4]. Within the UK, much of the current bioenergy feedstocks are imported, and this may have wider implications for a range of ecosystem services that we are only just beginning to quantify [5].

One of the main concerns surrounding commercial bioenergy deployment in the UK is the potential displacement of food production and disturbance of valuable landscapes and the ecosystem services they provide, including erosion and flood regulation, pest and disease control, pollination and habitat provisions and soil and water quality. If we are able to understand the effects of land-use change (LUC) to bioenergy cropping systems, in a UK specific context, more informed decisions can be made and more appropriate policies put in place to safeguard against use of unsuitable. The focus of the 'Ecosystem Land Use Modelling & Soil Carbon GHG Flux Trial' (ELUM) project is to assess the potential scale of a future UK bioenergy sector based on domestic production of 2G feedstocks.

The ELUM project is a consortium of seven UK partners; the Centre for Ecology & Hydrology (CEH), University of Aberdeen, University of Southampton, Forest Research, Aberystwyth University, University of Edinburgh and University of York, and is commissioned and funded by the Energy Technologies Institute (ETI), a public-private partnership between global energy and engineering companies and the UK Government. The 3-year, £4m project is coordinated by CEH and consists of four work packages (Figure 2.1a): a data mining exercise, meta-analysis and extensive literature review; cataloguing of soil carbon and changes in soil carbon pools using a paired site approach across the UK; establishment of paired field sites to assess the effects of direct LUC to bioenergy; processbased modelling to determine the effects of LUC to bioenergy in the UK using inputs from the other work packages. The ultimate product of the project will be a user-friendly and freely available meta-model that can predict the impacts of LUC to bioenergy spatially across the UK out to 2050. The transitions of interest are those from a primary land use, including grassland/pasture, arable and forestry to second generation bioenergy crops, including Miscanthus, short rotation coppice (SRC) willow and short rotation forestry (SRF). The model can also consider transitions into arable from forest and grassland. The

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ELUM project is unique in that firstly, the majority of experimental work is located on commercial farms across the UK rather than in controlled plot-scale experiments and secondly, that model and experimental work are brought together under one project.

## 2.3 Data mining exercise

The preliminary focus of this project is nearing completion and was to assess the current state of knowledge that exists in the literature for the effects of LUC to bioenergy on soil carbon and GHG emissions. This work was undertaken by conducting a systematic search of the literature using three commonly used search engines, namely Google Scholar, Science Direct and Web of Science, coupled to a meta-analysis. Exclusion criteria were clearly defined to allow papers to be selected and included in review and meta-analysis if appropriate. The findings of the literature review and meta-analysis showed that there are significant gaps in our knowledge surrounding LUC to bioenergy [6]. There were particular data gaps for transitions to SRF and high levels of uncertainty around transitions from grasslands to non-food bioenergy crops, largely reflecting the importance of management activities such as fertiliser input and tillage. This work confirmed the need for further studies to generate datasets to fill these gaps, from which to derive evidence-based models; which are scheduled to take place as part of the ELUM project. A systematic review was also conducted into the effects of bioenergy cultivation on a wide range of ecosystem services, highlighting the levels of uncertainty which surround our understanding of these changes, with significant data gaps in most types of ecosystem service [5].

#### This work is being undertaken by the University of Southampton.



**Figure 2.1:** (a) An overview of the project structure, with key tasks, responsible organisations and individuals, their linkage and outputs. (b) An overview of the network sites and the measurements made: eddy covariance, static soil GHG emissions and dynamic CO<sub>2</sub> soil respiration.

# 2.4 Field studies

The empirical studies in this project take four approaches: the large-scale sampling of paired field sites (original land use versus bioenergy) across the entirety of the UK; the establishment of a network of six paired sites where transitions to bioenergy are assessed for GHG emissions and soil C changes; an experimental plot field trial assessing the potential of different *Miscanthus* genotypes as future bioenergy crops; the use of  ${}^{13}CO_2$  pulse labelling of vegetation to track shorter term carbon flow through the plant and soil system.

#### 2.4.1 Paired site approach

A paired site approach is a method used to assess the effects of a treatment, in this case LUC to bioenergy crops, on a particular variable, soil carbon, over an extended duration of time. These 'space-for-time' replacements have been used in many ecological and environmental studies and are highly appropriate for measuring landscape-scale soil processes [7]. Fields where bioenergy crops of several different ages are cultivated were sampled to assess the changes in soil carbon over time. Critically, this technique overcomes the common challenge of repeated sampling of the establishment phase of perennial energy crops in the first few years, and thereby generates unique and meaningful data on the mature crop in different locations. The sampling gave good spatial coverage of the UK, with a total of 70 sites representing 117 sampled fields. A detailed soil analysis was conducted on over 10,000 soil samples on cores taken to depths of up to 1 m. Assessment of the soil carbon to depth below 30 cm is important as there have been several studies which have over- or under- estimated soil carbon due to insufficient sampling depths [8,9]. In addition, under SRF transitions laboratory experiments measured potential GHG fluxes under controlled temperature and moisture conditions, in combination with assessments of soil microbial populations. Physiochemical fractionation of soil samples is being undertaking to allow assessment of any changes in soil carbon pools and the likely direction of any future changes in soil carbon.

The wealth of soil data from the paired site approach will provide an extensive data set for model testing and parameterisation, whilst the fractionation work will allow the assessment of model performance in modelling soil carbon portioning and residence times.

This work is being undertaken by CEH, Forest Research, and the University of Edinburgh.

#### 2.4.2 A network of paired GHG sites

A network of six paired experiments at four sites has been established across the UK (Figure 2.1b), with contrasting climate and soil types; Scotland, Wales, northern and southern England. These sites cover five transitions to bioenergy including, arable to SRC willow, arable to Miscanthus, grass to Miscanthus, grass to SRC willow and grass to SRF. These sites were all planted at commercial scale on commercial farms following typical cultivation practices, and are representative of the crop species that would be cultivated in that area, such as SRF in Scotland, as shown in Figure 2.1b. Over 24 months all sites undergo periodic monthly gas sampling to assess soil fluxes of carbon dioxide (CO<sub>2</sub>) by infra-red gas analyser and nitrous oxide (N<sub>2</sub>O) and methane (CH<sub>4</sub>) using the static chamber approach with subsequent sample analyses by gas chromatography. At selected field sites, higher temporal resolution measurements of soil respiration are being made using automated chambers. As well as soil fluxes, each site is equipped with state-of-the-art eddy covariance systems which allows continuous measurements of whole crop ecosystem CO<sub>2</sub> exchange at a scale of a few 100 m<sup>2</sup> [10]. These instruments measure the 'covariance' of wind turbulence, air temperature and  $CO_2$  concentration 20 times a second to calculate  $CO_2$ fluxes across the crops upwind at any moment. Automatic meteorological stations at the sites measured weather variables to enable determination of the relationship with GHG fluxes.

These intensive measurements will not only produce a large amount of GHG flux data to allow us to understand the processes affected by LUC but will also feed into the modelling for parameterisation and validation.

This work is being undertaken by CEH, University of Southampton, University of York, Forest Research and Aberystwyth University.

#### 2.4.3 Miscanthus genotyping

The majority of *Miscanthus* planted commercially in the UK is the single clone *Miscanthus* x giganteus (Mxg). To determine the extent to which models based on Mxg will apply to future *Miscanthus* varieties under development, 15 diverse *Miscanthus* genotypes, including parental species *M. sinensis*, *M. sacchariflorus*, and their interspecific hybrids, were analysed to represent the morphological and physiological diversity present within the breeding population. Our aim is to understand the processes driving atmospheric CO<sub>2</sub> capture and carbon sequestration by *Miscanthus* crops and to assess genotypic variation in carbon sequestration potential. Phenotype analysis, GHG sampling and soil sampling have

been conducted across all genotypes and six have now been chosen for a more intensive analysis.

This work is being undertaken by Aberystwyth University and CEH.

# 2.4.4 <sup>13</sup>CO<sub>2</sub> pulse labelling

Large scale <sup>13</sup>CO<sub>2</sub>-pulse chase experiments have been conducted under adjacent SRC willow and *Miscanthus* plots in Lincolnshire and in diverse *Miscanthus* genotype plots at Aberystwyth. Crops were enclosed in large transparent tents and exposed to highly-enriched <sup>13</sup>CO<sub>2</sub> with plants, soils and soil-atmosphere CO<sub>2</sub> exchanges being sampled for months afterwards. These experiments will allow us to understand carbon allocation within the plant, the flow of carbon in soils and transformations of plant inputs to soils by microbial groups [11].

This work is being undertaken by CEH and Aberystwyth University.

# 2.5 Modelling

Detailed process modelling provides a mechanism to scale point values gained at the network of field sites, up to the whole UK, providing a resolution to 1 km<sup>2</sup> for the GHG balance of the land-use transitions to non-food bioenergy crops. Based on the outputs of the process model, which models yield, and soil processes, a user-friendly meta-model is being developed with a graphical user interface (GUI) to allow users to undertake spatial estimations for GHG emissions from bioenergy cropping. The model is able to predict effects out to 2050 for the whole UK, with areas unsuitable for transitions removed, such as urban areas or protected green sites. The user will be able to select an area of land anywhere in the UK, define a LUC transition and see the effects of the change on GHG and soil carbon (derived from the ECOSSE model [12]), with the addition of estimated yields (input derived from ForestGrowth[13,14], Forest-GrowthSRC [15] and Miscanfor models[16]). The meta-model and GUI will be available for download from the CEH and project webpages, enabling use by the wider scientific community, policymakers, land planners, and those with a commercial interest in developing sustainable bioenergy feedstock crops.

This work is being undertaken by the University of Aberdeen.

# 2.6 Future perspectives

The outcomes of the ELUM project include an increased understanding of the way in which transition to 2<sup>nd</sup> generation non-food crops impacts soil and crop processes related to GHG emissions. New data include GHG measurements and soil carbon stocks of relevance to national inventories. The meta-model and associated GUI will provide a valuable resource for the scientific community and a wide range of stakeholders including growers, land managers, energy companies and policy makers. The meta-model is due for public release at the end of the project in 2014. Overall the outcomes of the project should enable a better understanding and informed decision making around the deployment of sustainable bioenergy. For further information on the ELUM project please visit: <u>www.elum.ac.uk</u>.

# 2.7 Executive summary

- An extensive literature review and meta-analysis highlighted significant gaps in our current understanding of the impacts of land use change to bioenergy cropping systems on GHG balance.
- Paired site comparisons of land-use transitions into bioenergy has occurred at 70 land-use transitions across the UK, assessing the longer term effects of established bioenergy plantations on soil carbon stock compared to their original land use.
- A network of new sites has been established across the UK to monitor GHG emissions and soil carbon changes occurring after/with a transition to bioenergy cropping systems, representing a world-leading infrastructure from which to assess long-term impacts of land-use change on GHG emissions.
- Plot experiments are being used to assess the mechanisms underpinning *Miscanthus* soil carbon sequestration and test if there is a difference in the carbon sequestration and yield potential of 15 *Miscanthus* genotypes representing the diversity within the UK *Miscanthus* breeding programme.
- Modelling efforts combine all aspects of the ELUM project and data collected to
  provide detailed effects of land use conversion for bioenergy in the UK. The final
  product will be a user-friendly model intended to provide an industry standard for
  land planners, industry and policymakers.

# 2.8 Financial & competing interests disclosure

Funding for this project was provided by the Energy Technologies Institute (ETI). The Centre for Ecology & Hydrology, Forest Research also directly funded elements of the ELUM project. The authors declare no other financial interests or conflicts.

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# CHAPTER 3: LAND USE CHANGE TO BIOENERGY: A META-ANALYSIS OF SOIL CARBON AND GHG EMISSIONS

Harris ZM, Spake R, Taylor G (2015) Land use change to bioenergy: A meta-analysis of soil carbon and GHG emissions. *Biomass & Bioenergy*, **82**, 27–39.

Supplementary material can be found in Appendix B.

**Statement of contribution:** This study was conceived as part of workpackage 1 in the project ELUM. I performed all data collection and data selection. I conducted the metaanalysis with the help of Rebecca Spake who wrote the script for the R programming environment. I drafted the manuscript, with all authors contributing.

# 3.1 Highlights

• Meta-analyses were conducted on 27 publications concerning land use change to bioenergy.

• Transitions from arable to 2G bioenergy crops showed an increase in soil carbon.

- Transitions from forest to 2G bioenergy crops showed a decrease in soil carbon.
- Uncertainty exists in predicting the impact of transition to 2G crops from grasslands.
- Significant knowledge gaps exist for GHG balance associated with transition to 2G energy crops.

### 3.2 Abstract

A systematic review and meta-analysis were used to assess the current state of knowledge and quantify the effects of land use change (LUC) to second generation (2G), non-food bioenergy crops on soil organic carbon (SOC) and greenhouse gas (GHG) emissions of relevance to temperate zone agriculture. Following analysis from 138 original studies, transitions from arable to short rotation coppice (SRC, poplar or willow) or perennial grasses (mostly *Miscanthus* or switchgrass) resulted in increased SOC ( $+5.0 \pm 7.8\%$  and  $+25.7 \pm 6.7\%$  respectively). Transitions from grassland to SRC were broadly neutral (+3.7)  $\pm$  14.6%), whilst grassland to perennial grass transitions and forest to SRC both showed a decrease in SOC ( $-10.9 \pm 4.3\%$  and  $-11.4 \pm 23.4\%$  respectively). There were insufficient paired data to conduct a strict meta-analysis for GHG emissions but summary figures of general trends in GHGs from 188 original studies revealed increased and decreased soil CO<sub>2</sub> emissions following transition from forests and arable to perennial grasses. We demonstrate that significant knowledge gaps exist surrounding the effects of land use change to bioenergy on greenhouse gas balance, particularly for CH<sub>4</sub>. There is also large uncertainty in quantifying transitions from grasslands and transitions to short rotation forestry. A striking finding of this review is the lack of empirical studies that are available to validate modelled data. Given that models are extensively used in the development of bioenergy LCA and sustainability criteria, this is an area where further long-term data sets are required.

# **3.3 Introduction**

Over the last three hundred years, more than half of the global land surface has been impacted by human activity [1] and [2]. Land Use Change (LUC) is a major driver of global environmental change [3] and [4] and also an important driver of increased greenhouse gas (GHG) emissions, contributing to the  $180 \pm 80$  Pg C rise in atmospheric CO<sub>2</sub> between 1750 and 2011 [5]. LUC may lead to altered soil organic carbon (SOC) and changes in a host of ecosystem services [6], [7], [8], [9] and [10]. The majority of LUC is driven by demand for food, fibre and fuel and the nexus between water, energy and food is now clear, with much on-going debate amongst scientists and policy makers on how we can achieve intensification of land use whilst at the same time preserving natural capital [11].

There is an urgent need to mitigate the impacts of LUC, through sustainable land management strategies that include renewable energy technologies such as bioenergy, which has the potential to provide both carbon sequestration and a displacement of fossilbased fuels. Renewable energy targets across Europe and in both national [12] and international [13] future energy scenarios, suggest a central role for bioenergy where 10–20% of primary energy supply is provided from green plants in some form, including to generate electricity, heat and liquid transport fuel [14]. In order to reach future targets, a substantial increase in bioenergy crop plantings will be required. In the UK, for example, recent estimates show that there is a potential 35 000 km<sup>2</sup> of land available for dedicated lignocellulosic bioenergy cropping that would not impact on the highest quality agricultural land [15], with the potential to supply 66% and 62% of the total heat and electricity demand, respectively [16]. It is therefore important to quantify the direct impacts of LUC for GHG balance, SOC and other landscape scale effects, so that appropriate land management strategies can be put in place.

The carbon and GHG balance associated with dedicated bioenergy crops has been the subject of considerable debate in recent years. Empirical measurements on the direct impacts of land use change to bioenergy are only just starting to emerge [e.g. [17] and [18], with the indirect impacts of land-use change remaining difficult to quantify [19], [20], [21] and [22]. There are also conflicting messages from a fragmented literature regarding LUC, as these effects can vary depending on the starting land use, the initial carbon stocks, the management regime and the climatic region where the land exists. Quantitative syntheses are lacking which are able to bring this body of research together in a succinct analysis. Meta-analysis provides a useful approach to identify the general trends in the effects of
LUC to bioenergy cropping on GHG emissions and SOC. Meta-analyses are becoming increasingly common in the scientific literature, expanding out from the traditional subject area of clinical medicine into ecology and environmental science [23]. Meta-analyses are a robust statistical method of identifying trends and patterns that exist within the literature which may be overlooked or undervalued in a traditional narrative review [24]. Gou and Gifford [25] performed a highly-cited meta-analysis of the effect of LUC on SOC and found that transitions from forest or to arable resulted in decreased SOC, with several other large scale meta-analyses taking place in this research area following this [26], [27] and [28]. Here we are able to complement these studies by focussing our investigation on the effects of land use change to bioenergy cropping in temperate zones, relevant to recent policy development including the Renewable Energy Directive (RED) which requires a better understanding of LUC to bioenergy cropping for GHG savings [29].

The aim of this study was to assess the state of the current literature on LUC effects to bioenergy and to quantify the scale of these effects specifically focusing on SOC and GHG emissions. Our 'controls' were existing land uses - arable, grassland and forests, and our 'treatments' were the bioenergy land uses - '1<sup>st</sup> generation' crop ('1G'; food crops, e.g. wheat, corn, sugar beet etc.) and '2<sup>nd</sup> generation' crops (2G) grouped into short rotation coppice 'SRC' (short rotation coppice willow or poplar), 'perennial grasses' (e.g. *Miscanthus*, switchgrass etc.) and short rotation forestry 'SRF' (e.g. poplar, alder, birch, beech etc.; Table 3.1). The outcomes from this study will assist decision making for both land managers and policy makers regarding the effects of LUC to bioenergy cropping in temperate regions. In addition, we identify existing knowledge gaps which may be present to help direct future research efforts to close such knowledge gaps.



Bioenergy Land Use Type	Inclusive Species				
1 <sup>st</sup> Generation	Wheat	Triticale			
	Oilseed Rape	Canola			
	Corn	Sugar Beet			
	Barley				
Short Rotation Coppice (SRC)	Willow				
	Poplar				
Perennial Grasses	Miscanthus				
	Switchgrass				
	Reed canary gras	Reed canary grass			
Short Rotation Forestry (SRF)	Eucalyptus	Conifer			
	Alder	Beech			
	Birch	Poplar			
	Sycamore				

## 3.4 Materials & methods

#### 3.4.1 Systematic review scope

We followed standard systematic review methodologies [30] to collate empirical studies from temperate regions that measured SOC or GHGs in 'treatment' bioenergy plantations, relative to 'control' existing common land uses - arable, grassland and forests (see Appendix B.1 for glossary). The land use transitions of interest were grouped to cover a conversion from arable, grassland or forest to 1G, SRC, perennial grasses or SRF (Table 3.1).

We used a structured search string to ensure all relevant literature was captured without bias (see Appendix B.2 for systematic search query methodology). To ensure meaningful comparisons, publications had to satisfy strict inclusion criteria. These were as follows: (1) appropriate response metrics must be measured in the publications: SOC measures as C (carbon) in units of t ha<sup>-1</sup> y<sup>-1</sup> (or a convertible figure) and GHG emissions for crop life cycle, partitioned into CO<sub>2</sub>, N<sub>2</sub>O, CH<sub>4</sub> or 'all' measured as carbon dioxide equivalents (CO<sub>2</sub>e) in units of t ha<sup>-1</sup> y<sup>-1</sup> (or convertible figures); (2) studies featured transitions of interest (Table 3.1); (3) studies had to report both pre-existing (control) and post-conversion land-use (treatment) values for the response metric(s) of interest. Studies were also eligible if they documented a land conversion not strictly for use as bioenergy, but used similar land management practices as would be used for bioenergy cultivation. (4) study locations were relevant to a temperate climate i.e. within the 23.5° and 66.5° latitudinal band and (5) the species were inclusive of 1G and 2G bioenergy crops (Table 3.1), but only those able to be cultivated in a temperate region.

Data from relevant publications were extracted in pre-defined units for the meta-analysis; standard unit conversions were performed where necessary. Authors were contacted in instances where data were insufficiently reported for inclusion in the meta-analysis. For those studies that reported data in figures only, numerical information was extracted using DATATHIEF [31].

## 3.4.2 Statistical analysis

## 3.4.2.1 Effect size calculation

Three key values are required to perform a meta-analysis, a mean  $(\bar{x})$ , a standard deviation (SD) and a sample size (n) for the control and treatment. For each comparison, the log

response ratio (*lnR*) of SOC was calculated between a pre-existing land use (control group) and bioenergy (treatment group):

$$ln R = ln(\bar{x}_{bioenergy}) - ln(\bar{x}_{control})$$
(2.1)

where  $\bar{x}_{bioenergy}$  treatment is the mean SOC of bioenergy, post-conversion land use and  $\bar{x}_{control}$  is the mean SOC of the control, pre-conversion land use. The *lnR* describes the proportional difference in the response metric between control and treatment groups. The natural log transformation of the response ratio both linearizes the metric, treating deviations in the denominator and the numerator as equal, and normalises its otherwise skewed distribution [32]. *lnR* values can be transformed to show change more intuitively as percentage difference from control groups.

A negative effect size (lnR) indicates loss in SOC as a result of LUC to bioenergy; a positive effect size indicates an increase in SOC as a result of LUC to bioenergy.

## 3.4.2.2 Meta-analysis

Random-effects models [33] were applied to calculate overall effect sizes for the following LUC for SOC: Arable to perennial grasses, arable to SRC, forest to SRC, grass to perennial grasses and grass to SRC. Studies included in this meta-analysis differ intrinsically in the methods used, site characteristics, sampling depth etc. Random-effects models allow for different study-specific effect sizes and assumes that heterogeneity among studies in their true effect sizes is due to random variation around the overall mean effect of the population of studies [33]. Each study included in the meta-analysis is assumed to be a random sample of a relevant distribution of effects, and the combined effect estimates the mean effect in this distribution. If the 95% confidence intervals did not overlap zero, the treatment bioenergy land use transition was regarded as having significantly different SOC content than the control land use. The meta-analysis was weighted in that each study-wise effect size was weighted by the inverse of its variance [24] and [32]. All models used the restricted maximum-likelihood estimation (REML) estimate. Grand log response ratios characterising the mean log response ratio for a population of studies were backtransformed to represent more intuitive changes in terms of percentage difference in SOC relative to controls. We examined heterogeneity, the between-study variation, using a heterogeneity measure (Q), calculated by weighting the sum of squared differences between individual effects and the pooled effect, tested against a chi-square distribution. Restricted maximum-likelihood estimation was used to estimate  $T^2$  (see Appendix B.3 for

calculations [34]). All statistical analyses and calculations were performed in R version 3.0.2 [35] using the METAFOR package 1.9-3 [36].

Publication bias may be suspected if small positive studies are present without small negative studies [37]. This was tested by assessing funnel plots of effect size vs. standard error of the effect size (see Appendix B.4 [38]) using the METAFOR package [36]. Weighted regression with multiplicative dispersion using standard error as the predictor did not detect funnel plot asymmetry, (t=-1.66.  $_{df}$  = 136, p = 0.0994), indicating no evidence of publication bias.

A meta-analysis on the effect of LUC to bioenergy on GHG emissions was not conducted due to insufficient reporting of error terms. Therefore, with the data that were available, an arithmetic mean of the studies were calculated and presented in a standard histogram.

# 3.5 Results

Contrary to traditional statistical tests, in the case of a meta-analysis, the magnitude of the effect size is more important for interpretation of the results than the *p*-value [33]. *p*-values are able to indicate, with 95% confidence, that the result differs to the null hypothesis, and when read are rarely considered with the sample size. Effects sizes, produced as a result of meta-analysis, take into account the sample size of the included studies and weight them accordingly, thereby relieving the study of any bias due to larger sample sizes. Therefore whilst a grand mean may not be statistically significant it should not be discounted as not being of relevance to the research question; as the magnitude of the effect size indicates the general trends of the effects of LUC on SOC as observed in the literature. The Q statistic, indicated that all transitions studied showed a significant degree of between-study heterogeneity.

The literature search yielded c.8000 publications. Of these, 27 satisfied inclusion criteria concerning climate, LUC, bioenergy crops and appropriate SOC and GHG data (Fig. 3.1). For SOC there were 13 publications amounting to a total of 138 observations. There were insufficient data to conduct a meta-analysis on GHG data; therefore a summary table of the available data was produced representing 14 publications containing 188 observations. Of all the land use transitions initially targeted, it was only possible to perform meta-analysis on 5 transitions for SOC, and 8 transitions contributed to a summary figure for GHG emissions (Table 3.2) of the total 12 possible transitions we aimed to cover.

	Soil Organic Carbon	GHG Emissions
Arable $\rightarrow$ SRC	$\checkmark$	Summary figure
Grass → SRC	$\checkmark$	Summary figure
Forest $\rightarrow$ SRC	$\checkmark$	×
Arable→ Perennial Grasses	$\checkmark$	Summary figure
Grass $\rightarrow$ Perennial Grasses	$\checkmark$	Summary figure
Forest $\rightarrow$ Perennial Grasses	×	Summary figure
Arable $\rightarrow$ 1 <sup>st</sup> Gen Crops	×	Summary figure
Grass $\rightarrow$ 1 <sup>st</sup> Gen Crops	×	Summary figure
Forest $\rightarrow$ 1 <sup>st</sup> Gen Crops	×	Summary figure
Arable $\rightarrow$ SRF	×	×
$Grass \rightarrow SRF$	×	×
Forest $\rightarrow$ SRF	×	×

Table 3.2: Summary of data sufficiency for meta-analysis for land-use change to bioenergy cropping systems. Where this was not possible a summary figure was constructed.





Although SOC and GHG emissions are likely to vary with time since LUC and sampling depth, it was not possible to partition the studies according to these variables. The average time since transition across all studies was 5.5 years ( $X_{max} = 16$ ,  $X_{min} = 1$ ) for SOC. It was also not possible to partition by soil sampling depth, since the majority of studies considered SOC at the 0–30 cm profile only, although further depths were covered (ranges of 0–150 cm), these were inadequate for meta-analysis. Conclusions drawn from this meta-analysis can therefore be considered appropriate for the 0–30 cm sampling depth and c.6 years after transition to bioenergy cropping (Table 3.3). Longer-term experimental studies are lacking beyond this time-frame.

**Table 3.3:** Summary of changes in SOC as a result of LUC to bioenergy, showing time since transition and soil depth of included studies

Transition	Change in SOC	Average time since transition (years)			Average soil depth (number of studies at each depth)			
	(% ± SE)	Mean	X <sub>min</sub>	X <sub>max</sub>	0-30 cm 30-60 cm 60 cm+			
Arable – Perennial Grasses	25.7 ± 6.7	5.4	2	16	12 15 36			
Arable – SRC	5.0 ± 7.8	5.7	1	9	3 1 14			
Forest – SRC	-11.4 ± 23.4	5.7	2	9	1 1 5			
Grass – Perennial Grasses	-10.9 ±4.3	5.8	3	6	0 43			
Grass - SRC	3.7 ± 14.6	7.1	2	15	2 0 5			

## 3.5.1 Soil organic carbon

Sufficient data were available to analyse the effects of LUC on SOC from arable to both perennial grasses and SRC, both showing that a transition to 2G cropping resulted in an increase in SOC (Table 3.4, Fig. 3.2 and Fig. 3.3). Arable to perennial grasses showed a significant increase in SOC of +25% ( $\pm$ 6.7%). Arable to SRC showed an increase in SOC of +5.0% ( $\pm$ 7.8%), though this was not significant. As for forest transitions, there were only sufficient data for a transition to SRC, showing a loss in SOC of -11.4% ( $\pm$ 23.4%), though this was not significant. There was not a consensus on the effect of LUC to bioenergy cropping on SOC for grassland transitions. A transition from grass to perennial grass showing a significant decrease in SOC of -10.9% ( $\pm$ 4.3%) whilst a transition to SRC showed a slight increase in SOC of +3.7% ( $\pm$ 14.6%), though this was not significant.

Table 3.4: Meta-analysis outputs for land use transitions to bioenergy on Soil Organic Carbon
(SOC). Negative % change denotes a loss in SOC. n=number of studies.

	In(F	२)	% change				
	Effect Size	SE	Percentage Change	SE	<i>p</i> value	n	Refs
Arable – Perennial Grasses	0.23	0.03	25.7	6.7	<0.0001	63	[39]-[42], [48], [50]- [51]
Arable – SRC	0.05	0.04	5.0	7.8	0.2003	18	[41], [43]- [47]
Forest – SRC	-0.1209	0.11	-11.4	23.4	0.2589	7	[43], [46]
Grass – Perennial Grasses	-0.1158	0.022	-10.9	4.3	<0.0001	43	[42], [49]
Grass - SRC	0.04	0.07	3.7	14.6	0.6003	7	[46]-[47]



**Figure 3.2:** Percentage change of SOC as a result of land-use change to bioenergy crops, a) arable to perennial grass and b) grass to perennial grasses. Individual study data are shown and summary effect sizes are shown in red with the mean and 95% confidence intervals.



**Figure 3.3:** Percentage change of SOC as a result of land-use change to bioenergy crops, c) arable to SRC, d) grass to SRC and e) forest to SRC. Individual study data are shown and summary effect sizes are shown in red with the mean and 95% confidence intervals.

## 3.5.2 Greenhouse gas emissions

Meta-analysis of GHG emissions between control and treatment land uses was not possible due to inadequate reporting standards concerning error terms. Sufficient data were available to assess the effects on all GHGs of interest but not all transitions were covered. Fig. 3.4, Fig. 3.5 and Fig. 3.6 show the general trends of GHG changes as a result of LUC to bioenergy crops in the form of a summary histogram. The effect of LUC to bioenergy on CO<sub>2</sub> emissions can be seen in Fig. 3.4, showing that transitions from arable to 2G crops results in reduced emissions of CO<sub>2</sub>, -2.1 and -2.2 t  $ha^{-1} y^{-1}$  for SRC and perennial grasses respectively. The transition from arable to 1G cropping was broadly neutral with the few differences likely to be due to management regime, rather than crop species planted. Grassland to perennial grasses showed a slight reduction in  $CO_2$  emissions of -0.8 t ha<sup>-1</sup>  $y^{-1}$  and grass to 1G showed a slight increase in CO<sub>2</sub> emissions of 1.9 t ha<sup>-1</sup>  $y^{-1}$ . Grassland to SRC showed a more pronounced increase in CO<sub>2</sub> emissions of 6.7 t ha<sup>-1</sup> y<sup>-1</sup>, though this transition represents a change after only 7 years, whereas the previous grass transitions were around 25 year post-transition (Table 3.5). Forest transitions to bioenergy show the most pronounced changes in CO<sub>2</sub> emissions, with a transition to perennial grasses resulting in an increase of 20.8 t  $ha^{-1}$  y<sup>-1</sup> and a transition to 1G cropping showing the most pronounced emissions at 26.5 t  $ha^{-1} v^{-1}$ .



**Figure 3.4:** The effect of land-use change to bioenergy on CO<sub>2</sub> emissions. Standard errors are shown with *n* denoting the number of observations. Positive values represent emissions and negative values represent sequestration. Refs: [18], [41], [53]-[54], [57], [59]-[63].

**Table 3.5:** Summary of change GHG emissions as a result of LUC to bioenergy and the time since transition.

Transition	Change in C	GHG emissions as CO <sub>2</sub> -eq	Average time since transition		
	GHG (t ha <sup>-1</sup> y <sup>-1</sup> )		(years)		
	CO <sub>2</sub>	-2.2	14.8		
Arable – Perennial Grasses	$N_2O$	-0.2	12.7		
	$CH_4$	-0.4	9		
	CO <sub>2</sub>	-2.1	12.5		
Arable – SRC	N <sub>2</sub> O -0.2		11.9		
	$CH_4$	-0.2	6.5		
Arable – 1 <sup>st</sup> Gen	CO <sub>2</sub>	-0.1	20		
	$N_2O$	-0.1	20		
Grass – Perennial Grasses	CO <sub>2</sub>	-0.8	26.5		
	CO <sub>2</sub>	6.7	7.1		
Grass - SRC	N <sub>2</sub> O	2.5	2.2		
	$CH_4$	-0.007	1.5		
Grass – 1 <sup>st</sup> Gen	CO <sub>2</sub>	1.9	24.6		
	N <sub>2</sub> O	0.5	20		
Forest – Perennial Grasses	CO <sub>2</sub>	20.8	30		
Forest – 1 <sup>st</sup> Gen	CO <sub>2</sub>	26.45	30		

Fig. 3.5 shows the effect of LUC to bioenergy on N<sub>2</sub>O emissions for 5 transitions; there were insufficient data for the other land use transitions, as indicated on the graph. Similarly the effect of conversion from arable to 2G bioenergy cropping was a very small reduction of -0.2 t ha<sup>-1</sup> y<sup>-1</sup> for both SRC and perennial grasses for N<sub>2</sub>O. There was little effect on the conversion from arable to 1G cropping of -0.1 t ha<sup>-1</sup> y<sup>-1</sup> which again may be due to a change in management regime. The only transition where there was sufficient data for LUC from grassland to 2G cropping was grass to SRC which showed a slight increase in N<sub>2</sub>O emissions (2.5 t ha<sup>-1</sup> y<sup>-1</sup>), a transition to 1G showed an emission of 0.5 t ha<sup>-1</sup> y<sup>-1</sup>.

There were very limited data to assess the effects of LUC to bioenergy on methane emissions, with only 3 transitions being covered (Fig. 3.6). All transitions showed a very slight reduction in CH<sub>4</sub> emissions; arable to perennial grasses and SRC with -0.4 and -0.2 t ha<sup>-1</sup> y<sup>-1</sup> respectively, and grass to SRC with -0.007 t ha<sup>-1</sup> y<sup>-1</sup>. Current literature [17], [18], [64] and [65] and work currently being undertaken in the UK [66] indicates that methane only plays a minor role in the overall GHG balance during LUC to bioenergy cropping systems.



**Figure 3.5:** The effect of land-use change to bioenergy on N<sub>2</sub>O emissions. Standard errors are shown with *n* denoting the number of observations. Positive values represent emissions and negative values represent sequestration. Refs: [18], [41], [52], [54]-[56], [58].



**Figure 3.6:** The effect of land-use change to bioenergy on CH<sub>4</sub> emissions. Standard errors are shown with *n* denoting the number of observations. Positive values represent emissions and negative values represent sequestration. Refs: [18], [41], [59].

## 3.6 Discussion

## 3.6.1. Main outcomes

Using a total of 13 publications, containing 138 studies we have quantified the effects of LUC to bioenergy cropping for 5 out of 12 possible transitions for SOC. There were insufficient data to conduct a strict meta-analysis on GHG data, so a summary figure was constructed using 14 publications containing 188 studies for  $CO_2$ , covering 8 of the 12 LUC transitions, N<sub>2</sub>O and CH<sub>4</sub>, covering 5 and 3 of the 12 transitions, respectively. The transition across Europe and the USA. It is unlikely that we will see land converted from forest or agricultural croplands to bioenergy cropping, in these areas, with the most likely transitions from grasslands, ex-set aside lands or degraded lands that are unsuitable for crop production. This is concerning since most studies consider conversions from croplands and forests, in the case of transitions related to tropical ecosystems [27] where conversion from primary forests to sugarcane and maize resulted in more than a 25% loss of SOC. Here we have focused entirely on temperate zone LUC and provided a firm evidence base for policy and land management strategies.

For GHG emissions the effect of a conversion to bioenergy cropping is usually seen immediately, with land preparation and planting resulting in increased emissions [67]. After establishment, the crop may enable a net gain in SOC, until the net sequestration by the crop is equal to that of the initial emission event. It is only past this point, when the 'carbon debt' has been paid, that the crop be considered to be actively adding to the carbon sink. A number of LCA studies overlook the importance of the establishment phase of bioenergy planting following land conversions, as these will have a large influence over the resulting carbon debt which has to be repaid and similarly do not take management events into account, such as harvesting and fertilisation [68]. Several studies have shown that the initial landscape conditions and land-use history are key to determining the time required to repay the carbon debt as a result of LUC to bioenergy cropping systems [69]. Arable to bioenergy cropping showed decreases in emissions of CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub> in this analysis, across a timeframe of 1.5–23 years. Though the difference between 1.5 years and 23 years post-conversion is rather large the general trend is a decrease in emissions, with the mean time since transition approximately 10 years (Table 3.5). This change may reflect a difference in structure of the species, with 2G crops accumulating more biomass with a deeper rooting system [70] and as the result of change in management practice with reduced inputs such as fertiliser. Recent work on SRC and *Miscanthus* suggests that nitrogen fertiliser application may be the most significant management practice determining GHG balance [68] and [71].

For soil carbon a much longer time frame is often required to restore the land to its original, or new equilibrium, carbon stock as this pool develops much slower over time compared to the rate of GHG emissions [72]. The amount of carbon present in soil depends on the rate of decomposition of SOC to CO<sub>2</sub> by micro-organisms and the rate of organic matter input from plant residues; in temperate climates a new equilibrium is often achieved with an exponential change time constant of 30-40 years [73]. Soil carbon assimilation rates will vary from site to site and depend on the existing carbon pool, the soil properties and climatic region [61] and [74]. It is estimated that a conversion from annual to perennial rotations, or vice versa, will influence the SOC in mineral soils over a period of 30-50 years in temperate regions [75]. In our study we found that a conversion from arable to perennial grasses and SRC resulted in a net increase in the SOC of 25.7% and 5.0% respectively. Higher carbon accumulation rates are observed in perennial crops than annual crops across the literature [74], with the management and inputs largely influencing this difference [72] and [76]. The limited data on forest conversions indicate that a LUC from forest to SRC resulted in an 11.4% decrease in SOC, but in the UK this would not be a likely transition given policy initiatives to increase forest cover. Whilst we observed that a transition from arable to 1G cropping was broadly neutral for GHG emissions, there is likely more research needed here. This LUC represents a change from a 'food use' of the land - in all cases wheat cultivation, transitioning to sugar and oil crops for biofuel production which tend to have higher associated GHG emissions and are more intensively managed [54]. In this type of analysis it is very difficult tease out the effects of management on SOC and emissions when considering arable or 1G, although several meta-analyses have completed in an attempt to elucidate these effects [27].

This analysis delivers a mixed message on the overall effect of converting grassland to 2G bioenergy cropping, with no definitive change being indicated. SOC was found to decline by 10.9% for grass to perennial grass and increase by 3.7% for grass to SRC. This difference may be explained by soil sampling depth, where transition to perennial grasses only considered in the top 30 cm of the soil and transition to SRC had some studies which considered the 30–60 cm profile. For GHG emissions there were also mixed messages as a result of LUC. For CO<sub>2</sub> there was a small decrease of 0.8 t ha<sup>-1</sup> y<sup>-1</sup> emissions, for a conversion from grass to perennial grasses and a conversion to SRC or 1st generation

bioenergy cropping showed increased emissions for  $CO_2$  and  $N_2O$ . This result was not entirely surprising given that grassland are known to be highly variable in both quality and soil carbon stocks [77]. There are also very large differences in how grasslands are managed which will have a large impact on both SOC and GHG emissions [78].

Results from previous meta-analyses may allow us to infer the effect of transitions which we were unable to capture in these analyses. Gou and Gifford [25] measured 'plantations' which are managed forests which may result in similar effects of planting to SRF and found that LUC from pasture and forest to plantation resulted in a decrease in SOC of -10% and -13%, whereas a transition from arable cropping to forest plantation resulted in an 18% increase in SOC. Lagnière *et al.* [26] showed that the positive effects of afforestation on arable land on SOC was more pronounced than that in pastures and grasslands, which is in agreement with our findings where the most pronounced effects are as a result of LUC to 2G cropping whereas transitions from grassland to 2G show both increases and decreases in SOC.

## 3.6.2 Limitations

The main limitation of this review was that a meta-analysis could not be conducted for GHG emissions because the available data were largely unsuitable for meta-analysis techniques. Studies that measure whole ecosystem GHG emissions, such as eddy covariance, require expensive equipment resulting in low replication, in many cases yielding an n = 1. There is also the need for the pre-existing land use to allow comparison of a transition. Many studies measure the carbon and GHG balance of individual fields, forests and arable land and several look at bioenergy cropping, however few look at them together where they are comparable. Even fewer studies have measured the existing land use and capture the conversion process to the new bioenergy plantation.

The data included in the analysis were annualised to allow comparisons across different studies. Since the largest impact of LUC may occur over the first few years post conversion [64], [65] and [68], conversions studied over a shorter time frame are likely to show exaggerated changes in SOC and GHG emissions compared to those over a longer time-course and may be a source of error in the work reported here. However, an advantage of the studies included in this analysis is that they were all over similar time scales, up to approximately 15 years which for land use and SOC is relatively short term. However, the median time since LUC was only 3 years, and our analysis was limited by available data, which in future could be improved as new longer-term studies emerge. This

highlights the importance of taking into account the amount of time needed, postconversion, to determine the overall effect on the ecosystem, and if a loss of carbon is seen how long is will then be to repay this carbon debt. It should be noted that RCUK grants are rarely long enough for such experiments where at least 6–10 years data are needed.

## 3.6.3 Knowledge gaps & future research

This review has revealed a knowledge gap concerning the existence of robust, empirical studies investigating both the short-term and long-term consequences of LUC to bioenergy on SOC and GHG emissions in temperate regions. Just 13 and 14 published studies were available for meta-analysis for SOC and GHG emissions, respectively. A reason for this small number is that many studies incorporated experimental designs that suffered from pseudoreplication [79]. Furthermore, several studies had investigated SOC and GHG emissions in response to the LUC, but did not report the summary statistics that are required for meta-analysis ( $\bar{x}$ , n and SE). We urge that studies on LUC to bioenergy report such statistics to allow their inclusion in future meta-analyses. We assert the framework proposed by Whitlock [80] which states data should be archived with enough clarity and supporting information that they can be accurately interpreted by others.

Whilst research in this area is increasing, the most valuable data sets will come in two forms (1) replicated long term monitoring of an existing land use measuring the change of interest, monitoring of the conversion process and monitoring of the bioenergy crop and (2) a paired-site approach measuring two sites in parallel, with one representing an initial land use and the other representing the post-conversion bioenergy crop. The ideal design for assessing the impacts of LUC to bioenergy would be in the form of a Before-After, Control-Impact (BACI) design as this allows for both a change in the land use but the maintenance of a control site to allow any climatic variability to be taken into account [81]. Whilst these study designs are most desirable they are extremely difficult to execute on field scale due to the space and funds required, especially to measure whole ecosystem GHG balance.

For the transitions covered here there are two that demand further consideration. Firstly, transitions from grassland and secondly the lack of publications on transitions to SRF. There is large uncertainty surrounding transitions from grassland, a potentially very large carbon sink [82] with a global land converge of 25% of the earth's land surface [83]. Grassland degradation is a large threat to these sinks as recent results show globally almost 50% grassland have been degraded, with climate change and human activities being the

dominant causes resulting in 45.5% and 32.5% degradation respectively [84]. Grasslands have been shown to be extremely variable in their carbon stocks across different climatic gradients and management regimes [77] with sampling depth and bulk density considerations varying across published studies. In particular the effect of management of these grassland, including fertiliser application, type and intensity of grazing and rotation length, greatly affecting the overall GHG balance, especially with regard to N<sub>2</sub>O and CH<sub>4</sub> emissions which are more radiatively active than CO<sub>2</sub> [85]. Understanding these effects and applying the appropriate land management strategy, such as planting system and grazing intensity can help to manage the land more effectively for carbon sequestration [86]. As the average rotation of SRF is 18–20 years, it is difficult to cover the whole rotation period, with many studies thus far reporting mainly on biomass yields and effects of management regimes [87]. It is likely that transitions to SRF, from arable and (with less certainty) grassland will result in net GHG savings and increase SOC [88], [89], [90] and [91].

Based on the limitations and knowledge gaps discussed above we recommend [66] and [92]:

1. Studies should be designed to monitor the entire transition since capturing the effects of the conversion process would enhance our understanding of LUC to bioenergy.

2. Monitoring experiments at commercial scale should be maintained to assess the long-term effects of LUC.

3. Increased empirical research on the effects of LUC, especially for grassland transitions including rotational and permanent grass, and for SRF where there are limited data.

4. Authors are urged to make all data freely available with appropriate error terms, for meta-analysis.

In summary, we have quantified the impacts of LUC to bioenergy cropping on SOC and GHG balance. This has identified LUC from arable, in general to lead to increased SOC, with LUC from forests to be associated with reduced SOC and enhanced GHG emissions. Grasslands are highly variable and uncertain in their response to LUC to bioenergy and given their widespread occurrence across the temperate landscape, they remain a cause for concern and one of the main areas where future research efforts should be focussed.

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# CHAPTER 4: IDENTIFYING THE GOOD BIOENERGY OPTIONS - LAND USE CHANGE FROM GRASSLAND TO SRC WILLOW HAS AN IMPROVED CARBON BALANCE

Harris ZM, Alberti G, Jenkins JR, Rowe RL, McNamara NP and Taylor G (*Submitted*) Identifying the good bioenergy options - Land use change from grassland to SRC willow has an improved carbon balance.

Supplementary material can be found in Appendix C.

**Statement of contribution:** This research was conceived as part of workpackage 3 in the project ELUM. I performed all data collection and data analysis, with assistance from JRJ, GA, RLR who collected and analysed data from the automated soil respiration system (JRJ and GA) and undertook some soil and GHG analysis (RLR, NPMN). I conceived the message for the paper and drafted the manuscript, with input from all authors.

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## 4.1 Abstract

The effect of a transition from grassland to second-generation (2G) bioenergy on soil carbon and greenhouse gas (GHG) balance is uncertain, with limited empirical data on which to validate landscape-scale models, sustainability criteria and energy policies. Here we have quantified soil carbon, soil GHG emissions and whole ecosystem carbon balance for SRC bioenergy willow and a paired grassland site, both planted at commercial scale. We quantified the carbon balance of both sites for a two year period and captured the effects of a commercial harvest in the SRC willow. Soil fluxes of nitrous oxide (N<sub>2</sub>O) and methane (CH<sub>4</sub>) did not contribute significantly to the GHG balance of these land uses. Soil respiration was lower in SRC willow (911.6 gC  $m^{-2} y^{-1}$ ) than in grassland (1521.7 gC  $m^{-2} y^{-1}$ ) <sup>1</sup>). Net ecosystem exchange (NEE) reflected this with the grassland a net source of carbon with mean NEE of 118.9 gC m<sup>-2</sup> y<sup>-1</sup> and SRC willow a net sink, mean NEE -620.0 gC m<sup>-2</sup>  $y^{-1}$ . When carbon removed from the ecosystem in harvested products was considered (Net Biome Productivity) SRC willow remained a net sink of carbon. Despite negative NEE, soil carbon stocks (0-30 cm) were higher under the grassland possibly due to negative impacts of the soil disturbance associated with SRC crop establishment. There was a larger NEE and increase in ecosystem respiration ( $R_{eco}$ ) in the SRC willow after harvest, however the site still remained a carbon sink. Our results indicate that once established, significant carbon savings are likely in SRC willow compared to the minimally managed grassland at this site. Although these observed impacts on carbon balances may be site and management dependent, they add to the accumulating evidence that land use transition to 2G bioenergy has the potential to provide a significant improvement on the ecosystem service of climate regulation.
# 4.2 Introduction

Dedicated second-generation (2G) feedstocks offer the opportunity to provide biomass for both electricity and biofuels without utilising or competing with land for food crops (Stoof et al., 2015). Short rotation coppice (SRC) willow and poplar, and perennial grasses such as Miscanthus are likely to have positive impacts on soil properties (Kort et al., 1998), greenhouse gas (GHG) emissions (see refs within Rowe et al., 2009) and a whole basket of associated ecosystem services (Holland et al., 2015). Although recent reports suggest that energy and food may be produced in a multi-functional landscape in a sustainable way (Souza et al., 2015; Manning et al., 2015), many of these positive effects are dependent on land management, vegetation type, and in particular, the land use change (LUC) implemented when the bioenergy crop was planted (Milner et al., 2015). It is therefore important to consider how these crops will be placed within the landscape (Dauber et al., 2010) and the impacts of particular land use transitions on ecosystem services, of which climate regulation is of outstanding importance (Anderson-Teixeira et al., 2012). In 2013, 51 x  $10^3$  ha (0.8% total arable land) were used to grow bioenergy in the UK (DEFRA, 2014) and at the same time, it is estimated that there are  $3.5 \times 10^6$  ha of land currently available to grow bioenergy crops without impacting food production (Lovett et al., 2014), with estimated yields ranging from 6-12 t ha<sup>-1</sup> y<sup>-1</sup> for SRC willow (Hastings *et al.*, 2014). Adoption of bioenergy will inevitably result in large scale LUC, therefore it is important to consider which land classes are most suited to the conversion to minimise environmental damage and competition with food crops.

LUC, irrespective of crop type, may have many direct consequences on climate regulation, such as altered GHG emissions (IPCC, 2007a), changes in soil carbon (Guo & Gifford, 2002) as well as impacts on other ecosystem services and biodiversity (Sala *et al.*, 2000). Additionally for bioenergy crops, the impacts of indirect land use change (iLUC; Searchinger *et al.*, 2008; Melillo *et al.*, 2009; Finkbeiner, 2014) and that of quantifying the counterfactual land use (DECC, 2014; Mathews *et al.*, 2014) are increasingly recognised and considered in land use conversions. St. Clair *et al.* (2008) found that former land use is the most important consideration determining whether a transition to 2G bioenergy will result in a net source or net sink of carbon. A number of studies and meta-analyses have suggested that, although dependant on site, LUC from arable cropping to 2G bioenergy is most likely to result in neutral or net increases in soil carbon (Harris *et al.*, 2015; Qin *et al.*, 2012; Dimitriou *et al.*, 2012). Similarly, reductions in GHG emissions have also been reported for LUC from arable to 2G bioenergy (Drewer *et al.*, 2012; Gauder

et al., 2012; Zona et al., 2013a; Palmer et al., 2014), a proportion of which is attributable to change in management and land use intensity. However, there is much more uncertainty surrounding the effects of LUC from grassland to 2G bioenergy crops (Harris et al., 2015; Qin et al., 2015), partly reflecting the considerable variability that is found amongst grassland types with significant differences in management which can dictate GHG balance (Soussana et al., 2010). Though grassland may be managed to encourage a carbon sink (defined here as an ecosystem in which the net absorbance of carbon is greater than the net loss; Smith et al., 2014), other management practices such as fertiliser addition and grazing lead to large emissions of nitrous oxide (N<sub>2</sub>O) and methane (CH<sub>4</sub>). Ciais et al. (2010) suggested that emissions of N<sub>2</sub>O and CH<sub>4</sub> arising due to management practices, may offset approximately 70-80% of the net carbon sink in European grasslands. This indicates that conversion to 2G bioenergy cropping may result in additional GHG savings. Moreover, Styles and Jones (2007) demonstrated that initial cultivation emissions associated with LUC from grassland to SRC willow could be offset by GHG emissions savings from replacing fossil fuel usage, although the timescale for this 'payback' remains the subject of debate (Fargione et al., 2008; Don et al., 2012).

Two limitations are apparent when considering much of the literature in this controversial area. The first is that many studies rely entirely on modelled data with extremely limited or no validation (Cherubini et al., 2009) and this is worrying, given that outputs from such models, often parameterised for non-bioenergy 'exemplar' arable, grass and tree ideotypes, may be used to develop sustainability criteria and policy instruments in this complex area (Creutzig et al., 2012; Buchholz et al., 2014). Secondly, when empirical data have been captured for model validation, they have often been small research-scale plots of limited commercial relevance (e.g. Nikema et al., 2012; Zatta et al., 2014). Additionally, there are methodological considerations which may affect the conclusions drawn about LUC, such as soil sampling depth (Dolan et al., 2006; Blanco-Canqui & Lal, 2008) and calculation of soil carbon stocks using a fixed depth method (Walter et al., 2015). The aim of this study was to quantify the impacts of a land-use change at commercial scale from a grassland with limited management intervention, to that of SRC willow and to quantify the ecosystem GHG balance of this change seven years after conversion. During three years of measurement the SRC willow was harvested at commercial scale, and the impact of this activity on GHG balance and whole ecosystem carbon balance was also quantified.

# 4.3 Materials and Methods

### 4.3.1 Site description and management

This study was conducted in the south of England ( $50^{\circ}$  58' 38"N,  $0^{\circ}$  27' 33"W) in an established SRC willow plantation (8.1 ha) and permanent grassland with low inputs (7.4 ha).

SRC willow was planted in June 2008 on a grassland field, previously defined as set-aside (2000-2007; set-aside defined as land which is taken out of agricultural production to prevent over production and encourage land restoration) at a density of 15,000 stems ha<sup>-1</sup> in double rows with distances of 0.75 m in the row and 1.4 m between the rows (Forestry Commission, 2002). Prior to planting, the site was ploughed in 2007 and treated with herbicide (*Glyphos Supreme* at 3.5 1 ha<sup>-1</sup>, Headland Agrochemicals, Flintshire, UK) and insecticide (*Dursban* at 1 kg ha<sup>-1</sup>, Dow AgroSciences Ltd, Herts, UK). In 2008 the site was power harrowed and there was a further application of herbicide (*Glyphos Supreme* at 3.5 1 ha<sup>-1</sup>, BASF, Cheshire, UK) and insecticide (*Dursban* at 1 kg ha<sup>-1</sup>). The SRC willow was cut back in March 2009, further treated with herbicide (*Weedazol* at 10 1 ha<sup>-1</sup>, Nufarm, Bradford, UK) and then underwent a rotation of 5 years prior to harvest in April 2014.

The grassland site was enlisted in the set-aside scheme until 2004 and was maintained as low input grassland thereafter. There were no inputs to the site other than an addition of a total of 10 t of manganese lime across the site in April 2011. Management was variable year to year, with grazing by sheep once per year (2-4 weeks), or if this did not occur, the grass was mown to control grass height. During the experiment, the site was grazed for 2 weeks in 2012 and the grass was mown in August/September in 2013 and 2014. Mowed grass was left at the site and was not removed.

Mean annual rainfall at the sites is 794 mm and mean annual temperature is  $11.0 \,^{\circ}C$  (1960-2010; Met Office, 2015). The soil is silt loam (7% clay, 53% silt, 40% sand) with a pH of 5.5. Predominant winds occur from the south westerly position in both fields therefore eddy covariance towers were established in the north-easterly corner of the grassland and SRC willow in order to ensure enough fetch (Fig 4.1).



**Figure 4.1:** Site maps of grassland and SRC willow, including wind rose for each site showing a predominant north-easterly wind. Black circle indicates location of eddy covariance tower and meteorological station. Grey circles indicate experimental plots where soil GHG, litter fall, litter decomposition measurement were taken. 100 m rule indicated for scale.

### 4.3.2 Experimental timeline

The aim of this side-by-side comparison was to develop an intensive data set for all components of the ecosystem GHG balance from a commercial plantation over a period of two years, including bioenergy SRC harvest. The experimental set up was established in November 2011 and measurements continued through until December 2014 (Fig 4.2). Periodic sampling with a portable soil respiration system to assess soil CO<sub>2</sub> efflux started in November 2011. Static chambers to measure soil GHG fluxes (N<sub>2</sub>O and CH<sub>4</sub>) were also installed at both sites in November 2011 (Fig 4.1). Automated soil respiration chambers were installed in April 2012 in SRC willow only to measure both total and heterotrophic respiration through root-exclusion subplots (Ventura *et al.*, 2015). Eddy covariance towers were established in August 2012 and November 2012 for SRC willow and grassland, respectively. Aboveground biomass was taken four times in the SRC willow and twice in the grassland. Belowground biomass was measured twice in the grassland and estimated for the SRC willow. Leaf litter fall and leaf decomposition were measured in the SRC willow.



Figure 4.2: Timeline of experimental measures taken in grassland (G, green) and SRC willow (W, brown). Dots indicate spot measurements and line indicate continuous measurements. Arrows indicate when sites underwent harvest events. Blue box indicates timeframe of measurement over which annualised carbon balance figure (Fig. 4.8) was constructed.

### 4.3.3 Micrometeorological measurements

A meteorological station was installed in SRC willow in August 2011 and in grassland in November 2011 (Fig 4.2). Each station measured soil temperature and heat flux at three depths (5, 10 and 15 cm; TCAV, Campbell Scientific, Logan, UT, USA; HFP01SC heat flux plates, Campbell Scientific), soil water content using time-domain reflectometers (TDR CS616, Campbell Scientific), incoming photosynthetically active radiation (SKP215 quantum sensor, Skye Instruments, Powys, UK), net radiation (NR-LITE, Kipp and Zonen, Delft, The Netherlands), air temperature and humidity (HMP155A, Vaisala, Vantaa, Finland). Additionally, precipitation (52203, Young, Traverse City, Michigan, USA) and wind speed and direction (05103-5, Young) were measured at the SRC willow site only. At both stations, variables were measured at 0.1 Hz and then collected and averaged half-hourly using a CR1000 datalogger (Campbell Scientific). The 50 year (1960-2010) average monthly temperature and rainfall for the region were obtained from the UK Met Office (Met Office, 2015).

### 4.3.4 Soil GHG fluxes

Eight plots were established in random locations in the SRC willow and grassland in November 2011 to measure soil GHGs, soil chemistry, aboveground and belowground biomass; litter fall and litter decomposition (Fig 4.1). Within these plots, soil CO<sub>2</sub> efflux was measured at monthly intervals using a portable chamber (SRC-1, PP Systems, Amesbury, MA, USA) coupled with an IRGA (EMG-4, PP Systems). Every effort was made at each sampling date to measure soil efflux on bare soil at both sites, although it remains possible that small amounts of vegetation were inside the chamber and therefore soil CO<sub>2</sub> flux may be overestimated. Air temperature, soil temperature (stab probe, Testo, UK; 0-10 cm) and soil moisture (Theta probe, Delta-T, UK; 0-6 cm) were also measured around the chamber at the time of sampling. As soil temperature is generally a good predictor of soil respiration, a continuous data set was constructed using a simple exponential function and continuous soil temperature data measured at each weather station (Raich & Schlesinger, 1992; Raich *et al.*, 2002).

At each of the eight sampling locations,  $N_2O$  and  $CH_4$  soil fluxes were measured using closed vented static chambers (Hutchinson and Mosier, 1981; Smith *et al.*, 1995; Smith & Mullins, 2000) made of PVC base rings (8 cm high with a diameter of 40 cm), inserted in the soil to 5 cm depth, and chamber lids (20 cm high with a diameter of 40 cm). To determine GHG fluxes, headspace gas (10 ml) was sampled from a self-sealing septa in the

chamber lid using gas-tight syringes, at 0, 15, 30 and 50 min after closure; it was immediately stored in pre-evacuated gas-tight vials (3 ml, Labco Ltd, UK). Gas samples were analysed on a PerkinElmer Autosystem XL Gas Chromatograph (GC) fitted with a flame ionisation detector (FID) for CH<sub>4</sub> and an electron capture detector (ECD) for N<sub>2</sub>O. All results were calibrated against certified gas standards (BOC, UK; Case et al., 2014). N<sub>2</sub>O and CH<sub>4</sub> flux rates were determined by linear regression of the four sampling points for each chamber and by applying a temperature and pressure correction (Holland *et al.*, 1999). The analytical precision of the GC for standards at ambient concentration was approximately 2%, using two standard deviations as a measure of mean error. Sampling for soil GHG fluxes took place every month from November 2011 until December 2014. Sampling of the grassland initially took place in a smaller grassland site from November 2011 until August 2012 when sampling was moved to an alternative larger site (to accommodate eddy covariance equipment). Grassland sites were both sampled for GHG fluxes for 3 months to compare fluxes and there was no significant difference between the sites ( $t_{(4)}$ =-0.06, p=0.95). Non-CO<sub>2</sub> GHG fluxes were first converted into CO<sub>2</sub> equivalents using the global warming potentials over a 100 year horizon of 298 for N<sub>2</sub>O and 25 for CH<sub>4</sub> and then to carbon equivalents using a conversion factors of 0.2727 (IPCC, 2007b).

Six (two per plot: one root excluded, one total respiration) automated soil chambers were also established in the SRC willow in February 2012 (Ventura *et al.*, 2015). These chambers measured soil  $CO_2$  flux every 4 hours, and 3 of the chambers were placed in root exclusion chambers to allow the partitioning of autotrophic and heterotrophic respiration. Data from automated chambers were regressed against periodic measurements to test the agreement between datasets.

## 4.3.5 Soil analysis

Soil C was measured for 0-30 cm (15 cores) and to 1 m depth (3 cores) in both grassland and SRC willow (and initial grassland). Samples were only taken once during the experiment in October 2012. Fresh soil was sieved to 2 mm before being frozen at -80°C and subsequently freeze dried for minimum of 24 hours. A subsample of the freeze dried soil (20-30 ml) was milled to a fine powder in a ball mill (Planetary Mill, *FRITSCH*, Idar-Oberstein, Germany). A 200 mg subsample of the milled soil was used for the assessment of C concentration using an elemental analyser (Leco Truspec CN, Milan, Italy). Total soil C stock for the 0-30 and 0-100 cm fractions was calculated on an equivalent soil mass basis (Keith *et al.*, 2015).

## 4.3.6 Aboveground and belowground biomass and net primary production

### 4.3.6.1 Aboveground Biomass

In SRC willow, aboveground biomass was estimated from the stem:volume index (Pontailler *et al.*, 1997) which was calculated for 160 trees using stem diameter (22 cm from ground height; Rae *et al.*, 2004) and dominant stem height. Non-destructive sampling took place every year in winter during the experiment. Destructive sampling of SRC willow was also conducted prior to commercial harvest in November 2013, to allow an estimation of actual biomass from stem:volume index values. A linear regression, of stem:volume index against fresh weight, allowed estimation of total dry weight (kg tree<sup>-1</sup>) from trees which were non-destructively sampled (see table C1 for raw data). Total C contained in aboveground biomass was calculated by assuming that the amount of C contained in woody biomass was approximately 49.3%, calculated from an assessment on measured values in the literature for SRC (Fahmi *et al.*, 2007; Bridgeman *et al.*, 2008; Gudka, 2012; Sannigrahi *et al.*, 2010).

Willow leaf litter was collected in trays during the months of litter fall, July-December, to quantify leaf biomass. Leaf litter was oven dried at  $80^{\circ}$ C for 48 hrs, weighed and extrapolated from tray to tonnes per hectare. Litter decomposition was measured over two years in SRC willow. Mesh bags (20 x 10 cm; 1 mm aperture) each containing 5 g leaf litter (picked green leaves) were placed by each of the GHG chambers in November 2011. Bags were collected at several points post-insertion - 2 weeks then 1, 2, 4, 6, 9, 12, 18 and 24 months. Leaf litter was gently washed with water, then dried at 85°C for 24 hours before dry weight was recorded. See table C2 for extrapolation from measured values to t ha<sup>-1</sup>.

Aboveground biomass was estimated in the grassland by cutting all biomass within a 50x50 cm quadrat with hand shears flush to ground. Samples were taken twice during the experiment, in August 2013 and August 2014 prior to the mowing of the field. Samples were oven dried at 80°C for 48 hours, weighed and extrapolated from quadrat to tonnes per hectare.

#### 4.3.6.2 Belowground Biomass

Belowground biomass in SRC willow was estimated using a root:shoot ratio of 0.5:1 for annualised aboveground biomass according to (Pacaldo *et al.*, 2013a), where the belowground component encompasses belowground stool, fine root and coarse roots and the aboveground portion included aboveground stool and stem biomass. In the grassland,

belowground biomass was measured using 5 cm diameter auger and taken at three depths (0-10 cm, 10-20 cm and 20-30 cm). Roots were sieved consecutively through sieves of decreasing mesh size (3350, 2000 and 500  $\mu$ m), oven dried at 85°C for 24 hours, weighed and extrapolated from quadrat to tonnes per hectare (see table C3). Total biomass was calculated by summing total aboveground biomass and belowground biomass; for SRC willow the aboveground components included stem, branch and leaf biomass. Net primary production (NPP) was calculated on an annual basis using two consecutive harvest datasets.

#### 4.3.7 Eddy Covariance measurements

Eddy covariance towers were installed in SRC willow in April 2012 and in grassland in August 2012 to measure ecosystem  $CO_2$  fluxes. Each system consisted of an open path infrared gas analyser (Li-7500A, Licor, Lincoln, NE, USA) and a sonic anemometer (Windmaster Pro, Gill, Hampshire, UK). Data were logged at 20 Hz to an industrial grade USB stick in the LiCor interface box. Instrument height was 2.5 m from the ground for the grassland site. For SRC willow, instrument height was 8 m at the start of the experiment and extended as the crop grew to a maximum measuring height of 9.3 m in March 2014. After harvest the instrument height was reduced to 3.6 m above ground level.

Eddy covariance data were processed using Eddy Pro (Licor) and averaged over 30-minute intervals. The applied methodology was based on the EuroFlux protocol (Aubinet et al., 2000). Data were then elaborated and quality controlled using Stata IC 10 (StataCorp LP, College Station, Texas, USA). Data were rejected when fluxes came from outside the flux footprint which was between 135-262° for SRC willow and 140-290° for grassland. Gapfilling to estimate Net Ecosystem Exchange (NEE) and flux partitioning, intro Ecosystem Respiration (Reco) and Gross Primary Production (GPP), were done according methodology to the standard used in Fluxnet (http://www.bgcjena.mpg.de/~MDIwork/eddyproc/; Reichstein et al., 2005).

### 4.3.8 Ecosystem GHG Balance

A conceptual summary figure was constructed to represent the whole system GHG balance for both grassland and SRC willow for two whole years during the measurement period, January 2013-December 2014 (Fig 4.2, blue box). All gas flux data were expressed as gC  $m^{-2} y^{-1}$  and soil storage terms presented as standing stock (g m<sup>-2</sup>). The terminology used is as defined by Chapin *et al.* (2006), however we assigned a positive sign to emission of C to the atmosphere and a negative sign to an uptake of C by the ecosystem. Briefly, Net Ecosystem Exchange (NEE) was defined as the CO<sub>2</sub> exchange between the ecosystem and the atmosphere, measured using the eddy covariance technique. Gross Primary Productivity (GPP) was defined as the fixation of carbon by autotrophic organisms and Ecosystem Respiration ( $R_{eco,}$ ) is the net respiration by both autotrophic and heterotrophic organisms in the ecosystem. Soil respiration is the sum of CO<sub>2</sub> respired by roots (autotrophic respiration) and by microbes (heterotrophic respiration). Net Biome Production (NBP) describes the difference between Net Ecosystem Productivity (NEP, negative sign of NEE; Reichstein *et al.*, 2012) and the carbon removed through harvest. The sign of NBP is opposite to all other measures described above, where negative indicates a release to the atmosphere and a positive value indicates an uptake by the ecosystem. NBP was only calculated for SRC willow where harvested biomass was removed from the system; grassland biomass was cut and remained on the surface therefore leaving the C to remain in the system.

### 4.3.9 Statistical Analysis

A *t*-test was performed to detect any significant difference in soil carbon stocks at 0-30 cm (n=15) and 0-100 cm (n=3) between land uses, using SigmaPlot 12.5. All statistical analyses for GHG and eddy data were conducted in the R programming environment (R version 3.1.3). GHG data were analysed using linear mixed models (Bates *et al.*, 2014) where fixed effects were treatment, year, soil temperature and soil moisture. Air temperature and soil temperature exhibited collinearity so could not both be included in the model. Chamber number was used as a random factor to account for repeat sampling over time. Main effects were tested in addition to all second order interactions. Analysis of N<sub>2</sub>O and CH<sub>4</sub> reveal normality of residuals and homoscedasticity, however there was heteroscedasticity detected in the CO<sub>2</sub> data therefore log-transformation was performed. Model selection was performed according to Crawley (2007) using AIC to construct the minimum adequate model (See table C4).

For eddy covariance data a global model was constructed to assess the effects of land use and climate variables (Fixed effects: treatment, photosynthetically active radiation (PAR), wind speed, rain, soil temperature, relative humidity (RH) and soil water content; random effect: date) on NEE using daily averaged data. There was collinearity between air temperature and soil temperature so only one was used in the model, likewise for soil water content at both depths. Data were then partitioned by site and two separate models were constructed for each data set to see if the drivers of NEE differed between fields.

# 4.4 Results

# 4.4.1 Weather patterns

Air temperatures in 2012 were close to average values for the region. Spring of 2013 was cooler than average, whereas winter 2013 and spring of 2014 experienced higher than average temperatures (Fig 4.3). The spring/summer of 2012 and winter 2013 were notably wet years with above average rainfall for the region, whilst in contrast the spring and summer of 2013 were drier than average (Fig 4.3). 2013 was cooler and much drier than both 2012 and 2014 with an average air temperature of 9.9°C and rainfall of 673.3 mm. 2012 was slightly cooler but wetter (10.6°C and 1318 mm) than 2014 which experienced an average temperature of 11.1°C and 1023 mm rainfall.



Figure 4.3: (a) Monthly mean values of air temperature (°C) showing 50 year average (1960-2010; black line) and values measured by site met station (grey line). (b) Sum of rainfall (mm) for 50 year average (1960-2010; black line) and measured on site (grey line).

## 4.4.2 Net Primary Production

Total aboveground biomass in SRC willow increased from the first measurement, March 2012, to the final measurement before the harvest, November 2013 (Fig 4a). Biomass was rapidly accumulated after the harvest in April 2014 with total aboveground woody biomass reaching  $11.4\pm1.1$  t ha<sup>-1</sup> (mean±SE) by the end of 2014. Leaf litter was similar for 2012 and 2013 with 5.6±0.2 and 5.8±0.2 t ha<sup>-1</sup> y<sup>-1</sup> respectively. There was a decrease in leaf litter fall after the harvest in 2014 with only  $2.1\pm0.2$  t ha<sup>-1</sup> y<sup>-1</sup>. The majority of SRC willow leaf litter decomposed within the first year, with only 17% leaf litter remaining after 12 months and only 8% remaining after 2 years (Fig C1). Total grassland biomass was over double that in 2014 compared to 2013, for both aboveground and belowground biomass (Fig 4b). Total biomass in 2013 was higher in SRC willow ( $55.7\pm2.9$  t ha<sup>-1</sup>) than grassland ( $8.7\pm1.5$  t ha<sup>-1</sup>), but was similar in 2014 after SRC willow had been harvested and begun regrowing ( $18.3\pm1.4$  and  $20.8\pm1.6$  t ha<sup>-1</sup> for SRC willow and grassland respectively). The NPP post-harvest was slightly reduced compared to 2012 and 2013 (Fig 4c). In 2014, the NPP in grassland, 4.9 t C ha<sup>-1</sup> y<sup>-1</sup>, was less than that of SRC willow, 6.8 t C ha<sup>-1</sup> y<sup>-1</sup> (Fig 4c).



**Figure 4.4:** (a) Total biomass for SRC willow (t ha<sup>-1</sup>) including measured stem (checkerboard hatching) and leaf biomass (diagonal hatching), and estimated root biomass (grey fill). Error bars show standard error. (b) Grassland biomass from measured aboveground (checkerboard hatching) and belowground sampling (grey fill). Error bars show standard error. (c) Net primary productivity (NPP) for SRC willow (white bars) and grassland (black bars).

# 4.4.3 Soil respiration

CO<sub>2</sub> accounted for the majority of soil GHG flux, c.96% and c.99% for grassland and SRC willow respectively. Mean soil respiration (2012-2014) was higher in grassland (1532.4  $\pm$  38.9 gC m<sup>-2</sup> y<sup>-1</sup>) than in SRC willow (896.0  $\pm$  41.5 gC m<sup>-2</sup> y<sup>-1</sup>; Fig 4.5, Table S1, *p*=0.03). Year, soil temperature and soil moisture were all factors affecting soil respiration (*p*<0.001), as well as second order interactions involving treatment and year (*p*<0.001), treatment and soil temperature (*p*<0.001), and year and soil moisture (*p*=0.007; Table C5). According to continuous soil respiration measurements (Ventura *et al.*, 2015), heterotrophic respiration accounted for 84% of total soil respiration in the SRC willow.



**Figure 4.5:** Soil CO<sub>2</sub> flux (gC m<sup>-2</sup> d<sup>-1</sup>) for SRC willow (a; white circles) and grassland (b; black circles). Periodic sampling events (circles) and modelled CO<sub>2</sub> flux (black line; using soil temperature) are shown. Green line indicates 5<sup>th</sup> and 95<sup>th</sup> percentiles around the modelled values. Additionally, modelled CO<sub>2</sub> data are regressed against measured CO<sub>2</sub> data for both sites and the relationship shown on the graph as R<sup>2</sup>.

### 4.4.4 Eddy flux measurements

After quality control checks and footprint analysis of eddy covariance data, there was 40% for grassland and 37% for SRC willow of data remaining in 2013. In 2014, the remaining data for each site was 46% and 20% for grassland and SRC willow respectively. These data were subsequently gapfilled to give a complete dataset as described in methods. The energy balance closure for the sites was a 73% for grassland (Fig 4.6A) and 77% for SRC willow (Fig 4.6B).

For grassland, the NEE over two years (2013-2014) was 237.8 gC  $m^{-2}$ . In year one (2013), the grassland was a net source of carbon, 246.4 gC  $m^{-2}$  y<sup>-1</sup>, whereas in year two (2014) it was a net sink, -8.6 gC m<sup>-2</sup> y<sup>-1</sup>. In year one there was a small uptake of carbon during the growing season from June 2013 to the end of July 2013 (Fig 4.7a), however in year two there is a more defined uptake period starting from March 2014. This early onset of carbon fixation could be attributed to the higher mean monthly temperature in January-March 2014 compared to 2013 (Fig 4.3). SRC willow was a C sink for the two year duration of the experiment with a mean annual NEE of -620.0 gC m<sup>-2</sup> (Fig 4.7b). In the first year, which corresponded to the 4<sup>th</sup> year of growth, the site was a large sink of carbon (-901.4 gC m<sup>-2</sup> y<sup>-1</sup>). The NEE for the second year, after harvest in April 2014, was smaller (-338.7 gC m<sup>-2</sup> y<sup>-1</sup>). NEE was consistently lower in the SRC willow than in grassland (p < 0.001). Analyses of eddy covariance data also revealed that NEE in grassland and SRC willow were driven by different components (Table C6). In the grassland, PAR (p < 0.001), year, (p<0.001) soil (and air) temperature (p<0.001), wind speed (p=0.005) and rain (p=0.022)were factors affecting NEE. Whilst in the SRC willow only PAR (p < 0.001), year (p < 0.001) and soil water content (p < 0.001) were also factors affecting NEE. Relative humidity was not found to be a factor affecting NEE at either site.

There were also differences in the modelled estimates of  $R_{eco}$  and GPP between grassland and SRC willow.  $R_{eco}$  was 33% higher in 2014 than in 2013 in grassland (1261.3 gC m<sup>-2</sup> y<sup>-1</sup> and 1674.8 gC m<sup>-2</sup> y<sup>-1</sup> for year one and year two respectively).  $R_{eco}$  in SRC willow in year one was lower than both years in grassland at 971.4 gC m<sup>-2</sup> y<sup>-1</sup>. In 2014,  $R_{eco}$  was larger than year one in SRC willow and both years in the grassland site at 1970.6 gC m<sup>-2</sup> y<sup>-1</sup>. GPP in grassland was 1014.9 and 1683.4 gC m<sup>-2</sup> y<sup>-1</sup> for year one and two, respectively. In SRC willow, GPP was higher than the grassland for both years at 1872.8 and 2309.3 gC m<sup>-2</sup> y<sup>-1</sup> for year one and year two respectively.



**Figure 4.6:** Energy balance closure for grassland (a; black circles) and SRC willow (b; white circles) for 2013-2014, where H is sensible heat flux, LE is latent heat flux, G is soil heat flux and Sg is soil heat storage showing 30-min averaged data. Strength of regression indicted on graph by R<sup>2</sup> value. Red line shows 1:1 line, or 100% energy closure.



**Figure 4.7:** Net Ecosystem Exchange (NEE; gC m<sup>-2</sup> d<sup>-1</sup>) for grassland (a; black circles) and SRC willow (b; white circle) for 2013-2014. Harvest events at both sites are indicated by dashed arrows.

### 4.4.5 Belowground carbon pools

Soil carbon stocks (Table 4.1) for the 0-30 cm profile were higher in the grassland than in the SRC willow ( $t_{28}$ =-5.30, p<0.001), 63.4 ± 3.5 (mean±st.err.; n=15 cores) t C ha<sup>-1</sup> and 42.62 ± 1.8 t C ha<sup>-1</sup> respectively. The low replication of the 0-100 cm profile, however, did not detect a difference in soil C at this depth ( $t_2$ =-3.84, p=0.062; n=3 cores), where samples had 107.6 ± 1.8 t C ha<sup>-1</sup> in grassland and 77.3 ± 7.7 t C ha<sup>-1</sup> in SRC willow. Interestingly, the grassland which was used initially for chamber measurements had a similar C stocks to grassland in the upper 30 cm (61.2 ± 2.8 t C ha<sup>-1</sup>), but did not differ detectably in C in the 100 cm profile with the SRC willow (63.8 ± 4.1 t C ha<sup>-1</sup>; Table 4.1).

**Table 4.1:** Soil carbon stocks (t C ha<sup>-1</sup>) under grassland and SRC willow, calculated on an equivalent soil mass basis, for 0-30 cm and 0-100 cm. n=15 for 0-30 samples and n=3 for 0-100 cm samples.

	Grassland	SRC willow		Initial Grassland
Soil depth (cm)	Mean ± SE (t C ha⁻¹)			Mean ± SE (t C ha⁻¹)
0-30	63.35 ± 3.51	42.56 ± 1.75	*	61.2± 2.84
0-100	107.58 ± 1.82	77.28 ± 7.68	ns	63.84 ± 4.14

# 4.5.6 Soil GHG fluxes

N<sub>2</sub>O and CH<sub>4</sub> were not important contributors to the GHG balance of these two particular sites, accounting for less than 4% (3.4% N<sub>2</sub>O and 0.4% CH<sub>4</sub>) for grassland and less than 1% (0.77% N<sub>2</sub>O and 0.07% CH<sub>4</sub>) for SRC willow. Mean N<sub>2</sub>O fluxes at both sites (2012-2014) were very low (within detection limit of equipment) with emissions of 4.4 gC m<sup>-2</sup> y<sup>-1</sup> and 4.9 gC m<sup>-2</sup> y<sup>-1</sup> for grassland and SRC willow respectively (Fig C.2). There was no difference between N<sub>2</sub>O fluxes between the sites (*p*=0.81; Table C7). N<sub>2</sub>O flux was significantly affected by year across both sites (*p*=0.003), as well as an interaction between year and soil moisture (*p*=0.007). CH<sub>4</sub> was also very small at both sites, however there was a difference between the sites with an emission of 0.2 g C m<sup>-2</sup> y<sup>-1</sup> from grassland and uptake of -0.2 g C m<sup>-2</sup> y<sup>-1</sup> in SRC willow (*p*=0.003), Table C8, Fig S4). For both sites, soil temperature significantly affected CH<sub>4</sub> flux (*p*<0.001), as well an interaction between soil moisture and soil temperature (*p*=0.02).

# 4.5.7 Conceptual summary figure

Data from January 2013 to December 2014 were summarised in a conceptual summary figure to allow visualisation of a side-by-side comparison of the grassland and SRC willow

(Fig 4.8). This figure allows visualisation of the movement of carbon through the ecosystem. The harvested carbon is shown, 344.0 g C m<sup>-2</sup> y<sup>-1</sup> and 405.8 g C m<sup>-2</sup> y<sup>-1</sup> for grassland and SRC willow, respectively. However, as the mowed grass was not removed from the site, NBP is equal to NEE. Thus, mean NBP (2013-2014) was -118.9 g C m<sup>-2</sup> y<sup>-1</sup> for grassland and 214.2 g C m<sup>-2</sup> y<sup>-1</sup> for SRC willow which despite the removal of 405.8 g C m<sup>-2</sup> y<sup>-1</sup> biomass from the SRC field, remained a net sink for carbon.

## 4.5.8 Impact of harvest in SRC willow

The SRC willow was harvested in April 2014 which corresponded to year 5 of the first rotation. There was no detectable effect of the harvest on soil moisture or soil temperature in the SRC willow, compared to pre-harvest measurements. The effect of the harvest on the NEE can be seen in Fig 4.7b (dashed arrow indicated harvest date), where NEE is decreasing into the growing season then the harvest occurs and the NEE quickly increases. The smaller NEE and increased  $R_{eco}$  observed in SRC willow in 2014 compared to 2013 is likely attributable to the disturbance caused by the harvest. The site quickly became a net C sink again as there was a rapid re-sprout of willow stumps and understory vegetation. There was no noticeable effect on soil CO<sub>2</sub> and CH<sub>4</sub> emissions as a result of the harvest, which may be linked to the harvesting process (Fig C.2b).



**Figure 4.8:** Annual GHG budget for grassland and SRC willow for measurement period January 2013 to December 2014. All fluxes are in gC m<sup>-2</sup> y<sup>-1</sup> in square boxes and soil storage terms presented as standing stock (g m<sup>-2</sup>) in oval boxes. Measured values are presented as mean  $\pm$  standard error. Note: for all fluxes, apart from NBP, a negative flux indicates a gain to the ecosystem and a positive flux indicates a loss to the atmosphere.

# 4.6 Discussion

This research has demonstrated that over a two year period (including the harvest operation in SRC willow), during a side-by-side commercial-scale comparison, SRC willow was a net sink for carbon, whilst the minimally managed grassland, was a net source for carbon. We have also shown that there was lower total soil respiration in SRC willow compared to grassland. N<sub>2</sub>O and CH<sub>4</sub> emissions were generally low for both sites, contributing little to the total GHG balance for these contrasting land-use types in southern England. Thus we can conclude that seven years post land use transition and this site and pending further replication, SRC willow, had an improved GHG balance relative to the adjacent grassland. This suggests that not only did this LUC provide bioenergy as a net provisioning ecosystem service, but was also able to provide improved climate regulation through the generation of a net carbon sink relative to the original land use. In the controversial area of bioenergy science, this is an important empirical finding and suggests that in temperate climates, where reasonable land use transitions are considered, bioenergy may add positively to the multi-functional landscape, as suggested recently by those such as Manning *et al.* (2015) and Souza *et al.* (2015).

Improved grasslands are important sources of terrestrial carbon storage, holding the second largest store after bogs, with approximately 274 x  $10^6$  t C (Ostle *et al.*, 2009). In a UK context, conversion of semi-permanent, permanent or managed grassland to bioenergy cropping systems probably represents one of the most controversial land-use transitions, since grassland is also a significant part of the UK landscape (4-5 x  $10^6$  ha; DEFRA, 2007) and because management of grasslands can vary widely in the UK, particularly with respect to fertiliser input and grazing. This can have a dramatic effect on consequential GHG and carbon balance as a result of LUC. To our knowledge there has been only one previous limited study of eddy covariance measurements over SRC willow for bioenergy (Drewer *et al.*, 2012), our study being the first to have a paired site comparison of SRC willow and grassland. There have been studies which have used the eddy covariance technique on SRC poplar. These studies have observed SRC poplar to be a net sink of carbon at the ecosystem level (Arevalo *et al.*, 2011; Jassal *et al.*, 2013).

Retention of cut grass on the surface, which results in no C exports from the system, is fairly uncommon with the most common practice to have the land grazed or mowed for hay or silage (Smit *et al.*, 2008). Qun & Huizhi (2015) investigated similarly managed

grassland where there were no exports of carbon and found that the site was a net source of carbon, with a NBP of -138.4 g C m<sup>-2</sup> y<sup>-1</sup>. This is less than half that of our grassland which is likely due to reduced rainfall and lower biomass observed in the Qun & Huizhi study (2015). We identified PAR and soil moisture to be the main climatic drivers of NEE in grassland and SRC willow, which has been found in other studies (Ruimy *et al.*, 1995; Qun & Huizhi, 2015; Shao *et al.*, 2015). Others have identified leaf area index (LAI) to be the main biophysical driver of NEE in SRC poplar (Broeckx *et al.*, 2014; Zenone *et al.*, 2015). Data syntheses from a network of sites such as FLUXNET have already begun identifying driving factors of NEE, GPP and R<sub>eco</sub> over a number of biomes (Law *et al.*, 2002), and as the amount of flux data from bioenergy crops increases there is potential for syntheses in these biomes in future.

Grasslands have been shown to be both temporally variable (Soussana *et al.*, 2007) and spatially variable (Imer *et al.*, 2013), for GHG emissions and carbon balance. Evidence of this variability can be seen in this study, where we observed grassland to be a net source of carbon in 2013 and a net sink in 2014. This difference could be attributable to the higher temperatures observed in January-March 2014 compared to 2013. Grass has been shown to begin growth when air temperature exceeds  $5^{\circ}$ C (Robson *et al.*, 1988), which was achieved in early 2014. This combination of increased temperature with an increase in winter rainfall (which resulted in increased soil moisture) could explain the higher aboveground biomass in grassland and consequently why the site was a net sink in 2014 (Pitt & Heady, 1978).

As well as large variability, there are also large uncertainties surrounding the carbon balance of temperate grasslands (Janssens *et al.*, 2003). Within the literature there are reports that grasslands are acting as both carbon sources and carbon sinks (Scurlock & Hall; 1998; Bellamy *et al.*, 2005; Soussana *et al.*, 2007; Ciais *et al.*, 2010; Merbold *et al.*, 2014; Schipper *et al.*, 2014; Rutledge *et al.*, 2015). The carbon balance of grasslands is quite often dictated by favourable management regimes (Smith, 2014) however changes in management can cause grasslands to switch from a source to a sink (Merbold *et al.*, 2014). Grassland management practices such as fertilisation, grazing and mowing lead to large N<sub>2</sub>O and CH<sub>4</sub> emissions which counterbalance this CO<sub>2</sub> sink (Ciais *et al.*, 2010; Imer *et al.*, 2013). For our study, N<sub>2</sub>O and CH<sub>4</sub> did not make up a significant part of the whole GHG balance of either land uses and both were present in small quantities. SRC willow has been found to be a net sink for CH<sub>4</sub> in other studies to a similar extent to that found here (Kern *et al.*, 2012; Drewer *et al.*, 2012). For both sites, there was an effect of soil moisture, and a significant interaction for soil moisture and soil temperature on  $CH_4$  fluxes. Other studies have confirmed the influence of soil moisture on  $CH_4$  emissions under bioenergy crops (Kern *et al.*, 2012; Drewer *et al.*, 2012) and grasslands (Kammann *et al.*, 2001; Imer *et al.*, 2013).

Here we found that grassland had significantly higher soil carbon stocks than the SRC willow to 30 cm but examination to 1 m depth revealed no significant difference in soil C stock between land use types. Sampling depth is a recurrent problem in studies which attempt to quantify soil carbon (Dolan *et al.*, 2006; Blanco-Canqui & Lal, 2008) and it is essential that the whole profile is sampled to draw robust conclusions (Harrison *et al.*, 2011). We were able to sample two grassland sites and whilst they were in agreement for the top 30 cm, the data down to 100 cm showed higher stocks for one grassland (compared to SRC willow) and lower carbon stocks for the other; though neither was significantly different from the SRC willow. This demonstrates the large variability that exists amongst grasslands, even those in a similar location, on the same soil type and under similar management. A larger sample size would be needed to demonstrate if there is a difference in soil C to 100 cm at these sites, as a true difference may not have been detected in this case due to a low sample size.

At our site, the higher soil C observed in grassland may be attributable to the amount of organic material left on the soil surface after mowing (Post & Kwon, 2000). There have been reports in the literature of both increased soil carbon under SRC compared to grassland (Arevalo et al., 2009; Zan et al., 2001), as well as others which have found no significant difference (Grigal & Berguson, 1998; Walter et al., 2015). Walter et al. 2015, from a chronosequence of SRC sites, suggested this transition results in a redistribution of carbon through the profile, despite total SOC stock not being significantly different. After seven years post-conversion, we may be beginning to see redistribution of C in the soil profile. We found that at the two grasslands sites 59% and 96% carbon was stored in the top 30 cm, whereas in SRC willow 54% carbon was stored in the top 30 cm of the whole 100 cm profile. Whilst these differences are not large, it may be the early stages of C redistribution through the soil profile. Chronosequence data also suggest that after initial conversion from grassland to SRC willow, there can be a loss of soil carbon for up to 5 years, which is followed by recovery up to 19 years (Pacaldo et al., 2013b). Our site is only seven years post-conversion, therefore is likely still in the recovery phase with respect to soil carbon.

In this study we were able to capture the effects of a commercial harvest on the soil and ecosystem GHG balance. This is an essential management practice which takes place every three to four years in SRC willow and exerts major changes on the plantation; changes about which little is known for GHG balance in SRC willow (Vanbevern et al., 2015). Harvesting is recognised as one of the most energy intensive stages of the SRC willow life cycle due to the large consumption of diesel fuel (Murphy et al., 2014). From our study we have shown that whilst there is an increase in Reco after the harvest, the site is able to quickly become a large carbon sink as soon as three weeks after harvest. Other LCA findings have shown that whilst the harvest can increase emissions due to the harvest machinery, the carbon sink created by SRC willow is able to offset these emissions and result in a negative GHG balance (reported in the range of -138.4 to -52.9 kg CO<sub>2</sub>-eq. per odt biomass; Caputo et al., 2014). We also observed a one off peak in N<sub>2</sub>O emissions, two months post-harvest, which was the largest emission observed across both sites for the duration of the experiment. Other studies have observed little effect of harvest on N<sub>2</sub>O emissions from SRC cultures (Zona et al., 2013b). It is possible that this emission arose as a result of increased soil exposure after harvest and increased rainfall in May and June 2014, relative to 2013. Soil N<sub>2</sub>O fluxes are known to vary spatially and temporally and to arise quickly after changes in rainfall, temperature and management (Skiba & Smith, 2000). N<sub>2</sub>O emissions, therefore, require more intense monitoring to be able to capture these emissions, as one large emission can account for a large proportion of total N<sub>2</sub>O fluxes over a measurement period (Zona et al., 2013b).

To conclude, we have found that LUC to SRC willow from grassland can result in reduced GHG emissions. At our site we found that grassland was a net carbon source and SRC willow was a net carbon sink seven years after land conversion. However, soil carbon stocks were likely still in recovery as soil C at the SRC site remained significantly lower than grassland, even after this time since establishment. Whilst grasslands have been shown to be highly variable, there is evidence that this LUC may result in climate mitigation advantages. As our sites were both low input and were not fertilised,  $N_2O$  and  $CH_4$  emissions did not make a significant contribution to the whole GHG balance. Additional research is however, required on the effects of LUC from grasslands which consider a range of management options, particularly with respect to fertiliser and rotation length, so that wide-ranging conclusions can be made on this important land use type.

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## CHAPTER 5: INFLUENCE OF LAND USE, LITTER FALL AND LITTER DECOMPOSITION ON SOIL CHEMISTRY

**Statement of contribution:** I performed all data collection, CEH undertook analysis of soil samples, whilst I completed data analysis and wrote the chapter. Willow genotyping by SSR was kindly undertaken by Dr Steve Hanley at Rothamsted Research (see Appendix E for SRC willow genotyping).

## 5.1 Abstract

Land use change from grassland to SRC willow results in a change to vegetation and a change in the type of organic inputs which will likely result in changes belowground. The aim of this chapter is to assess the effect litter fall and decomposition on soil chemistry.

Litter fall and litter decomposition was measured for the duration of the experiment in SRC willow using litter traps and mesh decomposition bags. The rate of litter fall and litter decomposition can inform us about the movement of carbon from aboveground pools to belowground pools. SRC willow produced on average 5.5 t ha<sup>-1</sup> y<sup>-1</sup> of litter for the 4/5 year of growth and 2.1 t ha<sup>-1</sup> y<sup>-1</sup> litter for the first year post harvest. This is higher than other values found in the literature indicating high productivity at this site. Litter decomposition was found to be quite rapid, with over 80% litter decomposed within the first year. This was similar to what has been found in the literature.

Soil cores were taken throughout the experiment in both grassland and SRC willow and were analysed for soil chemistry. Analysis of soil chemistry revealed there was an increase in bulk density, a reduction in soil water content and a decrease in dissolved organic carbon as a result of land use change from grassland to SRC willow. There were more nitrate in grassland and more ammonium in SRC willow. There was no significant difference in water filled pore space or C:N ratio between sites. The change in bulk density did not appear to have an effect on SRC willow rooting behaviour as there was high productivity of the crop. A reduction in soil water is a positive change as the grassland was found to be a sink for CH<sub>4</sub>. A decrease in soil carbon content is a negative effect of LUC to SRC willow. The literature indicates a loss in SOC can be observed for up to 5 years post conversion, followed by recovery up to 19 years after conversion. It has been 7 years since the site was converted, indicating SOC in SRC willow may still be in the recovery phase.

## **5.2 Introduction**

The soil is an essential medium for all life on earth allowing the cultivation of primary producers and playing a vital role in biogeochemical cycling. The soil is a complex medium which mediates feedbacks between aboveground and belowground productivity.

Soil quality is defined as the ability of a soil to sustain environmental quality, foster biological productivity, and stimulate plant and animal health (Weinhold et al., 2005). The quality of the soil will be dependent on the intended use of the field and the expected production from land owners. There are several types of assessments of soil health through assessing both physical soil characteristics and chemical characteristics (Table 5.1; Larson & Peirce, 1994; Schoenholtz et al., 2000; Wienhold et al., 2005). The idealised soil composition is predominantly made up of solid matter in the form of minerals (45%) and organic matter (5%), with the remainder made up of water (25%) and air (25%). The amount of solid matter per volume of total soil is referred to as *bulk density*. A lower bulk density (BD) is preferable for root growth; when BD exceeds a certain value root activity is restricted; this upper bound of BD differs depending on soil type. The spaces between the solid matter (or aggregates) is known as pore space; the amount of pores within the solid medium is measured as *soil porosity*. We can estimate the amount of water present within the soil medium by measuring the amount of pore space saturated with water, or the water-filled pore space (WFPS). For WFPS there is an optimal range which is suitable for microbial activity at approximately 60% WFPS, which corresponds to a moist, wellaerated soil (Fig 5.5; Wienhold et al., 2005; Linn & Doran, 1984). At 80% WFPS anaerobic conditions dominate and result in processes such as denitrification. Soil water content, measured here as gravimetric moisture, indicates how much water is held in the soil medium. Soil moisture affects movement of gases through the soil and can dictate if soil is an aerobic or anaerobic environment. These physical measures allow an understanding of the soil composition and arrangement which give an indication of the ability of the soil to support root growth, host soil fauna and determine decomposition rates of organic material.

Chemical properties which are important indicators for soil health, are soil organic carbon status, nutrient availability, soil acidity and salinity (Schoenholtz *et al.*, 2000). Soil organic carbon has been found to influence all aspects of soil health as it is the primary source of energy for soil organisms and affected nutrient availability though mineralization.

**Table 5.1:** Potential soil properties for assessing soil quality and function. Taken from Weinhold *et al.* (2005).

Soil function	Physical Properties	Chemical Properties	Biological Properties
Substrate for plant	Bulk density	Electrical conductivity	
growth	Aggregate stability	Sodium adsorption	
	Soil depth		
Nutrient reservoir		Inorganic N content	Potential
		Extractable P	mineralization
		рН	
Atmospheric interactions	Water-filled pore space	CO <sub>2</sub> flux	
		NO <sub>x</sub> flux	
		Methane flux	
		NH <sub>4</sub> flux	
Storage and purification	Infiltration rate	NO <sub>3</sub> -N concentration	
	Water-holding capacity	Extractable P	
Biological activity	Water-filled pore		Microbial biomass
	space		Soil respiration
			Potential mineralization
			Earthworm population

Nutrient availability again is essential for biogeochemical cycling as well as plant productivity. Nitrogen is the main limiting nutrient in terrestrial ecosystems. Nitrogen in the soil is utilised by both the plant, taken up through the roots, and by soil microbes, metabolised as an energy source. There are 5 main pathways by which nitrogen is transformed in the soil, detailed in Table 5.2. The main forms by which nitrogen is utilised by plants and microbes is as nitrate ( $NO_3^-$ ) and ammonium ( $NH_4^+$ ). Plants preferentially take up N in the form of nitrate, though are able to take up ammonium; conversely microbes preferentially assimilate ammonium over nitrate or nitrite ( $NO_2^-$ ). If conditions in the soil are predominantly aerobic, nitrate will be the dominant form of nitrogen in the soil. If oxygen is limited, therefore creating anaerobic conditions, nitrate will be reduced and ammonium will be produced which may then lead to production of N<sub>2</sub>O and N<sub>2</sub> gases. The nitrate ion is extremely soluble; therefore excessive nutrients (which often arise as a result of fertilisation) can lead to contamination of surface and ground water as a result of runoff or leaching.



**Figure 5.1:** Hypothetical relationship between water filled pore space and microbial activities. Taken from Wienhold *et al.* (2005), originally from Linn & Doran, 1984.

Process	Mode of action	Equation
Mineralisation	(1) Organic N is hydrolysed to release ammonia $(NH_3)$ which is then protonated to	(1) Organic $N(R - NH_2) \rightarrow NH_3 \rightarrow NH_4^+$
(ammonification)	become ammonium (NH4 <sup>+</sup> )	
	(2) Urea is rapidly hydrolysed by the enzyme urease to produce carbon dioxide	(2) $H_{2}O + NH_{2}CONH_{2} \xrightarrow{Urease} CO_{2} + 2NH_{2}$
	and ammonia	
Assimilation	The incorporation of inorganic N as $NH_4^+$ , $NO_3^-$ or $NO_2^-$ into biomass	$NH_4^+ \rightarrow Organic N (R - NH_2) \text{ OR}$
(immobilisation)		$NO_3^- \text{ or } NO_2^- \rightarrow NH_4^+ \rightarrow Organic N (R - NH_2)$
Nitrification	The oxidation of reduced inorganic and organic N to $NO_2^-$ or $NO_3^-$ via an	(1) Simplified autotrophic nitrification:
	autotrophic or heterotrophic pathway.	$NH_{4}^{+} \leftrightarrow NH_{2} \xrightarrow{Nitrosomonas} NO_{2}^{-} \xrightarrow{Nitrobacter} NO_{3}^{-}$
	(1) Autotrophic nitrification is a two-step process which carried out by	+ H <sup>+</sup> 5 2 5
	chemolithotrophic bacteria, examples given in equation.	(2) Simplified betarotrophic pitrification:
	(2) Heterotrophic nitrification is carried out by chemoheterotrophic bacteria and	(2) Simplified Helefoldophic minimulation. $Organic N(P - NH) \rightarrow NO^{-} or NO^{-}$
	fungi (e.g. Arthrobacter, Streptomyces and Aspergillus), usually in soils too acidic	$Organic N(K - NH_2) \rightarrow NO_2 OrNO_3$
	for autotrophic nitrification	
Nitrate reduction	Reduction of nitrate occurs in two ways, predominately via denitrification and	(1) $NO_3^- \xrightarrow{Nitrate \ reductase} NO_2^- \xrightarrow{Nitrite \ reductase} NO$
	dissimilatory nitrate reduction to ammonium (DNRA). Both these processes occur	Nitric oxide reductase Nitrous oxide reductise
	primarily under waterlogged and anaerobic conditions	$ N_2 U  N_2$
	(1) Denitrification is a multistep process by which nitrate are reduced to nitrogen	(2) $NO^{-} \rightarrow NO^{-} \rightarrow NU^{+}$
	gas $(N_2)$ by a series of enzymes.	$(2) NO_3 \rightarrow NO_2 \rightarrow N\Pi_4$
	(2) DNRA is the reduction of nitrate to ammonium $(NH_4^*)$	
N <sub>2</sub> fixation	Process by which nitrogen gas $(N_2)$ is energetically reduced to ammonia $(NH_3)$ by	$N_2 + 16Ma - ATP + 8H^+ \xrightarrow{Nitrogenase} 2NH_2$
	prokaryotic bacteria, either independently or in symbiosis with leguminous or	
	nonleguminous plant species.	$+ n_2 + 10ADP + 10Pl$
		$+ 16Mg^{2+}$

 Table 5.2: Process by which nitrogen is transformed in the soil. Adapted from Coyne & Frye (2005).

Soil acidity, measured as soil pH, affects both plant growth and microbial community composition and activity. Soil salinity is a measure of salts present in the soil solution, high levels of which have negative impacts on plant growth and can lead to reductions in water quality and soil erosion.

Finally, there are soil biological properties which affect the soils ability to decompose organic material and recycle nutrients from aboveground biomass to the belowground community. This is mainly assessment of the living microbial community in the soil, such as microbial decomposers and earthworms, which has been measured on site as part of the EUROCHAR project (Jenkins, unpublished).

The ratio of C:N in the soil is another important indicator of ecosystem function as it can indicate the rate of decomposition and quality of SOM. The C:N ratio of the soil is determined by the input of litter from vegetation. A high quality litter is one that has a low C:N ratio as it is most rapidly decomposed into SOM by microorganisms requiring nitrogen (Jahren, 2013). However, there are varying nitrogen requirements within the microorganism community. Fungi have a lower N requirement than bacteria, therefore it has been surmised that the ideal C:N ratio is 24:1 as this meets the requirements of all microorganisms. Above this ratio, net immobilisation occurs and additional N is required for decomposition to occur (Hodge, 2005).

Leaf litter fall and decomposition are the primary means by which carbon and nutrients are recycled from aboveground biomass back into the soil. It is estimated that over 50% of net primary productivity (NPP) is returned to the soil via decomposition of leaf material (Wardle et al., 2004). Quantification of leaf litter mass allows an understanding of what proportion of total aboveground biomass is allocated to the leaves, and consequently how much litter will fall to the ground to allow return of carbon and nutrients to the soil. It is understood that there are four main factors affecting leaf litter decomposition: climate, litter quality, soil quality and the composition and activity of soil communities (Wardle et al., 2004; Berg & McClaugherty, 2008). The ability of SRC willow to effectively recycle nutrients through leaf biomass is documented within the literature (Ericsson, 1994) and is one of the reasons why these plantations do not require nutrient supplementation in the form of fertiliser. Leaf litter inputs are essential for the formation of soil organic matter (SOM) and accumulation of soil organic carbon (SOC) in the top soils (Tolbert, 2002; Rytter, 2012; Hangs et al., 2014). Raich and Nadelhoffer (1989), and more recently confirmed by Davison et al., (2002), showed that total belowground carbon is at least double that of aboveground litter fall in mature forests demonstrating the importance of leaf litter in linking the aboveground and belowground carbon (Waddle *et al.*, 2004). Accurate quantification of litter fall in SRC willow plantations would improve carbon balance estimates of these bioenergy crops, as it is recognised as a known source of uncertainty in LCA calculations (Caputo *et al.*, 2014).

Plant species composition, management and land use change (LUC) can have marked effects on all aspects of soil structure, microbial community composition and chemistry (Post & Kwon, 2000). In this section I will measure some of the basic properties of the soil under both land uses and the influence of litter fall and decomposition in SRC willow.

## 5.3 Methods

#### 5.3.1 Leaf Litter Fall

Plastic trays measuring 35x45 cm were deployed into the field in June of each year and secured with pegs into the ground. Drainage holes were drilled into the trays prior to deployment to allow drainage of rain and escape of any small animals. Leaf litter was collected every 2-4 weeks and placed into labelled bags to be transported back to the lab. Leaf litter was collected until the trees were bare of all leaves and no more litter was accumulating in the trays. Leaf litter was oven dried at 80°C for 48 hours. Leaf litter was weighed on a balance and recorded. Total leaf litter from a collection point was summed and a cumulative total calculated. Cumulative total was plotted against week of the year to see if the timing of leaf fall differed between years. Mean litter fall per square hectare was calculated by taking an average of the total litter fall from all plots and dividing by the size of the collection tray.

#### 5.3.2 Leaf litter decomposition

Willow leaf litter was collected in autumn 2011 when litter had already begun to fall naturally from the trees. Leaves were removed from the stems of the trees by running hands up the stems and collecting any litter that came away easily. This was placed into plastic bags and transported back to the lab. All litter was air dried in aerated crates, tossed by hand twice a day, until fully dry. Exactly 5 g of dried leaf litter was then placed into pre-made mesh bags. Mesh bags were 10x20 cm; heat sealed to contain the litter then each bag was labelled using a plant label tagged to one corner. Mesh size was 1 mm aperture and was sufficient to allow access by macrofauna, as well as microfauna. A total of 72 bags were made to be placed at each experimental plot throughout the field, with 9 bags at each location. Bags were placed either within the row (in the middle of the double row of planted trees) or between the rows (between each pair of planted rows), as per the location of the chamber (Figure 4.1). Bags were placed on the soil surface in the willow field and kept in place with potting labels pushed into the ground. Litter decomposition bags were collected at several time points post insertion - 2 weeks then 1, 2, 4, 6, 9, 12, 18 and 24 months. At collection, litter bags were carefully taken from the ground and any visible mud, root or other debris was carefully removed before placing into a small labelled plastic ziplock bag and transported back to the University of Southampton.

Once back at the lab, the litter bags were inspected for any noticeable damage, for example large holes due to animal chewing, and any debris, such as mud/roots carefully removed from the outside of the bag. The litter bags were cut open along 3 sides and the leaf litter was washed with water to remove any mud or other debris, so only plant leaf material was collected. Tweezers were used to handle leaf litter and any small fragments were all collected and placed into a labelled paper bag. The leaf litter was then oven dried for 24 hours at 85°C, weighed, and weight recorded. This was repeated for each litter bag.

All litter bag removals were made on schedule apart from the 18 and 24 month collections as the field was harvested in April 2014 meaning the litter bags had to be removed from the field. The litter bags were out of the field for 43 days, from 18/03/14 to 30/04/14. During this time they were carefully removed from the ground, placed in a plastic zip-lock bag and transported back to the lab. Litter bags were stored at -20°C to ensure there was no further decay or decomposition during the time out of the field. Litter bags were replaced in as close as a position from which they were removed and the dates of collection were adjusted to account for the time out of the field, i.e. shifted back 43 days.

To calculate the average rate of litter loss of each sampling time point, all values were summed and divided by the total, n=8. Percentage loss over time was plotted to assess the rate of loss over time.

#### 5.3.3 Soil chemistry

#### 5.3.3.1 Taking soil cores in the field

Soil cores were taken every quarter of the year in November, February, May and August, starting in November 2011 (when the static chambers were first installed) until November 2014. Samples were taken on the same day as the soil GHG measurements to allow any link between soil chemistry and GHG flux to be identified. Both the grassland and SRC willow sites were sampled where 4 out of the 8 chamber locations were randomly chosen to be sampled. At each sampling event, plastic pipes of 15 cm depth and 5 cm diameter were hammered into the soil using a rubber mallet and removed using pliers. Soil samples were left in the pipes, placed into a labelled plastic bag and placed immediately in a cooler to reduce any effect on soil chemistry of the sample. Samples were transported back to the lab and stored at 4°C until processed.

#### 5.3.3.2 Processing soil cores in the lab

Soil cores were removed from their bags, weighed and depth of soil in the core recorded. Soil was removed from the core and cut longitudinally into 4 sections, each section was individually weighed. Each section was then sieved to remove any roots or stones and stored in a labelled plastic bag until ready for processing. Three of the four sections were used for extractions and the last section was split between two 50 ml labelled falcon tubes and stored at -80°C as a spare.

#### 5.3.3.3 Soil Physical Properties

For determination of gravimetric moisture, labelled crucibles were placed into an oven at 105°C for at least 1 hour prior to use to remove any moisture within the crucible. Once removed from the oven, crucibles were placed into a desiccator to cool. Cooled crucibles were weighed and 10 g of fresh soil was added, the soil and crucible were then weighed. Crucibles were placed in the oven at 105°C for 24 hours, cooled in a desiccator then reweighed.

The following calculation was used to estimate gravimetric moisture (GM):

Gravimetric Moisture (%) = 
$$\frac{Mass loss (g)}{Mass of oven dried soil (g)} \times 100$$
(5.1)

where:

$$Mass \ loss \ (g) = Wet \ soil \ \& \ crucible - Dry \ soil \ \& \ crucible$$
(5.2)

and:

Mass of oven dried soil 
$$(g) = dry$$
 soil & crucible weight – crucible weight

(5.3)

The bulk density (BD) of the soil can be calculated using the following equation:

$$Bulk \ Density \ (g \ cm^3) = \frac{Estimated \ dry \ mass \ of \ core}{Volume \ of \ core}$$
(5.4)

where:

Estimated dry core mass = soil core weight 
$$x \left(\frac{Xg \, dry \, soil}{10}\right)$$
(5.5)

and :

*Volume of core* = 
$$\pi r^2 d$$

(5.6)

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Total porosity (TP) can be calculated as below, using bulk density from equation 5.6 and a particle density of 2.65  $Mg/m^3$  (for most mineral soils; Freeze and Cherry, 1979):

$$Total Porosity (\%) = \left(1 - \left(\frac{Bulk Density}{Particle Density}\right)\right) \times 100$$
(5.7)

Water-filled pore space (WFPS) can be calculated as per below using gravimetric moisture from equation 5.3, bulk density from equation 5.6 and total porosity using equation 5.9.

$$WFPS(\%) = Grav Moisture \times \frac{Bulk Density}{Total Porosity} \times 100$$
(5.8)

#### 5.3.3.4 Estimation of Soil Dissolved Organic Carbon (DOC)

After sieving, 10 g of soil were mixed with 70 ml deionised water for 10 minutes on an orbital shaker. The homogenised mixture was filtered through Whatman No.1 filter paper; this step sometimes took several hours, so filtration was left in a 4°C cold room overnight. Samples were vacuum filtered through cellulose filter paper, collected into two 50 ml falcon tubes and stored at -20°C. The analysis of the extracts took place at CEH Lancaster due to a desire for consistency among sample analysis for all partners in the ELUM consortium.

#### 5.3.3.5 Estimation of Soil Ammonium (NH<sub>4</sub><sup>+</sup>) and Nitrate (NO<sub>3</sub><sup>-</sup>)

After sieving, 10 g of soil were mixed with 100 ml of 6% analytical grade potassium chloride (KCl) and left to stand for 15 minutes with occasional stirring. The mixture was filtered through Whatman No. 44 filter paper and the first few millilitres rejected. Filtered extract was collected into two 50 ml falcon tubes and stored at -20°C. As with the DOC extraction (section 5.4.2.4), all samples were processed by CEH Lancaster.

#### 5.3.4.6 Estimation of Soil Carbon and Nitrogen Content

After sieving, soil samples were placed at -80°C for a minimum of 24 hours before being freeze dried for a further 24 hours. Freeze dried soil was ball milled for 1 minute at 300 RPM to grind the soil into a fine dust in preparation for analysis. Soil samples were analysed using a LECO TruSpec C/N Analyser. Ten blanks were run prior to loading any

samples; while this was completed the samples were prepared for analysis. Soil was weighed into foil cups to approximately 0.2 g on scales and the weight sent to the analyser software. The foil cup, containing soil, was shaped into a ball and placed into the loading dock of the analyser. This was repeated for all samples, including a standard which was run before the samples, and after every 20 samples to account for any machine drift. All samples were drift corrected according to the standards. Samples were automatically loaded into the analyser where they were combusted in a furnace operating at 950°C. As a result of dry combustion, carbon was estimated via infrared detection and nitrogen through thermal conductivity detection. Data were outputted as % of C and N, downloaded into a USB and analysed in Microsoft Excel. The C:N ratio was calculated by dividing %C by %N.

#### 5.3.4 Statistical Analysis

All statistical analyses were conducted in the R programming environment (R version 3.1.3). Data were analysed using linear mixed models (Pinheiro *et al.*, 2015) where chamber, nested within row type, was used as a random factor in all models to account for repeat sampling over time. There were 8 chambers in total, 4 of which were placed within a row and 4 placed outside a row.

Differences in total annual litter fall data were tested using a one-way ANOVA with posthoc Tukey testing to confirm differences between years. For litter fall and litter decomposition in SRC willow, a random effects model was constructed to test for the effects of year, air temperature and genotype on litter production (eq 5.9 and 5.10). Air temperature was found to correlate with soil temperature and soil moisture, therefore due to collinearity, only air temperature is included in the model.

Litter fall ~ year + air\_temp + genotype, random= ~ 1 | row\_type/chamber 
$$(5.9)$$

Decomp ~ air\_temp+genotype, random = ~ 
$$1 | row_type/chamber$$
 (5.10)

For soil properties, a global model was used to test for effects of treatment on various measures of interest, then individual models specific to each site dataset were used to test for effects of other variables. Analysis of variance was used to determine effect of each variable in the model on the response variable.

For soil properties including, bulk density, total porosity, gravimetric moisture and water filled pore space there was collinearity detected between air temperature, soil temperature and soil moisture. The following model was constructed:

Soil property ~ site + month + year+ air.temp, random= ~ 
$$1 \mid cha$$
 (5.11)

Gravimetric moisture could not be tested for an effect of air temperature, soil temperature or soil moisture, as gravimetric moisture is a measure of soil water content as is soil moisture.

For dissolved organic carbon, ammonium and nitrate content there was collinearity detected between air temperature, soil temperature and soil moisture; BD and TP; and GM and WFPS. This means that only one of these terms can be included in the model; if one of these terms are found to be significant we can assume the variable which collineates is also 170

significant. The following models were constructed where 'chemical property' was DOC,  $NO_3^-$ ,  $NH_4^+$  or  $NO_3^-$ :  $NH_4^+$  ratio.

Global model: Chemical property ~ Treatment+Year+Month+air.temp+BD+WFPS, random=~1|Chamber (5.12) <u>Site specific model:</u> Chemical property ~ Year+Month+air.temp+BD+WFPS, random=~1|Chamber (5.13) For C:N ratio, the following models were constructed: <u>Global model:</u>

C:N ratio ~ Treatment+Year+Month+air.temp+BD, random=~1|Chamber

(5.14)

Site specific model:

C:N ~ Year+Month+air.temp+BD, random=~1|Chamber

(5.14)

The significance of the model terms were assessed using a Likelihood Ratio test. Post-hoc Tukey tests were used after ANOVA to confirm how yearly and monthly means differed in models where they were found to be statistically significant (Hothorn *et al.*, 2008).

Effects displays were constructed for all models (Fox & Hong, 2003) which can be found in Appendix D.

## 5.4 Results

#### 5.4.1 Leaf litter fall

The timing of leaf litter fall was similar for 2012 and 2013 (Figure 5.2). The first collection was made in early July (week 27/28) and the final collection in December (week 48-50). However for 2014, which was the year of the harvest, litter fall started much later in the year, in early September (week 36) and required less frequent collections than 2012/2013.



**Figure 5.2:** Cumulative litter fall for sampling years. Note harvest took place in April (week 17) of 2014.



**Figure 5.3:** Annual litter fall for SRC willow site with SE shown. Note harvest took place in April 2014.

Leaf biomass contributed to 13.8%, 11.3%, 15.7% of total aboveground biomass for 2012, 2013 and 2014, respectively (Figure 4.4). Assuming a carbon content of leaves of 50%, we could estimate that approximately 2.1 t C ha<sup>-1</sup> y<sup>-1</sup> is returned to the soil via leaf litter. Total litter fall was similar for 2012 and 2013 with 5.6 and 5.3 t ha<sup>-1</sup> y<sup>-1</sup> (Fig 5.3). A one-way ANOVA detected a difference in the litter fall between years ( $F_{2,21}$ =76.4, *p*<0.001), with a posthoc Tukey test confirming litter fall was lower in 2014 with 2.1 t ha<sup>-1</sup> y<sup>-1</sup> than in both 2012 and 2013 (*p*<0.001). There was no difference in litter fall between 2012 and 2013 (*p*=0.70). Statistical analyses revealed that there was no effect of year (*p*=0.89), air temperature (and by associated soil temp and spoil moisture; *p*=0.24) or genotype (*p*=0.57) on leaf litter fall.

#### 5.4.2 Leaf litter decomposition

The majority of leaf litter underwent decomposition within the first year, with only 17% leaf litter remaining after 12 months. After 2 years there was approximately 8% of the leaf litter remaining. There was relatively low variation around the mean rate of decomposition across the plots at the site (Fig 5.4). Statistical analyses revealed there was no effect of air temperature, and therefore soil temperature or soil moisture, on the rate of decomposition (p=0.68).



**Figure 5.4:** Willow leaf litter decomposition over time, averaged for 8 plots across the site. Max and Min values show variation across the site.

#### 5.4.3 Soil properties and soil chemistry

The average BD for the three year measurement (2012-2014) period was  $1.2 \pm 0.0$  g cm<sup>3</sup> for grassland and  $1.4 \pm 0.0$  g cm<sup>3</sup> for SRC willow (Table 5.3). BD was higher in SRC willow than in the grassland ( $F_{1.81}=77.94$ , p<0.001), however there was no effect of chamber, month, year or air temperature (soil temperature and soil moisture). GM was higher in grassland at 39.1  $\pm$  1.6% than SRC willow at 26.5  $\pm$  1.1% for the three year measurement period ( $F_{1,81}$ =80.52, p<0.001; Table 5.3), though there was no significant effect of any other tested variables. There was an effect of site on TP, averaging  $53.2 \pm 0.7\%$ in grassland and 44.9  $\pm$  0.6% in SRC willow ( $F_{1.81}$ =77.94, p<0.001; Table 5.3). WFPS was higher in grassland (82.9  $\pm$  2.7%) than in SRC willow though this was not significantly different ( $F_{1.81}$ =3.39, p=0.069; Table 5.3). Air temperature (and by association soil temperature and soil moisture), year and month were found to have a significant effect on WFPS ( $F_{1.81}$ =16.54, p<0.001;  $F_{2.81}$ =8.66, p<0.001;  $F_{3.81}$ =33.03, p<0.001 respectively). A posthoc Tukey test revealed that WFPS was higher in 2014 (p=0.003) and 2012 (p<0.001) than 2013. There was no significant difference in WFPS in 2012 and 2014 (p=0.85). Additionally, WFPS was found to be higher in February and November compared to August (p<0.001 for both comparisons).

Data for monthly, annual and a three year measurement average of DOC,  $NH_4^+$ ,  $NO_3^-$  and the ratio of ammonia to nitrate can be found in Table 5.4. DOC in the top 15 cm of soil was found to be higher in grassland (12.6 ± 1.3 mg/kg) than in SRC willow (9.3 ± 0.6 mg/kg;

F<sub>1,80</sub>=5.63, *p*=0.02; Figure 5.5, Table 5.4). In grassland there was no significant effect of any of the tested variables on DOC content; however in SRC willow year had a significant effect on DOC content. DOC content in 2014 was higher than 2012 or 2013 (F<sub>2,32</sub>=22.61, p<0.0001; Figure 5.6). NH<sub>4</sub><sup>+</sup> content was higher in grassland than in SRC willow (F<sub>1,78</sub> = 6.32, *p*=0.01; Figure 5.7, Table 5.4). NH<sub>4</sub><sup>+</sup> was lower in both fields in 2012 compared to 2013 and 2014 (F<sub>2,32</sub> = 5.11, *p*=0.011 for grassland and SRC willow; Table 5.4). NO<sub>3</sub><sup>-</sup> were higher in SRC willow (0.65 ± 2.03 mg/kg) than in grassland (0.34 ± 2.53 mg/kg; F<sub>1,79</sub> = 8.03, *p*=0.006). NO<sub>3</sub><sup>-</sup> were also lower in 2012 than 2014 in grassland (post-hoc Tukey test *p*=0.036) whereas NO<sub>3</sub><sup>-</sup> were lower in 2012 compared to both 2013 and 2014 in SRC willow (F<sub>2,32</sub> = 10.30, *p*=0.0004; post-hoc Tukey test: 2012-2013 *p*<0.01, 2012-2014 *p*=0.031; Table 5.4). The ratio of NO<sub>3</sub><sup>-</sup> : NH<sub>4</sub><sup>+</sup> is different between grassland and willow (F<sub>1,79</sub> = 9.36, *p*=0.003). Grassland has more NO<sub>3</sub><sup>-</sup> than NH<sub>4</sub><sup>+</sup> with a ratio of 1:0.86 whereas SRC willow had more NH<sub>4</sub><sup>+</sup> than NO<sub>3</sub><sup>-</sup> with a ratio of 1:1.84.

The C:N ratio was not significantly different between land uses, with 10.0:1 in grassland and 10.3:1 in SRC willow ( $F_{1,80} = 1.42$ , p=0.24). However there were differences within each site. For grassland, year and month were found to be significant variable affecting C:N ratio ( $F_{2,33} = 5.10$ , p=0.011 and  $F_{3,33} = 4.25$ , p=0.012 respectively). A post-hoc Tukey test revealed that C:N ratio, in grassland, was higher in 2014 than in 2012 (p=0.007) and C:N was lower in May than November across all measurement years (p=0.027). Air temperature (and by extension soil temperature and soil moisture) and bulk density (and by extension total porosity) were found to have a significant effect on C:N ratio in SRC willow ( $F_{1,32} = 4.84$ , p=0.035 and  $F_{1,32} = 16.36$ , p=0.0003 respectively).

**Table 5.3:** Physical soil properties under grassland and SRC willow showing mean  $\pm$  standard error. Annual average (*n*=16), monthly average for each year (*n*=4), and a three-year average (*n*=48) are shown.

Grassland	Bulk Density (a cm <sup>3</sup> )	Gravimetric Moisture (%)	Total Porosity (%)	Water-filled
2012	1.2 ± 0.0	39.3 ± 2.0	53.3 ± 1.1	86.7 ± 3.0
February	1.3 ± 0.0	37.9 ± 0.3	53.0 ± 2.2	91.4 ± 1.7
May	1.2 ± 0.1	41.7 ± 4.6	57.0 ± 1.3	92.1 ± 4.3
August	1.2 ± 0.1	30.4 ± 2.9	51.1 ± 0.8	70.8 ± 6.1
November	1.1 ± 0.0	47.1 ± 2.4	52.0 ± 3.0	92.6 ± 3.4
2013	1.2 ± 0.0	36.9 ± 3.2	51.9 ± 1.4	77.6 ± 6.2
February	1.2 ± 0.0	43.7 ± 1.1	53.1 ± 2.4	96.6 ± 2.2
May	1.3 ± 0.1	37.3 ± 6.2	51.1 ± 4.5	86.2 ± 8.2
August	1.1 ± 0.0	21.6 ± 1.0	49.9 ± 2.2	39.5 ± 2.0
November	1.2 ± 0.1	45.1 ± 7.0	53.4 ± 2.7	88.2 ± 5.6
2014	1.2 ± 0.0	41.0 ± 2.8	54.3 ± 1.0	84.4 ± 4.1
February	$1.2 \pm 0.0$	49.7 ± 2.0	53.4 ± 1.1	98.2 ± 1.8
May	1.2 ± 0.1	40.4 ± 4.2	54.9 ± 1.7	84.3 ± 7.6
August	1.3 ± 0.1	27.0 ± 3.4	56.8 ± 3.3	63.2 ± 5.9
November	1.1 ± 0.1	47.0 ± 4.9	52.1 ± 1.1	91.9 ± 2.1
Grass ('12-'14)	1.2 ± 0.0	39.1 ± 1.6	53.2 ± 0.7	82.9 ± 2.7
SRC Willow	Bulk Density (a cm <sup>3</sup> )	Gravimetric Moisture (%)	Total Porosity (%)	Water-filled
0.040	(9 )			90.1 + 4.2
2012	1.4 ± 0.0	27.0 ± 1.7	45.8 ± 1.0	00.1 ± 4.2
2012 February	1.4 ± 0.0 1.4 ± 0.1	27.0 ± 1.7 23.2 ± 4.6	45.8 ± 1.0 47.7 ± 0.6	66.5 ± 10.5
2012 February May	1.4 ± 0.0 1.4 ± 0.1 1.5 ± 0.0	27.0 ± 1.7 23.2 ± 4.6 27.4 ± 2.3	45.8 ± 1.0 47.7 ± 0.6 46.0 ± 3.2	$66.5 \pm 10.5$ $93.9 \pm 4.0$
February May August	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$	$66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$
February May August November	$1.4 \pm 0.0$ 1.4 \pm 0.1 1.5 \pm 0.0 1.4 \pm 0.0 1.4 \pm 0.1	$27.0 \pm 1.7$ 23.2 ± 4.6 27.4 ± 2.3 25.4 ± 2.4 31.8 ± 3.8	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$	$66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$ $89.5 \pm 1.3$
February May August November 2013	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$	$60.7 \pm 4.2$ $66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$ $89.5 \pm 1.3$ $68.8 \pm 5.6$
2012 February May August November 2013 February	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$ $29.8 \pm 1.9$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$ $45.7 \pm 1.1$	$60.7 \pm 4.2$ $66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$ $89.5 \pm 1.3$ $68.8 \pm 5.6$ $89.0 \pm 3.1$
February May August November 2013 February May	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$ $29.8 \pm 1.9$ $27.5 \pm 6.0$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$ $45.7 \pm 1.1$ $47.5 \pm 2.3$	$60.7 \pm 4.2$ $66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$ $89.5 \pm 1.3$ $68.8 \pm 5.6$ $89.0 \pm 3.1$ $70.5 \pm 9.1$
February May August November 2013 February May August	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.3 \pm 0.1$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$ $29.8 \pm 1.9$ $27.5 \pm 6.0$ $14.2 \pm 1.0$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$ $45.7 \pm 1.1$ $47.5 \pm 2.3$ $40.8 \pm 0.5$	$60.7 \pm 4.2$ $66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$ $89.5 \pm 1.3$ $68.8 \pm 5.6$ $89.0 \pm 3.1$ $70.5 \pm 9.1$ $38.7 \pm 6.2$
February May August November 2013 February May August November	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.3 \pm 0.1$ $1.4 \pm 0.1$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$ $29.8 \pm 1.9$ $27.5 \pm 6.0$ $14.2 \pm 1.0$ $25.3 \pm 1.2$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$ $45.7 \pm 1.1$ $47.5 \pm 2.3$ $40.8 \pm 0.5$ $43.5 \pm 2.2$	$60.7 \pm 4.2$ $66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$ $89.5 \pm 1.3$ $68.8 \pm 5.6$ $89.0 \pm 3.1$ $70.5 \pm 9.1$ $38.7 \pm 6.2$ $77.1 \pm 5.6$
February May August November 2013 February May August November 2014	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.3 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$ $29.8 \pm 1.9$ $27.5 \pm 6.0$ $14.2 \pm 1.0$ $25.3 \pm 1.2$ $28.3 \pm 2.0$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$ $45.7 \pm 1.1$ $47.5 \pm 2.3$ $40.8 \pm 0.5$ $43.5 \pm 2.2$ $44.5 \pm 1.3$	$60.7 \pm 4.2$ $66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$ $89.5 \pm 1.3$ $68.8 \pm 5.6$ $89.0 \pm 3.1$ $70.5 \pm 9.1$ $38.7 \pm 6.2$ $77.1 \pm 5.6$ $85.8 \pm 3.8$
February May August November 2013 February May August November 2014 February	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.3 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.0$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$ $29.8 \pm 1.9$ $27.5 \pm 6.0$ $14.2 \pm 1.0$ $25.3 \pm 1.2$ $28.3 \pm 2.0$ $33.6 \pm 3.1$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$ $45.7 \pm 1.1$ $47.5 \pm 2.3$ $40.8 \pm 0.5$ $43.5 \pm 2.2$ $44.5 \pm 1.3$ $43.4 \pm 1.5$	$60.7 \pm 4.2$ $66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$ $89.5 \pm 1.3$ $68.8 \pm 5.6$ $89.0 \pm 3.1$ $70.5 \pm 9.1$ $38.7 \pm 6.2$ $77.1 \pm 5.6$ $85.8 \pm 3.8$ $96.8 \pm 1.9$
2012FebruaryMayAugustNovember2013FebruaryMayAugustNovember2014FebruaryMay	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.3 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.0$ $1.4 \pm 0.0$ $1.5 \pm 0.0$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$ $29.8 \pm 1.9$ $27.5 \pm 6.0$ $14.2 \pm 1.0$ $25.3 \pm 1.2$ $28.3 \pm 2.0$ $33.6 \pm 3.1$ $28.6 \pm 3.1$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$ $45.7 \pm 1.1$ $47.5 \pm 2.3$ $40.8 \pm 0.5$ $43.5 \pm 2.2$ $44.5 \pm 1.3$ $43.4 \pm 1.5$ $44.6 \pm 3.3$	$\begin{array}{c} 80.7 \pm 4.2 \\ 66.5 \pm 10.5 \\ 93.9 \pm 4.0 \\ 70.7 \pm 6.9 \\ 89.5 \pm 1.3 \\ \hline 68.8 \pm 5.6 \\ 89.0 \pm 3.1 \\ 70.5 \pm 9.1 \\ 38.7 \pm 6.2 \\ 77.1 \pm 5.6 \\ \hline 85.8 \pm 3.8 \\ 96.8 \pm 1.9 \\ 90.9 \pm 3.8 \end{array}$
2012FebruaryMayAugustNovember2013FebruaryMayAugustNovember2014FebruaryMayAugustAugust	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.3 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.0$ $1.4 \pm 0.0$ $1.5 \pm 0.0$ $1.5 \pm 0.1$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$ $29.8 \pm 1.9$ $27.5 \pm 6.0$ $14.2 \pm 1.0$ $25.3 \pm 1.2$ $28.3 \pm 2.0$ $33.6 \pm 3.1$ $28.6 \pm 3.1$ $19.8 \pm 2.2$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$ $45.7 \pm 1.1$ $47.5 \pm 2.3$ $40.8 \pm 0.5$ $43.5 \pm 2.2$ $44.5 \pm 1.3$ $43.4 \pm 1.5$ $44.6 \pm 3.3$ $47.3 \pm 3.0$	$60.7 \pm 4.2$ $66.5 \pm 10.5$ $93.9 \pm 4.0$ $70.7 \pm 6.9$ $89.5 \pm 1.3$ $68.8 \pm 5.6$ $89.0 \pm 3.1$ $70.5 \pm 9.1$ $38.7 \pm 6.2$ $77.1 \pm 5.6$ $85.8 \pm 3.8$ $96.8 \pm 1.9$ $90.9 \pm 3.8$ $70.5 \pm 10.6$
2012FebruaryMayAugustNovember2013FebruaryMayAugustNovember2014FebruaryMayAugustNovember	$1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.5 \pm 0.0$ $1.4 \pm 0.0$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.1$ $1.3 \pm 0.1$ $1.4 \pm 0.1$ $1.4 \pm 0.0$ $1.4 \pm 0.0$ $1.5 \pm 0.0$ $1.5 \pm 0.1$ $1.4 \pm 0.0$	$27.0 \pm 1.7$ $23.2 \pm 4.6$ $27.4 \pm 2.3$ $25.4 \pm 2.4$ $31.8 \pm 3.8$ $24.2 \pm 2.1$ $29.8 \pm 1.9$ $27.5 \pm 6.0$ $14.2 \pm 1.0$ $25.3 \pm 1.2$ $28.3 \pm 2.0$ $33.6 \pm 3.1$ $28.6 \pm 3.1$ $19.8 \pm 2.2$ $31.3 \pm 4.5$	$45.8 \pm 1.0$ $47.7 \pm 0.6$ $46.0 \pm 3.2$ $46.8 \pm 2.0$ $42.7 \pm 1.6$ $44.4 \pm 1.0$ $45.7 \pm 1.1$ $47.5 \pm 2.3$ $40.8 \pm 0.5$ $43.5 \pm 2.2$ $44.5 \pm 1.3$ $43.4 \pm 1.5$ $44.6 \pm 3.3$ $47.3 \pm 3.0$ $42.9 \pm 2.4$	$\begin{array}{c} 80.7 \pm 4.2 \\ 66.5 \pm 10.5 \\ 93.9 \pm 4.0 \\ 70.7 \pm 6.9 \\ 89.5 \pm 1.3 \\ \hline 68.8 \pm 5.6 \\ 89.0 \pm 3.1 \\ 70.5 \pm 9.1 \\ 38.7 \pm 6.2 \\ 77.1 \pm 5.6 \\ \hline 85.8 \pm 3.8 \\ 96.8 \pm 1.9 \\ 90.9 \pm 3.8 \\ 70.5 \pm 10.6 \\ 85.0 \pm 6.2 \\ \end{array}$

**Table 5.4:** Concentrations of dissolved organic carbon (DOC; mg/kg), nitrate (NO<sub>3</sub><sup>-</sup>; mg/kg), ammonium (NH<sub>4</sub><sup>+</sup>; mg/kg) and the ratio of nitrate to ammonia (NO<sub>3</sub><sup>-</sup> : NH<sub>4</sub><sup>+</sup>) in grassland and SRC willow in the top 15 cm soil. Data are mean  $\pm$  standard error. Annual average (*n*=16), monthly average for each year (*n*=4), and a three-year average (*n*=52) are shown.

Grassland	DOC (mg/kg)	NO <sub>3</sub> <sup>-</sup> (mg/kg)	NH4 <sup>+</sup> (mg/kg)	$NO_3^-$ : $NH_4^+$ Ratio
2012	11.95 ± 2.54	0.12 ± 0.62	0.31 ± 0.04	1 : 0.44
February	11.52 ± 2.54	0.15 ± 0.23	0.23 ± 0.07	1 : 0.79
May	20.37 ± 9.22	0.03 ± 0.07	0.46 ± 0.19	1: 0.09
August	9.99 ± 0.42	0.05 ± 0.28	0.22 ± 0.03	1 : 0.21
November	5.91 ± 0.42	0.34 ± 0.57	0.39 ± 0.02	1 : 0.89
2013	11.25 ± 3.64	0.38 ± 0.78	0.53 ± 0.06	1 : 0.9
February	5.95 ± 0.51	0.32 ± 0.16	0.69 ± 0.15	1 : 0.52
Мау	23.07 ± 14.18	0.51 ± 0.83	0.35 ± 0.1	1 : 1.57
August	8.33 ± 0.59	0.35 ± 0.11	0.56 ± 0.12	1 : 0.69
November	7.65 ± 0.37	0.37 ± 0.99	$0.53 \pm 0.07$	1 : 0.83
2014	16.82 ± 2.04	0.57 ± 1.6	0.53 ± 0.21	1 : 1.34
February	26.91 ± 4.23	1.04 ± 6.26	$0.44 \pm 0.04$	1 : 3.28
Мау	14.57 ± 1.55	0.8 ± 1.35	0.77 ± 0.13	1 : 1.1
August	15.06 ± 3.64	0.25 ± 0.61	$0.46 \pm 0.04$	1 : 0.55
November	10.73 ± 1.01	0.18 ± 0.13	$0.44 \pm 0.04$	1 : 0.42
Grass ('12-'14)	13.44 ± 1.51	0.34 ± 2.53	0.45 ± 0.03	1 : 0.86
SRC Willow	DOC (mg/kg)	NO <sub>3</sub> <sup>-</sup> (mg/kg)	NH4 <sup>+</sup> (mg/kg)	$NO_3^-$ : $NH_4^+$ Ratio
SRC Willow 2012	<b>DOC (mg/kg)</b> 8.31 ± 0.87	<b>NO<sub>3</sub><sup>-</sup> (mg/kg)</b> 0.14 ± 0.74	NH4 <sup>+</sup> (mg/kg) 0.23 ± 0.06	<b>NO</b> <sub>3</sub> <sup>-</sup> : <b>NH</b> <sub>4</sub> <sup>+</sup> <b>Ratio</b> 1 : 0.61
SRC Willow 2012 February	<b>DOC (mg/kg)</b> 8.31 ± 0.87 12.16 ± 1.23	<b>NO<sub>3</sub> (mg/kg)</b> 0.14 ± 0.74 0.04 ± 0.27	<b>NH₄<sup>+</sup> (mg/kg)</b> 0.23 ± 0.06 0.15 ± 0.01	<b>NO<sub>3</sub><sup>-</sup>: NH<sub>4</sub><sup>+</sup> Ratio</b> 1 : 0.61 1 : 0.24
SRC Willow 2012 February May	DOC (mg/kg) 8.31 ± 0.87 12.16 ± 1.23 8.23 ± 2.34	<b>NO<sub>3</sub> (mg/kg)</b> 0.14 ± 0.74 0.04 ± 0.27 0.08 ± 0.35	NH₄ <sup>+</sup> (mg/kg) 0.23 ± 0.06 0.15 ± 0.01 0.22 ± 0.03	NO <sub>3</sub> <sup>•</sup> : NH <sub>4</sub> <sup>+</sup> Ratio 1 : 0.61 1 : 0.24 1 : 0.35
SRC Willow 2012 February May August	DOC (mg/kg) 8.31 ± 0.87 12.16 ± 1.23 8.23 ± 2.34 6.56 ± 0.56	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$	NH₄ <sup>+</sup> (mg/kg) 0.23 ± 0.06 0.15 ± 0.01 0.22 ± 0.03 0.27 ± 0.1	NO <sub>3</sub> <sup>•</sup> : NH <sub>4</sub> <sup>+</sup> Ratio 1 : 0.61 1 : 0.24 1 : 0.35 1 : 0.04
SRC Willow 2012 February May August November	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$	$\begin{array}{c} \mathbf{NH_4^+ (mg/kg)} \\ 0.23 \pm 0.06 \\ 0.15 \pm 0.01 \\ 0.22 \pm 0.03 \\ 0.27 \pm 0.1 \\ 0.29 \pm 0.06 \end{array}$	NO <sub>3</sub> : NH <sub>4</sub> * Ratio 1: 0.61 1: 0.24 1: 0.35 1: 0.04 1: 1.8
SRC Willow 2012 February May August November 2013	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$	$\begin{array}{c} \mathbf{NH_4}^+ \ (\mathbf{mg/kg}) \\ 0.23 \pm 0.06 \\ 0.15 \pm 0.01 \\ 0.22 \pm 0.03 \\ 0.27 \pm 0.1 \\ 0.29 \pm 0.06 \\ 0.4 \pm 0.14 \end{array}$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1:0.61 1:0.24 1:0.35 1:0.04 1:1.8 1:3.16
SRC Willow 2012 February May August November 2013 February	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$ $5.7 \pm 1.26$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$ $0.46 \pm 0.8$	$\begin{array}{c} \mathbf{NH_4^+ (mg/kg)} \\ 0.23 \pm 0.06 \\ 0.15 \pm 0.01 \\ 0.22 \pm 0.03 \\ 0.27 \pm 0.1 \\ 0.29 \pm 0.06 \\ 0.4 \pm 0.14 \\ 0.61 \pm 0.13 \end{array}$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1: 0.61 1: 0.24 1: 0.35 1: 0.04 1: 1.8 1: 3.16 1: 1.33
SRC Willow 2012 February May August November 2013 February May	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$ $5.7 \pm 1.26$ $5.34 \pm 1.12$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$ $0.46 \pm 0.8$ $0.99 \pm 2.22$	$\begin{array}{c} \mathbf{NH_4^+ (mg/kg)} \\ 0.23 \pm 0.06 \\ 0.15 \pm 0.01 \\ 0.22 \pm 0.03 \\ 0.27 \pm 0.1 \\ 0.29 \pm 0.06 \\ 0.4 \pm 0.14 \\ 0.61 \pm 0.13 \\ 0.3 \pm 0.04 \end{array}$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1: 0.61 1: 0.24 1: 0.35 1: 0.04 1: 1.8 1: 3.16 1: 1.33 1: 3.65
SRC Willow 2012 February May August November 2013 February May August	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$ $5.7 \pm 1.26$ $5.34 \pm 1.12$ $6.19 \pm 0.65$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$ $0.46 \pm 0.8$ $0.99 \pm 2.22$ $1.41 \pm 2.16$	$\begin{array}{c} \mathbf{NH_4}^+ \ (\mathbf{mg/kg}) \\ \hline 0.23 \pm 0.06 \\ 0.15 \pm 0.01 \\ 0.22 \pm 0.03 \\ 0.27 \pm 0.1 \\ 0.29 \pm 0.06 \\ \hline 0.4 \pm 0.14 \\ 0.61 \pm 0.13 \\ 0.3 \pm 0.04 \\ 0.45 \pm 0.03 \end{array}$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1:0.61 1:0.24 1:0.35 1:0.04 1:1.8 1:3.16 1:1.33 1:3.65 1:3.25
SRC Willow 2012 February May August November 2013 February May August November	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$ $5.7 \pm 1.26$ $5.34 \pm 1.12$ $6.19 \pm 0.65$ $9.57 \pm 0.94$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$ $0.46 \pm 0.8$ $0.99 \pm 2.22$ $1.41 \pm 2.16$ $1.03 \pm 2.5$	$\begin{array}{c} \mathbf{NH_4}^+ \ (\mathbf{mg/kg}) \\ \hline 0.23 \pm 0.06 \\ 0.15 \pm 0.01 \\ 0.22 \pm 0.03 \\ 0.27 \pm 0.1 \\ 0.29 \pm 0.06 \\ \hline 0.4 \pm 0.14 \\ 0.61 \pm 0.13 \\ 0.3 \pm 0.04 \\ 0.45 \pm 0.03 \\ 0.23 \pm 0.02 \end{array}$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1:0.61 1:0.24 1:0.35 1:0.04 1:1.8 1:3.16 1:1.33 1:3.65 1:3.25 1:4.39
SRC Willow 2012 February May August November 2013 February May August November 2014	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$ $5.7 \pm 1.26$ $5.34 \pm 1.12$ $6.19 \pm 0.65$ $9.57 \pm 0.94$ $13.7 \pm 0.78$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$ $0.46 \pm 0.8$ $0.99 \pm 2.22$ $1.41 \pm 2.16$ $1.03 \pm 2.5$ $0.83 \pm 3.57$	$NH_4^+ (mg/kg)$ $0.23 \pm 0.06$ $0.15 \pm 0.01$ $0.22 \pm 0.03$ $0.27 \pm 0.1$ $0.29 \pm 0.06$ $0.4 \pm 0.14$ $0.61 \pm 0.13$ $0.3 \pm 0.04$ $0.45 \pm 0.03$ $0.23 \pm 0.02$ $0.43 \pm 0.05$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1:0.61 1:0.24 1:0.35 1:0.04 1:1.8 1:3.16 1:1.33 1:3.65 1:3.25 1:4.39 1:1.76
SRC Willow 2012 February May August November 2013 February May August November 2014 February	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$ $5.7 \pm 1.26$ $5.34 \pm 1.12$ $6.19 \pm 0.65$ $9.57 \pm 0.94$ $13.7 \pm 0.78$ $15.12 \pm 1.5$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$ $0.46 \pm 0.8$ $0.99 \pm 2.22$ $1.41 \pm 2.16$ $1.03 \pm 2.5$ $0.83 \pm 3.57$ $1.82 \pm 6.07$	$\begin{array}{c} \mathbf{NH_4}^+ \ (\mathbf{mg/kg}) \\ \hline 0.23 \pm 0.06 \\ 0.15 \pm 0.01 \\ 0.22 \pm 0.03 \\ 0.27 \pm 0.1 \\ 0.29 \pm 0.06 \\ \hline 0.4 \pm 0.14 \\ 0.61 \pm 0.13 \\ 0.3 \pm 0.04 \\ 0.45 \pm 0.03 \\ 0.23 \pm 0.02 \\ \hline 0.43 \pm 0.05 \\ 0.54 \pm 0.02 \end{array}$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1:0.61 1:0.24 1:0.35 1:0.04 1:1.8 1:3.16 1:1.33 1:3.65 1:3.25 1:4.39 1:1.76 1:3.48
SRC Willow 2012 February May August November 2013 February May August November 2014 February May	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$ $5.7 \pm 1.26$ $5.34 \pm 1.12$ $6.19 \pm 0.65$ $9.57 \pm 0.94$ $13.7 \pm 0.78$ $15.12 \pm 1.5$ $13.72 \pm 1.05$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$ $0.46 \pm 0.8$ $0.99 \pm 2.22$ $1.41 \pm 2.16$ $1.03 \pm 2.5$ $0.83 \pm 3.57$ $1.82 \pm 6.07$ $1.03 \pm 1.84$	$\begin{array}{c} \mathbf{NH_4^+ (mg/kg)} \\ 0.23 \pm 0.06 \\ 0.15 \pm 0.01 \\ 0.22 \pm 0.03 \\ 0.27 \pm 0.1 \\ 0.29 \pm 0.06 \\ \hline 0.4 \pm 0.14 \\ 0.61 \pm 0.13 \\ 0.3 \pm 0.04 \\ 0.45 \pm 0.03 \\ 0.23 \pm 0.02 \\ \hline 0.43 \pm 0.05 \\ 0.54 \pm 0.02 \\ 0.43 \pm 0.07 \\ \hline \end{array}$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1:0.61 1:0.24 1:0.35 1:0.04 1:1.8 1:3.16 1:1.33 1:3.65 1:3.25 1:4.39 1:1.76 1:3.48 1:2.32
SRC Willow 2012 February May August November 2013 February May August November 2014 February May August	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$ $5.7 \pm 1.26$ $5.34 \pm 1.12$ $6.19 \pm 0.65$ $9.57 \pm 0.94$ $13.7 \pm 0.78$ $15.12 \pm 1.5$ $13.72 \pm 1.05$ $13.62 \pm 2.68$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$ $0.46 \pm 0.8$ $0.99 \pm 2.22$ $1.41 \pm 2.16$ $1.03 \pm 2.5$ $0.83 \pm 3.57$ $1.82 \pm 6.07$ $1.03 \pm 1.84$ $0.3 \pm 0.42$	$\mathbf{NH_4^+ (mg/kg)}$ $0.23 \pm 0.06$ $0.15 \pm 0.01$ $0.22 \pm 0.03$ $0.27 \pm 0.1$ $0.29 \pm 0.06$ $0.4 \pm 0.14$ $0.61 \pm 0.13$ $0.3 \pm 0.04$ $0.45 \pm 0.03$ $0.23 \pm 0.02$ $0.43 \pm 0.05$ $0.54 \pm 0.02$ $0.43 \pm 0.07$ $0.36 \pm 0.03$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1:0.61 1:0.24 1:0.35 1:0.04 1:1.8 1:3.16 1:1.33 1:3.65 1:3.25 1:4.39 1:1.76 1:3.48 1:2.32 1:0.82
SRC Willow 2012 February May August November 2013 February May August November 2014 February May August November	<b>DOC (mg/kg)</b> $8.31 \pm 0.87$ $12.16 \pm 1.23$ $8.23 \pm 2.34$ $6.56 \pm 0.56$ $6.29 \pm 0.72$ $6.7 \pm 0.63$ $5.7 \pm 1.26$ $5.34 \pm 1.12$ $6.19 \pm 0.65$ $9.57 \pm 0.94$ $13.7 \pm 0.78$ $15.12 \pm 1.5$ $13.72 \pm 1.05$ $13.62 \pm 2.68$ $12.34 \pm 0.57$	NO <sub>3</sub> (mg/kg) $0.14 \pm 0.74$ $0.04 \pm 0.27$ $0.08 \pm 0.35$ $0.02 \pm 0.14$ $0.43 \pm 0.47$ $0.97 \pm 1.24$ $0.46 \pm 0.8$ $0.99 \pm 2.22$ $1.41 \pm 2.16$ $1.03 \pm 2.5$ $0.83 \pm 3.57$ $1.82 \pm 6.07$ $1.03 \pm 1.84$ $0.3 \pm 0.42$ $0.17 \pm 0.04$	$\begin{array}{c} \mathbf{NH_4}^+ \ (\mathbf{mg/kg}) \\ \hline 0.23 \pm 0.06 \\ 0.15 \pm 0.01 \\ 0.22 \pm 0.03 \\ 0.27 \pm 0.1 \\ 0.29 \pm 0.06 \\ \hline 0.4 \pm 0.14 \\ 0.61 \pm 0.13 \\ 0.3 \pm 0.04 \\ 0.45 \pm 0.03 \\ 0.23 \pm 0.02 \\ \hline 0.43 \pm 0.05 \\ 0.54 \pm 0.02 \\ 0.43 \pm 0.07 \\ 0.36 \pm 0.03 \\ 0.4 \pm 0.01 \\ \end{array}$	NO <sub>3</sub> : NH <sub>4</sub> + Ratio 1:0.61 1:0.24 1:0.35 1:0.04 1:1.8 1:3.16 1:1.33 1:3.65 1:3.25 1:4.39 1:1.76 1:3.48 1:2.32 1:0.82 1:0.43



**Figure 5.5:** Concentrations of soil dissolved organic carbon (DOC) in grassland (grey bar) and SRC willow (white bar).



Figure 5.6: Dissolved organic carbon (DOC) in top 15 cm soil for grass and SRC willow for each year.



**Figure 5.7:** Concentrations of soil ammonia  $(NH_4^+; dark grey bars)$  and nitrate  $(NO_3; light grey bars)$  in grassland and SRC willow.

## 5.5 Discussion

Here we have shown that litter fall made up approximately 15% of total SRC willow biomass, and after 1 year 83%, of the leaf litter had decomposed. Analysis of soil physical properties showed that grassland had lower bulk density, higher gravimetric moisture and lower porosity than SRC willow. There was no significant difference in WFPS between sites. There were more nitrate in grassland and more ammonium in SRC willow meaning there was a higher ratio of  $NO_3^-$ :  $NH_4^+$  in SRC willow. There was no significant difference in C:N ratio between sites. C:N ratio in grassland was higher in 2014 than 2012 and for all measurement years, C:N was lower in May than November. Air temperature, soil temperature, soil moisture, bulk density and total porosity all affected C:N ratio in SRC willow.

Pre-harvest, year 4 and 5 leaf litter fall averaged 5.5 t ha<sup>-1</sup> y<sup>-1</sup> for the SRC willow, which is markedly higher than some other estimates in the literature which have reported approximately 1.5 t ha<sup>-1</sup> y<sup>-1</sup> (Rytter, 2012) and another reported a 4<sup>th</sup> year production of 3.3 t ha<sup>-1</sup> (Hangs *et al.*, 2014). The post-harvest litter fall is also higher in this experiment at 2.1 t ha<sup>-1</sup> y<sup>-1</sup> compared to Rytter (2012) who reported 1.3 t ha<sup>-1</sup> y<sup>-1</sup> for the first year. These data would suggest that the SRC willow at this site are particularly productive in terms of leaf litter accumulation.

In this experiment we observed that the majority, 83%, of the leaf litter had decomposed within the first year. This is similar to what has been reported for SRC willow in other studies (Hangs *et al.*, 2014). Whilst it has been observed that the willow genotype has an effect on decomposition (Šlapokas, 1991; Šlapokas & Granhall, 1991; Hangs *et al.*, 2014) we did not observe any effect of genotype on litter decomposability. We also found that there was no effect of climatic variables on litter decomposition which are known to control decomposition rate (Berg & McClaugherty, 2008). Leaf litter quality is also known to affect the rate of decomposition, where it has been shown that litter quality primarily drives decomposition during the first year after leaf drop(Berg & McClaugherty, 2008). The C:N ratio of the leaf litter is known to affect decomposition by the decomposer community (Wardle *et al.*, 2006) and a recent meta-analysis showed that for macrofuana decomposition the decomposition rate was highest when there was an intermediate C:N ratio (between 20-30; Frouz *et al.*, 2015). SRC willow has been shown to have high quality litter (i.e. a low C:N ratio) as well as high abundance of soil fauna and microbial biomass which may explain their rapid decomposition as observed in this experiment (Heděnec *et* 

al., 2014). In this experiment the C:N ratio was 10:1 for SRC willow. A ratio of 10:1 tells us that there is rapid turnover of organic material into the soil (Hodge, 2005). C:N ratio was found to be higher in SRC willow in 2014 compared to previous years; this may be due to the increased aboveground inputs as a result of the SRC willow harvest where a lot of material was left on the ground. The ratio was lower in May than in November; again this is likely attributable to a change in organic inputs. SRC willow experiences litter fall beginning early July and ending in December, therefore only experiences organic inputs on a seasonal basis and not all year round. Litter decomposition has been shown to be very important for soil carbon sequestration and recycling of nutrients back into the soil, some studies finding that litter fall is responsible for up to 70% of annual nitrogen input (Bauer et al., 2000). In a comparison between woody bioenergy crops (willow, poplar and black locust) and herbaceous energy crops (giant reed, Miscanthus and switchgrass) it was found that there was higher SOC accumulation in the top 10 cm under woody crops which has been attributed to litter fall input (Chimento et al., 2014). LUC to bioenergy cropping systems have been shown to influence both the direction and magnitude of decomposition dynamics (Kallenbach & Gandy, 2014). Pairwise comparisons of SRC willow and arable fields have shown that decomposition is higher in SRC willow; likely due to the increased abundance of macrofauna such as earthworms and woodlice and microbial communities in the soil (Rowe et al., 2013; Makeschin, 1994). Given the predicted change in climate and rise in CO<sub>2</sub> concentrations there have been some studies that have shown that in forested ecosystems under elevated CO<sub>2</sub> there was little to no effect on litter production but increased decomposition rates (Cotrufo et al., 2005). It was also observed in this study that increased CO<sub>2</sub> in SRC plantations result in lower leaf nitrogen concentrations, therefore causing a reduction in N entering the soil via leaf litter which could have implications for future nutrient cycling under increased atmospheric CO<sub>2</sub> concentrations.

There was no significant difference in the C:N ratio between land use types, with both having a ratio of around 10:1. The ideal ratio to accommodate the nutrient requirements of all soil microbes is 24:1 (Hodge, 2005), and the ratio observed here is markedly lower than this. Stauffer *et al.* (2014) reported C:N values similar to that reported in this study with 9.6 for SRC willow and 10.2 for grassland. Jenkins (unpublished) found that the species richness of both bacteria and fungi were higher in the grassland than in the willow at these sites, though this does not seem to have a functional effect on the C:N ratio. Jenkins did however find there was an increase in the fungi *Basidiomycota* (20%), of an unidentified *Agaricales* family which are usually related to tree root symbiosis and saprotrophic degradation of wood. This suggests that the shift in microbial community has occurred to

accommodate the change in organic inputs that the soil will be receiving in the form of woody material. Whilst abundance of bacterial and fungal microbial biomass has not been measured at this site, other studies have found an increasing fungal biomass in SRC soils relative to bacterial microbial biomass (Stauffer *et al.*, 2014; Liang *et al.*, 2012). This effect may be a result of the compounds contained within SRC willow leaf litter that promote fungal growth (Stauffer *et al.*, 2014).

In this study we found that there was a higher bulk density in SRC willow than in grassland. This is likely due to the effects of compaction when the site was converted and subsequently underwent harvest. Compaction in SRC willow has been documented in other studies, where it is discussed in the context of the effects on rooting and yield (Souch *et al.*, 2004; Kuzovkina *et al.*, 2004; Edelfeldt *et al.*, 2013). For the expected level of farm machinery and foot traffic there was no significant effect of compaction on willow rooting behaviour of yield (Kuzovkina *et al.*, 2004; Souch *et al.*, 2004). Bulk density is intimately linked to total porosity, where an increase in bulk density results in a decrease in total porosity, as observed here. A decrease in soil porosity can be detrimental as it affects the movement of water and gases through the soil profile (Nimmo *et al.*, 2005). Any potential waterlogging that may occur as a result of reduced total porosity will unlikely have negative impacts on an established field of SRC willow, as they have been shown to tolerate waterlogged conditions (Volk *et al.*, 2006) with no reduction in yield (Kuzovkina *et al.*, 2004).

Whilst SRC willow had a lower TP than grassland, there was no significant difference in WFPS between sites. This indicates that there was not a higher level of water retention in SRC willow relative to the grassland site. WFPS was found to be lower in 2013 compared to the other measurement years; this is attributable to reduced rainfall in 2013 relative to 2012/2014 (673 mm vs. 1318/1023 mm, respectively). Additionally, WFPS was higher in February and November compared to August; again this is due to the higher rainfall experienced in these months relative to August (Figure 4.3). Gravimetric moisture was found to be consistently higher in the grassland than in SRC willow. This was also observed at monthly GHG sampling events where soil moisture was measured with a stab probe. SRC willow have been shown to have a much deeper rooting system and show a higher water use efficiency compared to grassland and arable crops (Don *et al.*, 2012). A comparison of SRC willow and grassland showed that SRC willow consumes more water than grassland, but has a smaller water footprint (water use per unit of biomass produced) than grasslands (Borek *et al.*, 2010). Through observation during field visits, the grassland

was waterlogged for the majority of the year, which can be observed with WFPS exceeding 90% for the majority of measurement increments. Retention of water in the soil can lead to anaerobic conditions, which results in  $NO_3^-$  within the soil being reduced to  $NH_4^+$ . Grassland had more  $NH_4^+$  than  $NO_3^-$  compared to the SRC willow, which may be explained by anaerobic soil conditions. Additionally, grassland showed emissions of soil methane throughout the experiment which have likely arisen due to high soil moisture and soil  $NH_4^+$ . SRC willow, which experienced lower soil moisture content and lower WFPS had more  $NO_3^-$  than grassland. Haycock & Pinay (1993) compared grassland and poplar as riparian buffer strips and found there was less runoff of  $NO_3^-$  for poplar. They surmised that the higher aboveground inputs contribute to soil microbial biomass which are responsible for the reduction of  $NO_3^-$ .

Over the 0-15 cm profile, DOC was higher in grassland than in SRC willow for the three year measurement period. This is consistent with the findings in Chapter 4, where there is more carbon stored in the top 0-30 cm profile, though there is no significant difference down to 100 cm. The difference in carbon in these land uses is likely due to the amounts of organic carbon being received from aboveground biomass. It was shown in Chapter 4 that after mowing 344 g C m<sup>-2</sup> y<sup>-1</sup> was left on the ground in grassland, whereas leaf litter fall reached a maximum of 291.7 g C m<sup>-2</sup> y<sup>-1</sup> in 2013. DOC was higher in 2014 at both sites, compared to previous years. In grassland, this was likely due to an increase in aboveground biomass (Figure 4.4), which resulted in increased inputs to the soil. In SRC willow, the site underwent harvest in April 2014 and the shoots rapidly began re-sprouting. There was less leaf litter in 2014 than in previous years (Figure 5.2), however as a result of the harvest there was a lot of both leaves and woody debris left onsite. Reduction of SOC observed in SRC willow has likely occurred as a result of land preparation during the conversion from grassland. Site preparation for planting SRC willow usually includes ploughing to 30 cm and subsequent harrowing to make the land amiable for planting (Tubby & Armstrong, 2002). The effects of soil surface disturbance have shown to result in losses in soil carbon such as when arable land undergoes tillage (Kaiser et al., 2014). Kahle et al. (2013) showed a conversion from an established SRC plantation to arable cropping results in a loss of soil carbon due to the excessive soil disturbance associated with management practices such as tillage.

This chapter has demonstrated that land use change from grassland to SRC results in changes in soil properties. The positive changes have been a reduction in the soil moisture content of the soil; where it was observed in grassland these conditions led to methane emissions. The negative changes that have occurred as a result of this transition are an increase in bulk density which may subsequently impede root growth; though given the yields of SRC willow this has likely not been the case. Additionally, there was a reduction in soil carbon observed as a result of land use change to bioenergy. This is likely as a result of the management practice employed in the grassland where cut material is left on the surface, therefore affording the grassland higher C inputs. Other studies have found that the recovery time of soil carbon from a transition from grassland to SRC willow can take up to 19 years (Pacaldo *et al.*, 2013). Whilst there may have been an initial loss of carbon as a result of transition, the SRC willow is fixing carbon into its biomass on an annual basis, whereas the grassland is a small source of carbon (discussed in Chapter 4). Future work at this site should look to measure the decomposition rate of grass as this was not measured in this study.

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## **CHAPTER 6: FINAL DISCUSSION**

The aim of this study was to assess the effects of land use change to bioenergy on soil carbon and GHG emissions. This was achieved in two ways, firstly through systematic review and meta-analysis of the existing literature and secondly through a field study of an ex-set aside grassland and SRC willow bioenergy plantation.

This PhD project formed part of a seven-member consortium project whose aims were to assess the effects of land use change to bioenergy at a number of sites, employing several different measurement techniques (Harris et al., 2014; Chapter 2). The ELUM project has the strength of combining sampling at multiple sites to draw more robust conclusions than would be possible from single site evaluations. One of the main outputs of the ELUM project was a meta-model which can estimate the associated GHG emissions and soil carbon changes from conversions to bioenergy in the UK. This model comes with a user friendly interface to allow policy makers and land managers to select the land of interest and assess potential impacts of LUC. The model is also parameterised for future climate scenarios out to 2050. Model parameterisation and validation was completed using data collected from two sources; (1) Data which were collected as part of the data mining work package - forming this literature review and meta-analysis, and (2) Data collected from a network of field sites which took similar measures to those collected in Chapter 4, as well as multi-year chronosequence soil sampling campaign. The model has shown significant association between modelled and measured values of CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub>; and an ability to predict impacts of LUC to bioenergy on GHG at site and national level (Dondini et al., 2015).

The systematic review of the literature and meta-analysis of available data on the effects of LUC to bioenergy on soil C and GHG emissions demonstrated our current understanding and highlighted knowledge gaps (Harris *et al.*, 2015; Chapter 3). This analysis revealed that in general a transition from arable to 2G bioenergy was largely positive resulting in reduced GHG emissions and increased soil carbon. A transition from forest to 2G bioenergy resulted in an increase in GHG emissions and a loss of soil carbon. Transitions from grasslands were not harmonious for 2G bioenergy types; a transition to SRC willow resulted in a slight increase in soil carbon. One of the large knowledge gaps identified is that surrounding non-CO<sub>2</sub> GHGs. The most abundant data surrounding the effects of LUC to 2G bioenergy on GHG emissions is for CO<sub>2</sub>, which is appropriate since this is by far the

most important GHG in relation to LUC, in the context of 2 G bioenergy. Most of the transitions of interest had some data for N<sub>2</sub>O however there was a lack of data for CH<sub>4</sub> emissions. Methane is important to consider as it has a GWP of 24 and therefore may have a large influence on the GHG balance of these transitions. In this study, it was observed that SRC willow was a sink for methane whereas grassland is a source. This finding is significant as SRC willow may result in reductions of CH<sub>4</sub>, as well as CO<sub>2</sub>. These emissions may also arise as a result of management practice as a significant portion of anthropogenic CH<sub>4</sub> emissions arise as a result of grazing animals on permanent pastures (Miller et al., 2013). The other main knowledge gap that the meta-analysis highlighted was the lack of data on transitions to SRF, which is a potential candidate for bioenergy in the UK (Hardcastle et al., 2006; McKay, 2011; Leslie et al., 2012). A recent chronosequence showed that transitions from arable land to coniferous SRF for bioenergy there was an increase in soil carbon; whilst a transition to broadleaf SRF there was no change; and a transition to eucalyptus there was a loss of soil C (Keith et al., 2015). These results have helped fill this knowledge gap but have also opened up an opportunity for further research into these land use types and the effect of LUC to SRF.

The experimental work conducted as part of this research investigated the effects of LUC from grassland to SRC willow at a commercial site in Southern England (Harris et al., submitted; Chapter 4 and 5). Both sites were low-input systems; grassland was an ex-set aside field which was mowed once a year, and material was left onsite. SRC willow received no inputs other than initial herbicide applications at establishment; after harvest no fertiliser was applied. Results from this site showed that grassland was a net source of carbon and SRC willow was a net sink. Soil N<sub>2</sub>O and CH<sub>4</sub> fluxes were very low for both sites. Soil carbon was higher under grassland than SRC willow for the top 0-30 cm, though there was no significant difference to 100 cm, although the trend was similar to the 0-30 cm measurement. The loss of soil carbon in the top 0-30 cm likely occurred as a result of site establishment and is still in recovery from initial stocks in grassland. Grassland experiences higher inputs of carbon from mowed biomass compared to that received in SRC willow from leaf litter inputs. Despite this impact on soil carbon, the net ecosystem exchange of these land uses show that on an annual scale carbon was being removed from the atmosphere and fixed into SRC willow biomass, whereas the grassland ecosystem was losing carbon to the atmosphere.

## 6.1 Bioenergy and climate change

Global climate change is occurring rapidly with an increase in global temperature, increased frequency of extreme weather events and loss of biodiversity (IPCC, 2014). Bioenergy is a proposed solution to help mitigate climate change by fixing atmospheric carbon, and also to increase energy security in the light of diminishing fossil fuels (IPCC, 2011). SRC crops have been shown to produce 14 to 85 times more energy than coal per unit of fossil energy input, with GHG emissions that were 9–161 times lower than coal (Djomo *et al.*, 2011). Current estimates suggest that if all available land for cultivation in the UK, (8 Mha of low agricultural land classes) was utilised for *Miscanthus* and SRC cultivation this could provide over 60% total heat and electricity needs in the UK (Wang *et al.*, 2014). With a more realistic deployment scenario of 0.4 Mha, 2G bioenergy could provide more than 5% of the UK's heat and electricity demand by 2020 (Wang *et al.*, 2014), helping meet the EU commitment to have 20% energy demand met by renewables by 2020 (European Commission, 2009).

Several studies agree that any initial losses in soil carbon that arise as a result of LUC to bioenergy from arable cropping are likely to be repaid within a few years of crop establishment through increased C fixation into biomass and the displacement of transitional fossil fuels, although this 'payback' time may be variable (Fargione *et al.*, 2008; Mello *et al.*, 2014). Larger 'payback' times for conversions of native lands including forests and grasslands seem likely (Fargione *et al.*, 2008; Gelfand *et al.*, 2011; Elshout *et al.*, 2015). All these studies highlight the importance of the initial conditions, the location (payback times appear longer in the tropics) and the management practices which will change along with the LUC.

As well as the carbon balance associated with bioenergy crops, there are a whole suite of other ecosystem services (ES) which may be affected by large scale cultivation of bioenergy. A recent analysis showed, with high confidence, that a transition from arable to 2G bioenergy resulted in the greatest improvements in ES, including hazard regulation disease and pest control, pollination, soil quality and water quality (Holland *et al.*, 2015). The data surrounding the effects of LUC from grassland are less clear but there are anticipated positive impacts on hazard regulation, soil quality and water quality. Effects on disease and pest control and pollination are thought to be broadly neutral, and effects on water availability are thought to be negative (Holland *et al.*, 2015). Transitions from forests, again lack certainty but are found in general to have broadly negative impacts on ES.

Spatial mapping estimates that SRC willow can be planted on 71890 ha in the UK to obtain beneficial ES impacts, with the most beneficial area of planting to be north-west England (Milner *et al.*, 2015). Another common theme emerging from the available data on ES impacts of LUC to bioenergy is the site specific nature of these impacts.

# 6.2 Importance of management in determining impact of crop cultivation

One of the major themes emerging in the literature, and also from this study, is the importance of management in determining the impact of LUC to bioenergy on soil carbon and GHG emissions.

Management has been shown to be significant in affecting the payback time of the carbon and GHG debt incurred as a result of LUC to bioenergy. Gelfand *et al.* (2011) showed that LUC from grassland to 1G bioenergy would incur GHG debts three times higher if conventional tillage was utilised compared to non-tillage management. Strictly in terms of soil carbon, Mello *et al.* (2014) showed a payback time of only 8 and 3 years for LUC from native forest and pasture to sugarcane in Brazil, respectively. These payback times are much shorter than those observed elsewhere in the literature (Fargione *et al.*, 2008) and can be attributed to fertiliser applications which result in high biomass yields and return of carbon to the soil, but ignore any other LUC impacts such as N<sub>2</sub>O emissions. Similarly, Elshout *et al.* (2015) has shown a change from no-input to high-input farming, in terms of irrigation and fertiliser application, will reduce the greenhouse gas payback time by more than 100 years.

Management can be utilised to change the cultivation of a bioenergy species from being a carbon source to a carbon sink, and vice versa. This has been termed 'management swing potential' (Davis *et al.*, 2015). For SRC willow, rotation length is a key aspect for management swing to ensure maximum biomass production (Tubby & Armstrong, 2002; Davis *et al.*, 2013). For *Miscanthus*, management swing involves leaving the crop over winter to allow reallocation of nutrients into the soil and avoiding overfertilisation (Davis *et al.*, 2015). Management swing potential is not limited to bioenergy crops; it applies to all ecosystems including grassland (Smith, 2014). In this experiment, the grassland was managed in a fairly uncommon way where grass was mowed and left on the surface, resulting in large C inputs which can help maintain soil C stocks. Grasslands are most often grazed or mowed for hay or silage, and many are often subject to a fertilisation

regime. Sites which undergo these management regimes have been shown to be both net sinks, and net sources (Soussana *et al.*, 2007; Chang *et al.*, 2015). Fertilisation can improve carbon balance through increased biomass and soil C (Conant *et al.*, 2001). However, for this field site, application of fertiliser would most likely result in large soil N<sub>2</sub>O emissions due to the high soil moisture at the site. SRC willow at the site was managed fairly typically for 2G bioenergy, though the site was not cut on a 3-year cycle. The harvest was delayed until year 5, which may have resulted in a lower GHG mitigation potential of the SRC willow than was possible if it had been harvested earlier (Tubby & Armstrong, 2002; Davis *et al.*, 2015).

## 6.3 Policy to support bioenergy deployment

Despite there being unsolved challenges of bioenergy deployment and lacking a 'one rule fits all' for LUC effects, bioenergy will still have an important role in future energy mixes and reaching emission reduction targets. The EU, as part of the Renewable Energy Directive (RED), is currently dedicated to increasing the amount of renewable energy used to 20% of total energy consumption by 2020 whilst simultaneously reducing GHG by 20% by 2020, with 10% of all liquid transport fuel coming from biomass (Directive 2009/28/EC). Recent revisions to RED from December 2014 state that biofuels produced from dedicated 2G feedstocks will count double towards the 10% target for liquid biofuels for transport while capping at 5% the share that may be met through 1G feedstocks. Additionally, this revision looks to better address the impacts of Indirect Land Use Change (iLUC) by introducing mandatory iLUC reporting for producers. iLUC occurs when the cultivation of biofuels displaces an existing land use which will need to be relocated elsewhere. The amended legislation should help prevent the impacts of iLUC, not only in the EU but globally as a 7% impact threshold now applies to all imported biomass as well as domestic supply. Despite this policy implementation, in order for bioenergy to make a substantial contribution towards emissions reduction scenarios it is likely to require sustained investment, government incentives and appropriate policies alongside a commitment from industry. Energy markets are dynamic and the discovery of a new oil source which causes a rapid decline in the price of oil can have severe knock-on effects for renewables, such as that observed in the latter half of 2014 (REN21, 2015). The latest threat to renewable energy investments is the renewed interest in fracking for natural gas in the UK. The IEA (2011) warned that the use of fracking and natural gas resources will lead
to a lack of investment in renewable energy resources, with some evidence that there is already a curb in investment in renewables as a result (EREC, 2013).

Within recent years the available data on the effects of land use change to bioenergy has increased, adding to our knowledge and highlighting uncertainties. Watson *et al.* (2015) argue that in a time of rapidly changing environment and increasing pressures on global supplies, the addition of more data will not help us find a silver bullet solution. They acknowledge that there are uncertainties surrounding bioenergy deployment, but suggest these need to be managed and risks minimised to allow investment in these technologies to help reach future energy and sustainability targets.

#### 6.4 Study limitations

There were four main limitations to this study: (1) That we were unable to follow the whole transition from planting to harvest; (2) The limited duration over which measurements were taken; (3) The frequency of the soil GHG fluxes measurements. (4) The lack of replication of net ecosystem exchange measurements.

In order to fully quantify the effects of LUC to 2G bioenergy on GHG emissions and soil carbon, the whole process of the conversion should be monitored. As mentioned in Chapter 2, the ideal design for this would be in the form of a Before-After, Control-Impact (BACI) design (Block *et al.*, 2001). This type of experimental design would allow assessment of initial conditions, capture the immediate effects of land use change and take into account any climatic variability which is difficult to account for in paired-site studies. However, these studies are difficult to implement on field scale, in a real commercial setting, especially to measure whole ecosystem GHG balance using eddy covariance towers which would be costly. Implementation of this kind of experimental design would help inform us further on the effects of LUC to bioenergy and allow portioning on effects into actual LUC and other sources such as climatic, spatial and temporal variability.

The second limitation of this study is the short duration over which measurements were taken. This study was one of the first to have a paired site comparison of a grassland and SRC willow plantation using the eddy covariance technique. When the eddy covariance technique was first becoming popular in the scientific literature, publications would cover a growing season or a year (Baldocchi, 2014). As the field has advanced, publications have reported an increased time series and the technological advances and affordability of equipment have also increased. And thus, publication requirement has become more

stringent. In this study I was able to capture the effects of a commercial harvest on the GHG balance, showing that despite a slight increase in NEE, the site quickly became a sink 3 weeks later and was a net sink for the year. As this site continues to run, several mores years' worth of data will be accumulated over the second rotation which will further our understanding of these systems.

The third limitation is the frequency at which the soil GHG fluxes were measured. For soil  $N_2O$  and  $CH_4$  in the SRC willow, and all soil GHG fluxes in grassland, measurements were only taken on a monthly time step with static GHG chambers. In the SRC willow we were able measure soil  $CO_2$  flux from dynamic chambers taking measurements every 4 hours. Being able to measure GHGs on a finer temporal scale would have provided more certainty for these measurements. As both soil  $N_2O$  and  $CH_4$  were low in both land uses this may not have been a large problem for this study. However other studies have shown that soil GHGs, in particular  $N_2O$  emissions, can arise quickly after rain events or management intervention and a large one off emission can account for a large portion annual budget (Zona *et al.*, 2013a). Fast-sensors which can measure ecosystem  $N_2O$  and  $CH_4$  using the eddy covariance method have only recently become technologically sophisticated and affordable (Baldocchi, 2014). Use of these techniques would reduce uncertainties which may arise from measurement on a smaller spatial or temporal scale which are extrapolated to an annual budget (Kroon, 2010; Yu *et al.*, 2013; Zona *et al.*, 2013b).

The final limitation of this study is the lack of replication for the net ecosystem exchange measurements. The reason for this lack of replication is due to the expense of the kit required to make these measurements; to equip one field site with eddy covariance equipment and a meteorological station costs in the region of £40,000. This lack of spatial replication means that the results found cannot be accurately up-scaled to regional or national scales, and conclusions drawn are solely for the site being measured. Despite the lack of replication, these data are still informative and will contribute to global networks of data being collected. FLUXNET is a global database where researchers can upload data from individual sites; and other can subsequently use this data to detect global trends surrounding land use, management and disturbance; as well as quantify annual and regional variation (Baldocchi *et al.*, 2001).

### 6.5 Conclusions

This research has shown that there is not a common and consistent effect when it comes to LUC to bioenergy. The initial conditions of the land, the species transitioned to, the management employed and the regional climate will all dictate how LUC to 2G bioenergy alters the net GHG balance of the system. Future assessments of the effects of LUC to bioenergy will likely need to be decided on a site by site basis for maximum certainty of any consequences of cultivation. Bioenergy will have an important role to play in future energy mixes on a global scale, and if sited appropriately can bring multiple ES benefits here in the UK. Whilst there is still some uncertainty surrounding the effects of LUC to 2G bioenergy, in order to meet future targets progress must be made and lands managed as appropriately as possible to encourage GHG mitigation.

### 6.6 Future work

Future research in this area will need to focus on three main areas; (1) increasing empirical data on carbon and GHG movement in different land uses; (2) assessing a range of management options and effects; (3) fine-tuning model predicative capabilities, through improved parameterisation and validation at multiple sites and over multiple years and different crop types.

This research highlighted several important knowledge gaps in our understanding of the effects of LUC to bioenergy cropping in the UK. Future research should be focused on transitions to SRF, transitions from grassland and the effect on non-CO<sub>2</sub> GHG emissions. Management can be a key aspect to determining the net effect of LUC to bioenergy and this is not entirely understood as of yet. More research needs to be conducted into how management interacts with LUC effects. Finally, research efforts need to continue into integrating empirical measurements into predictive models. Modelling is a powerful means by which to try and understand future impacts of LUC in a changing environment. Modelling potential effects out to the future can help inform effective decision making.

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## **APPENDIX A: SYSTEMATIC SEARCH METHODOLOGY**

The initial search method was developed in 2010 by Mathew J Tallis and was adapted by Zoe M Harris in 2011 when the work began. Searches were conducted using three commonly used search engines, namely Google Scholar, Science Direct and Web of Science. The use of different search engines was to ensure that all publications that fall under the criteria of our search were captured and the search was truly exhaustive. For example, Google Scholar is able to capture grey literature, such as governmental reports, which the other search engines will not capture. Science Direct was used as peer-reviewed publications in its databases, provided by Elsevier, were excluded from results in Google Scholar searches at the time of the original searches, although this has now changed and may be one reason why between 2010 and 2011 the numbers of hits from Google Scholar showed an increase. Web of Science was searched using two techniques, one with quotation marks around the search terms and the other without, as differences were found in the papers retrieved from the search from using either method. For example when searching 'LUC, SOC and biofuel' in 2011, the search with quotation marks gave 5 results whereas the search without yielded 18 results. It is not entirely clear why the use of quotation marks yields slightly different results, it is likely due to the search algorithms employed by the search engine. This is shown in figures and in text using: "WoS" or WoS, for each search technique respectively. The ability of Google Scholar to act as a scholarly search engine has been called into question since its beta release in 2004 (Jacsó, 2005). An understanding of search engine algorithms is important, enabling users to have an idea of how searches are performed, to assess the reliability of any search for their own purpose. Google does not disclose what algorithm they use but from several studies it appears that it uses a combination of ranking factors (Beel & Gipp, 2009a; Beel & Gipp, 2009b), taking different weightings compared to other search engines which allow the user to select how the papers are ranked; for example Science Direct allows users to select between relevance and date (Beel & Gipp, 2009a). It is apparent now, 7 years after its release, that Google Scholar is a contender in the scholarly domain and is challenging the more conventionally used search engines, Science Direct and Web of Science (Yang & Meho, 2006).

Search terms were defined and searched in a standardised format across the search engines with slight modifications made to suit the searching preferences of the particular engine. The search string was made up of four tiers, which allowed filtering of the papers through the searches and also allowed us to highlight the difference in area of interest between crop species (Fig 1.1). The results from these search engines were uploaded into a database for systematic review, but in the first instance the number of hits from the search was recorded. Search terms were defined to capture all literature which would contribute to covering the assessment of the effects of LUC to bioenergy crops in a UK context. SRF was initially one of the species terms used in the ETI contract but it was agreed at a later date, following our consultation with the consortium, that the individual species under SRF would provide a more effective search term, as these individual species terms captured references not captured by applying the generic term "SRF".

This search stage was comprised of 1024 unique searches which resulted in a total of 5786 individual references once duplicates were removed. These papers were firstly 'raw processed' by assignment of the categories 'useful' and 'not useful' based on a pre-defined selection criteria as outlined in the ETI contract. The criteria for selection were:

- the location (to be UK applicable),
- the species concerned (inclusive of first and second generation bioenergy crops)
- the mention of the metrics which we used in the meta-analysis.

After this first round of processing, the papers were more carefully inspected to extract the data in pre-defined units for the meta-analysis, performing standard unit conversions if required. The data extraction parameters were chosen to ensure they cover soil processes, GHG emissions and LCA, shown in Figure A1.1.

	Paramter	Unit		Paramter	Unit
	Paper ID			Yrs after transition	Yrs
	Author			Soil Organic Carbon	kg C ha <sup>-1</sup> yr <sup>-1</sup>
	Year			Depth for SOC	ст
	Title			Total Soil Carbon	Kg C ha <sup>-1</sup> yr <sup>-1</sup>
	Transition from:	type or n/a		Correction for bulk density	Y/N
	Paper Type	Model/field/lab/database	e p <mark>e</mark> t	CO <sub>2</sub>	
	Measument year		r cro c v	N <sub>2</sub> O	Kg CO 2 eq ha -1 yr -1
	Location	Latitude	우문 등 풀	CH <sub>4</sub>	
		Longditude	. <u>.</u>	CO <sub>2</sub>	Kg CO <sub>2</sub> eq ha <sup>-1</sup> yr <sup>-1</sup>
	pН		t of t	N <sub>2</sub> O	
	Temp	°C	<u>5</u> ≥ −	CH <sub>4</sub>	
	Precipitation	mm yr <sup>-1</sup>		Whole LCA for energy	(MJ in :MJ out )
	Yield	t ha <sup>-1</sup> yr <sup>-1</sup>		Carbon Isotopic Soil Signature	‰
Different management Regimes	Fertilization	kg ha <sup>-1</sup> yr <sup>-1</sup>		Carbon Sequestration	kg C ha <sup>-1</sup> yr <sup>-1</sup>
	Tillage	Y/N		Dissolved Organic Carbon	μg C g <sup>-1</sup> soil
	Planting Density	plants ha <sup>-1</sup>		Below Ground Biomass	Kg ha <sup>-1</sup> yr <sup>-1</sup>
	Crop Rotation	Crop & Length		Above Ground Biomass	Kg ha <sup>-1</sup> yr <sup>-2</sup>
	Irrigation	Y/N or n/a		Litter Dry Matter	g yr -1
	Residue	Y/N or n/a		Litter Decomp Rate	k
Soil Texture	Class			Root Decomp Rate	k
	Sand	%		Fine Root turnover	yr <sup>-1</sup> or % yr <sup>-1</sup>
	Silt	%		Conversions Made	
	Clay	%		Other Measurements	
	Bulk Density	g cm <sup>-3</sup>			

Figure A1.1 - Data extraction parameters for meta-analysis including standard units for measurements

The data extraction parameters were chosen to allow the meta-analysis to be conducted, but also to feed into model parameterisation and validation which took place in the ELUM project.

# APPENDIX B: SUPPLEMENTARY MATERIAL FROM META-ANALYSIS PUBLICATION

### Appendix B.1 – definition of terms

1<sup>st</sup> Generation (1G) bioenergy crops: Energy crops which are primarily derived from food crops made up of simple sugars which most often are used to make biofuels. Examples include: wheat, corn, oilseed rape, canola and sugar beet.

2<sup>nd</sup> Generation (2G) bioenergy crops: Energy crops which are derived from deciduous lignocellulosic crops which are most often used as biomass for heat and electricity generation. Examples include: SRC poplar and willow, *Miscanthus* and short rotation forestry (SRF).

**Global Warming Potential (GWP):** A relative measure of how much thermal radiation a greenhouse gas traps in the atmosphere commonly calculated over a 100 year horizon in  $CO_2$ -equivalents. Over a 100 year horizon nitrous oxide has a GWP of 298 and methane has a GWP of 25 (IPCC, 2007)

**Greenhouse gases (GHG):** Gases which directly contribute to the greenhouse gas effect by absorbing and emitting thermal radiation. In this case we focus solely on the three major GHGs; carbon dioxide ( $CO_2$ ), nitrous oxide ( $N_2O$ ) and methane ( $CH_4$ ).

**Perennial grasses:** Perennials are plants which live for longer than one year (annuals). In the context of this work perennial grasses are a group of grasses which are typically used for bioenergy cultivation. Examples include: *Miscanthus*, switchgrass and reed canary grass.

**Short rotation coppice (SRC):** These trees are planted as cuttings and cut after a year to encourage coppicing. Biomass is rapidly accumulated in multistems and the crop is harvested after 3-4 years. Trees can typically be in the ground for up to 20 years with regular coppicing without losing productivity. Examples include willow and poplar.

**Short rotation forestry (SRF):** These trees are planted and grown until the trees have reached a certain diameter. Rotations are typically 15-20 years. Examples include: Eucalyptus, alder, ash, beech, birch, sycamore, conifer and spruce.

Soil organic carbon (SOC): The organic fraction of carbon contained within the soil



## Appendix B.2: Systematic search query methodology

Figure B.1 – Systematic search string used in ISI Web of Knowledge and Google Scholar to capture all data for literature and meta-analysis

## Appendix B.3 – Meta-analysis Calculations

The standard error of each study was calculated as:

$$SE_{lnR} = \sqrt{V_{lnR}} = \sqrt{S^2 \left(\frac{1}{n_{treatment}(\bar{x}_{treatment})^2} + \frac{1}{n_{control}(\bar{x}_{control})^2}\right)}$$

The Q statistic describes the degree of between-study heterogeneity in a pool of studies. A significant result indicated that the estimated effect sizes are more heterogeneous than would be expected by chance. Q is calculated as follows:

$$Q = \sum_{i=1}^{k} W_i Y_i^2 - \frac{\left(\sum_{i=1}^{k} W_i Y_i^2\right)^2}{\sum_{i=1}^{k} W_i}$$

Where Wi is the study weight, Yi is the effect size and k is the number of studies.



Appendix B.4 - Assessment of publication bias

**Figure B.2:** Funnel plot of *InR* versus standard error of the effect of LUC on SOC for all transitions. The presence of symmetry within the funnel plot indicates little or no publication bias (regression test: t=-1.66,  $_{df}$ =136, p=0.0994).



Forest to SRC

**Figure B.3:** Funnel plots of *InR* versus standard error of the effect of LUC on SOC plotted by transition. The presence of symmetry within the funnel plots indicates little or no publication bias (regression tests: Arable to perennial grasses: *t*=0.32, <sub>df</sub>=61, *p*=0.7522, Arable to SRC: *t*=1.71, <sub>df</sub>=16, *p*=0.1072, Grass to perennial grasses: *t*=-0.91, <sub>df</sub>=41, *p*=0.3700, Grass to SRC: *t*=-1.0591, <sub>df</sub>=5, *p*=0.3380, Forest to SRC: *t*=0.51, <sub>df</sub>=5, *p*=0.6309).

### Appendix B.5 – List of publications assessed for Meta-Analysis

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## APPENDIX C: IDENTIFYING THE GOOD BIOENERGY OPTIONS - LAND USE CHANGE FROM GRASSLAND TO SRC WILLOW HAS AN IMPROVED CARBON BALANCE



Figure C.1: Percentage mass loss from leaf litter decomposition in SRC willow over 24 months.



**Figure C.2:** Soil N<sub>2</sub>O flux (mg N<sub>2</sub>O m<sup>-2</sup> d<sup>-1</sup>) for (a) grassland, (b) SRC willow and (c) both sites where grassland is shown as black circles and SRC willow is white circles. Monthly sampling took place from 8 chambers per field, standard error shown.



**Figure C.3:** Soil CH<sub>4</sub> flux (mg CH<sub>4</sub> m<sup>-2</sup> d<sup>-1</sup>) for (a) grassland, (b) SRC willow and (c) both sites where grassland is shown as black circles and SRC willow is white circles. Monthly sampling took place from 8 chambers per field, standard error shown.

**Table C1:** SRC willow stem and branch biomass scaled up from measured values to t ha<sup>-1</sup>.

Site	Year	Plot	Total dry	Total dry Mean ± SD		Mean ± SD	
			weight	(kg stump <sup>-</sup> ')		(t ha'')	
Willow	Mar 2012	1	1.59				
Willow	Mar 2012	2	2.10	-			
Willow	Mar 2012	3	1.40				
Willow	Mar 2012	4	1.31		13333	22.84 ±	
Willow	Mar 2012	5	1.94	$1.71 \pm 0.4$		5.61	
Willow	Mar 2012	6	2.28				
Willow	Mar 2012	7	1.11				
Willow	Mar 2012	8	1.98				
Willow	Nov 2012	1	2.86				
Willow	Nov 2012	2	2.95				
Willow	Nov 2012	3	2.28			347+623	
Willow	Nov 2012	4	1.99	$260 \pm 0.5$	12222		
Willow	Nov 2012	5	2.47	$2.00 \pm 0.3$	10000	34.7 ± 6.23	
Willow	Nov 2012	6	3.23				
Willow	Nov 2012	7	2.05				
Willow	Nov 2012	8	2.99				
Willow	Nov 2013	1	3.63				
Willow	Nov 2013	2	3.68				
Willow	Nov 2013	3	3.13				
Willow	Nov 2013	4	2.61	$3.40 \pm 0.5$	13333	45.39 ±	
Willow	Nov 2013	5	3.33	5.40 ± 0.5		7.18	
Willow	Nov 2013	6	4.02				
Willow	Nov 2013	7	2.78				
Willow	Nov 2013	8	4.06				
Willow	Dec 2014	1	0.70				
Willow	Dec 2014	2	0.74				
Willow	Dec 2014	3	0.85				
Willow	Dec 2014	4	0.61	0.86 ± 0.3	13333	44 4 . 0.07	
Willow	Dec 2014	5	0.96	$0.00 \pm 0.3$	10000	$11.4 \pm 3.37$	
Willow	Dec 2014	6	1.42	1			
Willow	Dec 2014	7	0.74	1			
Willow	Dec 2014	8	0.81	1			

	Table	C2: Willow	leaf litter fa	all extrapolated fr	om measured	values to t ha <sup>-1</sup> .
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Year	Plot	Total leaf fall (g)	Litter trap size	Mean $\pm$ SD		
2012	1	75.547				
2012	2	65.7752				
2012	3	61.2177	-			
2012	4	65.5149				
2012	5	53.1042	0.12	5.6 ±0.7		
2012	6	78.1607	-			
2012	7	69.786				
2012	8	64.866				
2013	1	59.562				
2013	2	71.202		5.8 ± 0.7		
2013	3	76.046				
2013	4	69.309				
2013	5	62.355	0.12			
2013	6	81.909	•			
2013	7	78.712	-			
2013	8	60.976	-			
2014	1	19.746				
2014	2	15.095	-			
2014	3	32.414				
2014	4	31.095	0.40	0.4 × 0.0		
2014	5	26.458	0.12	2.1 ± 0.6		
2014	6	25.483	1			
2014	7	36.399	1			
2014	8	17.144	1			

**Table C3:** Above and belowground biomass measured in grassland scaled up from measured plot values to t ha<sup>-1</sup>.

Site	Year	Biomass	Plot	Dry weight	Area	Scaled up to t	Mean ±	
		measured		(g)	measured	ha⁻¹	SD	
					(m²)		(t ha⁻¹)	
Grass	2013	Aboveground	1	55.28		2.21		
Grass	2013	Aboveground	2	172.99	0.25	6.92	5.08 ±	
Grass	2013	Aboveground	3	130.79		5.23	2.03	
Grass	2013	Aboveground	4	148.93		5.96		
Grass	2014	Aboveground	1	327.82	0.25	13.11	12.12 ±	
Grass	2014	Aboveground	2	278.26	0.25	11.13	1.40	
Grass	2013	Belowground	1	0.48		2.43		
Grass	2013	Belowground	2	0.80	0.025	4.07	3.58 ± 0.88	
Grass	2013	Belowground	3	0.87	0.025	4.42		
Grass	2013	Belowground	4	0.67		3.39		
Grass	2014	Belowground	1	2.015		10.26		
Grass	2014	Belowground	2	1.43	0.025	7.28	8.69 ±	
Grass	2014	Belowground	3	1.641	0.025	8.36	1.24	
Grass	2014	Belowground	4	1.736		8.84		

Model sele	ction for metha	ane (CH	₄) soil flux													
#	Treatment	Year	Soil.T	Soil.Moist	Treatment : Year	Treatment : Soil.T	Treatment : soil.Moist	Year : soil.T	Year : soil.Moist	Soil.T : soil.Moist	1 Site	df	AIC	Δ AIC	Marginal R <sup>2</sup>	Conditional R <sup>2</sup>
Minimum Adequate Model	+	-	+	-	-	-	-	-	-	+	+	6	1170.5	-8.8	0.06	0.07
2	+	-	+	+	-	-	-	-	-	+	+	7	1179.3	-12.6	0.06	0.08
3	+	+	+	+	-	-	-	-	-	+	+	10	1192.0	-19.9	0.06	0.08
4	+	+	+	+	-	-	-	+	-	+	+	13	1211.9	-10.1	0.07	0.09
5	+	+	+	+	+	-	-	+	-	+	+	16	1222.0	-23.4	0.07	0.09
6	+	+	+	+	+	-	-	+	+	+	+	19	1245.4	-8.6	0.08	0.10
7	+	+	+	+	+	+	-	+	+	+	+	20	1254.0	-10.5	0.08	0.10
Global Model	+	+	+	+	+	+	+	+	+	+	+	21	1264.5	92.4	0.08	0.09
Model sele	ction for nitrou	s oxide	(N <sub>2</sub> O) soil	flux												
#	Treatment	Year	Soil.T	Soil.Moist	Treatment : Year	Treatment : Soil.T	Treatment : soil.Moist	Year : soil.T	Year : soil.Moist	Soil.T : soil.Moist	1 Site	df	AIC	Δ AIC	Marginal R <sup>2</sup>	Conditional R <sup>2</sup>
Minimum Adequate Model	-	+	-	+	-	-	-	-	+	-	+	10	-98.4	-8.1	0.04	0.05
2	+	+	-	+	-	-	-	-	+	-	+	11	-90.4	-12.1	0.04	0.05
3	+	+	+	+	-	-	-	-	+	-	+	12	-78.3	-10.5	0.04	0.05
4	+	+	+	+	-	+	-	-	+	-	+	13	-67.8	-11.1	0.04	0.05
5	+	+	+	+	-	+	+	-	+	-	+	14	-56.8	-10.3	0.04	0.06
6	+	+	+	+	+	+	+	-	+	-	+	17	-46.4	-20.2	0.05	0.07
7	+	+	+	+	+	+	+	+	+	-	+	20	-26.2	-15.6	0.06	0.08
Global Model	+	+	+	+	+	+	+	+	+	+	+	21	-10.6	138.9	0.07	0.08

Table C4: Model selection: variables included in linear mixed models developed to explain variation in soil GHG flux

Model selection for carbon dioxide (CO<sub>2</sub>) soil flux

#	Treatment	Year	Soil.T	Soil.Moist	Treatment : Year	Treatment : Soil.T	Treatment : soil.Moist	Year : soil.T	Year : soil.Moist	Soil.T : soil.Moist	1 Site	df	AIC	Δ AIC	Marginal R <sup>2</sup>	Conditional R <sup>2</sup>
Minimum Adequate Model	+	+	+	+	+	+	-	-	+	-	+	16	-47.3	-24.9	0.65	0.66
2	+	+	+	+	+	+	-	+	+	-	+	19	-22.3	-16.9	0.65	0.66
3	+	+	+	+	+	+	-	+	+	+	+	20	-5.4	-12.4	0.65	0.66
Global Model	+	+	+	+	+	+	+	+	+	+	+	21	7.0	- 454.3	0.65	0.66

Variable	F	Df	p
treatment	60.313	1, 571	3.78E-14
year	6.783	3, 572	0.000169
soil.T	291.284	1, 575	< 2.2e-16
soil.Moist	5.256	1, 576	0.022229
treatment:year	7.139	3 <i>,</i> 570	0.000103
treatment:soil.T	15.196	1 <i>,</i> 569	0.000109
year:soil.Moist	4.051	3, 572	0.007257

Table C5: Output table for minimum adequate model for soil CO<sub>2</sub> flux. Minimum Adequate Model: CO2 flux ~ treatment + year + soil.T + soil.Moist + treatment:year + treatment:soil.T + year:soil.Moist + (1/site).

**Table C6:** Output table of linear mixed models statistics on net ecosystem exchange data. Global model: (NEE~ treatment + year+ wind.speed+rain+tsoil.1+RH+SWC.1, data=eddy.data). Grassland model: (NEE~ year+wind.speed+rain+tsoil.1+RH+SWC.1, data=eddy.grass.data). SRC willow model: (NEE~ year+wind.speed+rain+tsoil.1+RH+SWC.1, data=eddy.grass.data). SRC willow model: (NEE~ year+wind.speed+rain+tsoil.1+RH+SWC.1, data=eddy.willow.data). Where 'tsoil' is soil temperature, 'RH' is relative humidity and 'SWC.1' is soil moisture. Collinearity was detected for soil and air temperature; and for soil water content at 2 depths therefore only one term was utilised in the model. Significance codes: ns = not significant, \* p ≤ 0.05, \*\* p ≤ 0.01, \*\*\* p ≤ 0.001.

	glo	obal model		Gra	ssland model		SRC willow model			
Variables	t	p	sig	t	р	sig	t	p	sig	
treatment	-15.102	< 2e-16	***	-	-	-	-	-	-	
year	2.921	0.003545	**	-6.75	3.27E-11	***	3.604	0.000338	***	
wind.speed	-0.071	0.943713	ns	-2.79	0.00542	**	0.495	0.621039	ns	
rain	1.966	0.049509	*	2.295	0.02206	*	-0.745	0.456724	ns	
tsoil.1	-3.472	0.000534	***	4.457	9.79E-06	***	-0.68	0.496799	ns	
RH	0.233	0.815815	ns	0.127	0.89902	ns	-1.34	0.18057	ns	
SWC.1	-1.931	0.053655	ns	1.061	0.28911	ns	10.173	< 2e-16	***	
PAR	-14.141	< 2e-16	***	-10.313	< 2e-16	***	-16.711	< 2e-16	***	

Table C7: Output table for minimum adequate model for soil  $N_2O$  flux. Minimum Adequate Model: *N2O flux ~ year + soil.Moist + year:soil.Moist + (1/site)*.

Variable	F	Df	a
year	4.6343	3, 591	0.003261
soil.Moist	1.5546	1, 594	0.212948
year:soil.Moist	4.0656	3, 591	0.007105

Table C8: Output table for minimum adequate model for soil  $CH_4$  flux. Minimum Adequate Model: *CH4 flux* ~ *treatment* + *soil*.*T* + *soil*.*T* :*soil*.*Moist* + (1/*site*).

Variable	F	Df	р
treatment	8.8689	1, 591	0.00302
soil.T	18.3989	1, 592	2.09E-05
soil.T:soil.Moist	10.12	1 <i>,</i> 594	0.001543

## **APPENDIX D: ADDITIONAL DATA**



Figure D1: Pairs plot from R showing correlations between variables for litter fall data.



Figure D2: Mixed model effects plot for leaf litter fall analysis



Figure D3: Pairs plot for DOC data



Figure D4: Pairs plot for NO<sub>3</sub>, NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub>: NH<sub>4</sub><sup>+</sup> ratio



**Figure D5:** Effects plot for global DOC model. DOC.model = lme(DOC~Treatment+Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=DOC.data, na.action="na.omit")



**Figure D6:** Effects plot for grassland DOC model. gDOC.model = Ime(DOC~Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=gDOC.data, na.action="na.omit")



**Figure D7:** Effects plot for willow DOC model. wDOC.model = lme(DOC~Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=wDOC.data, na.action="na.omit").



**Figure D8:** Effects plot for global NH<sub>4</sub><sup>+</sup> model. NH4.model = Ime(NH4~treatment+Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=gKCL.data, na.action="na.omit").



**Figure D9:** Effects plot for grass NH<sub>4</sub><sup>+</sup> model. gNH4.model = Ime(NH4~Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=gKCL.data, na.action="na.omit").



**Figure D10:** Effects plot for SRC willow NH<sub>4</sub><sup>+</sup> model. wNH4.model = Ime(NH4~Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=wKCL.data, na.action="na.omit").



**Figure D11:** Effects plot for global NO<sub>3</sub><sup>-</sup> model. NO3.model = Ime(NO3~Treatment+Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=KCL.data, na.action="na.omit")



**Figure D12:** Effects plot for grass NO<sub>3</sub><sup>-</sup> model. gNO3.model = lme(NO3~Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=gKCL.data, na.action="na.omit").



**Figure D13:** Effects plot for SRC willow  $NO_3^-$  model. > wNO3.model = Ime(NO3~Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=wKCL.data, na.action="na.omit").



**Figure D14:** Effects plot for global NO<sub>3</sub><sup>-</sup>: NH<sub>4</sub><sup>+</sup> model. > Ratio.model = Ime(Ratio~Treatment+Year+Month+air.temp+BD+WFPS, random=~1|Chamber, dat a=KCL.data, na.action="na.omit").



**Figure D15:** Effects plot for grassland  $NO_3^-$ :  $NH_4^+$  model. gRatio.model = Ime(Ratio~Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=gKCL.data, na.action="na.omit").



**Figure D16:** Effects plot for SRC willow  $NO_3^-$ :  $NH_4^+$  model. wRatio.model = Ime(Ratio~Year+Month+air.temp+BD+WFPS, random=~1|Chamber, data=wKCL.data, na.action="na.omit")



Figure D17: Pairs plot for C:N data.



**Figure D18:** Effects plot for global C:N model. CNRatio.model = Ime(CN~Site+Year+Month+air.temp+BD, random=~1|Chamber, data=CN.data, na.action="na.omit").



**Figure D19:** Effects plot for grass C:N ratio. gCNRatio.model = Ime(CN~Year+Month+air.temp+BD, random=~1|Chamber, data=gCN.data, na.action="na.omit").



**Figure D20:** Effects plot for SRC willow C:N model. wCNRatio.model = Ime(CN~Year+Month+air.temp+BD, random=~1|Chamber, data=wCN.data, na.action="na.omit").

## APPENDIX E: IDENTIFYING GENOTYPES OF SRC WILLOW

## Introduction

Willow (*Salix* spp) breeding programmes were established in 1987 in Sweden and 1996 in the UK (Lindegaard *et al.*, 2001), to create high yielding, disease- and pest-resistant crops suitable for mass cultivation and mechanical harvesting. At present there are several high performing lines which are typically planted in mixed stands to incur greater resistance; a list of the available and outclassed (commercial) genotypes in the UK can be seen in Table 5.1. As willow varieties are being selected based on different traits, the genotype of the willow may result in a difference in soil GHG fluxes, litter fall, soil chemistry and aboveground biomass. For example the unreleased *Endurance* tends to outperform other genotypes in all locations for yield but other varieties which are less highly yielding have other advantages, such as *Terra Nova* which is free from disease and pests, and is able to retain leaves even if shadowed by other willow genotypes (Lindegaard *et al.*, 2011).

The aim of this section is to establish which genotypes are planted in the SRC willow field.
**Table E.1:** SRC willow genotypes and their commercial availability. Data sourced from the Teagasc & AFBI (2012)

 and personal communication with Rothamsted Research (2015).

Genotype	Availability in UK	Reference	Tested for
Advance	Possible release 2014	Teagasc & AFBI, 2012	No
Asgerd	No longer available	Teagasc & AFBI, 2012	Yes
Astrid	No longer available	Teagasc & AFBI, 2012	Yes
Baldwin	Unknown		Yes
Beagle	Commercially available	Teagasc & AFBI, 2012	Yes
Bjorn	No longer available	Teagasc & AFBI, 2012	Yes
Bowles	Unknown		Yes
Discovery	No longer available	Teagasc & AFBI, 2012	Yes
Doris	No longer available	Teagasc & AFBI, 2012	Yes
Endeavour	Commercially available	Teagasc & AFBI, 2012	Yes
Endurance	Possible release 2015	Teagasc & AFBI, 2012	No
Gudrun	Commercially available	Teagasc & AFBI, 2012	Yes
Helga	Unknown		Yes
Inger	Commercially available	Teagasc & AFBI, 2012	Yes
Jorr	Commercially available	Teagasc & AFBI, 2012	Yes
Jorunn	No longer available	Teagasc & AFBI, 2012	Yes
Karin	No longer available	Teagasc & AFBI, 2012	Yes
Klara	Not yet trialled in UK	Teagasc & AFBI, 2012	Yes
Loden	No longer available	Teagasc & AFBI, 2012	Yes
Meteor	Possible release 2014	Teagasc & AFBI, 2012	No
Nimrod	No longer available	Teagasc & AFBI, 2012	Yes
Olof	Commercially available	Teagasc & AFBI, 2012	Yes
Orm	No longer available	Teagasc & AFBI, 2012	Yes
Quest	No longer available	Teagasc & AFBI, 2012	Yes
Rapp	No longer available	Teagasc & AFBI, 2012	Yes
Resolution	Commercially available	Teagasc & AFBI, 2012	No
Roth Chiltern	Commercially available	Teagasc & AFBI, 2012	Yes
Roth Cotswold	Commercially available	Teagasc & AFBI, 2012	Yes
RR04250	Unknown		Yes
Sherwood	No longer available	Teagasc & AFBI, 2012	Yes
Stott	No longer available	Teagasc & AFBI, 2012	Yes
Sven	Commercially available	Teagasc & AFBI, 2012	Yes
Terra Nova	Commercially available	Teagasc & AFBI, 2012	Yes
Tora	Commercially available	Teagasc & AFBI, 2012	Yes

Tordis	Commercially available	Teagasc & AFBI, 2012	Yes
Torhild	Commercially available	Teagasc & AFBI, 2012	Yes
Ulv	No longer available	Teagasc & AFBI, 2012	Yes

#### Methods

#### **Sampling location**

Sampling took place around the location of the 8 experimental plots where soil GHGs were measured (Figure 4.1). A total of 56 samples were taken from the field; 5 from around the location of each static chamber and 4 samples from the met station and 4 from the flux tower, one from the north, south, west and east of each piece of kit.

#### Leaf sampling from the field

Sampled trees were marked using a GPS, so co-ordinates of the location could later be mapped. Two juvenile-to-maturing leaves were carefully removed from mid-way up the stem of the tree and placed into a pre-made foil packet. The foil packets were sealed and placed immediately into a dewar of liquid nitrogen. Once all samples had been collected they were transported back to the lab and stored at -80°C until they were ready for processing.

#### Willow leaf litter DNA extraction

Leaf material for each sample was ground in liquid nitrogen using pre-chilled pestle and mortar to ensure samples did not defrost during grinding. Ground leaf material was transferred into 2 ml Eppendorf Safe-Lock tubes and stored at -80°C until ready for extraction. DNA was extracted from the ground leaf material using a modified hexadecyltrimethylammonium bromide (CTAB) extraction protocol (Doyle *et al.*, 1987). The quantity of DNA was measured using a Thermoscientific NanoDrop 1000 spectrophotometer v3.7. Any samples which were found to have less than 100 ng/µl of DNA were re-extracted. Some samples were re-extracted up to 4 times, if at the end of this time they still did not yield more than 100 ng/µl of DNA the highest yielding extraction was taken. All extracted samples were then sent to Steve Hanley at Rothamsted Research (Harpenden, UK) where they were screened with 8 microsatellite markers which can differentiate the varieties and compared to a database of 34 known varieties. This work was unable to be completed at the University of Southampton because the markers are not readily available and we do not have these facilities available.

# Results

Samples from the SRC willow field consisted of five different genotypes, two of which did not match anything in Rothamsted's database. The three identified genotypes were found to be *Tora*, *Terra Nova*, and *Tordis*. A field map showing the locations of the identified genotypes can be seen in Figure 5.1 and the genotypes for each chamber location can be found in Table 5.2. The samples which did not match any existing known genotypes were re-tested and the same result was found. It is believed that it is not a technical or contamination issue as an allele was found in one of the samples that is unique and not in any other varieties. As the willow field was harvested, there is no way to collect leaf material from exactly the same tree and repeat the protocol.

Table	E.2: Willow	aenotypes	identified f	or tree	closest to	each Statio	c Chamber	location in	willow	SRC f	ield.
1 4010		gonotypoo	aonanoa i	01 1100	0100001 10	ouon oluin	onambor	100ution ii		01101	ioia.

Chamber number	Tree genotype
1	Tora
2	Terra Nova
3	Terra Nova
4	Terra Nova
5	Tordis
6	Terra Nova
7	Terra Nova
8	Tora



Figure E.1: Field map of Gatewick field showing location of sampled SRC willow trees and their corresponding genotype.

## Discussion

The purpose of this experiment was to identify the genotypes of the SRC willow varieties. SRC willow crops are typically planted in rows using a mix of 4-6 different genotypes to confer pest, weather and disease resistance in the crop. The owner of the field was told by the contractors who planted the field: "The varieties planted were a mix of at least 4 taken from *Tora*, *Torhild*, *Tordis*, *Jorunn*, *Jorr*, *Sven*, *Olof* and *Sherwood*", with no further specifics. There is a possibility that the 'unknown' genotypes are one of the listed above but were unable to be identified due to low yielding DNA ('001' and 'Flux2' had 13.82 and 14.85 ng/µl of DNA respectively). However as the field was harvested it is very difficult to identify the exact tree from which these samples were collected and therefore no further efforts were be made to try to identify these two individuals. From Figure 5.1 it can be seen that the locations of the genotyped trees and the colouring of the crops match up very well. *Terra Nova* is located centrally in a lighter coloured canopy and *Tora* and *Tordis* on the field margins in a darker canopy.

*Tora* is the benchmark against which other varieties are compared due to its consistently high yields across all regions of the UK. It has some susceptibility to pests and a medium tolerance to frost, as well as a quite low calorific value resulting in a need for more chipping to produce the same amount of energy as other varieties. *Terra Nova* is one of the lowest yielding willow varieties but has a high conference for pest and disease resistance; it also has one of the highest calorific values compared to other varieties. Under good conditions, *Tordis* can have comparable yields to *Tora*, but on more depleted lands it does not perform as well. *Tordis* is considered one of the best varieties for cutting production and biomass productivity, with good yields and an above average calorific value (Teagasc & AFBI, 2012).

# References

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# APPENDIX F: SIMULATION OF GREENHOUSE GASES FOLLOWING LAND-USE CHANGE TO BIOENERGY CROPS USING THE ECOSSE MODEL: A COMPARISON BETWEEN SITE MEASUREMENTS AND MODEL PREDICTIONS

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**Statement of contribution:** This paper used my data to perform model parametrisation and validation, without my data this paper would not be possible.

### Abstract

This article evaluates the suitability of the ECOSSE model to estimate soil greenhouse gas (GHG) fluxes from short rotation coppice willow (SRC-Willow), short rotation forestry (SRF-Scots Pine) and Miscanthus after land-use change from conventional systems (grassland and arable). We simulate heterotrophic respiration  $(R_h)$ , nitrous oxide  $(N_2O)$  and methane  $(CH_4)$  fluxes at four paired sites in the UK and compare them to estimates of  $R_{\rm h}$  derived from the ecosystem respiration estimated from eddy covariance (EC) and R<sub>h</sub> estimated from chamber (IRGA) measurements, as well as direct measurements of N<sub>2</sub>O and CH<sub>4</sub> fluxes. Significant association between modelled and EC-derived R<sub>h</sub> was found under Miscanthus, with correlation coefficient (r) ranging between 0.54 and 0.70. Association between IRGA-derived  $R_{\rm h}$  and modelled outputs was statistically significant at the Aberystwyth site (r = 0.64), but not significant at the Lincolnshire site (r = 0.29). At all SRC-Willow sites, significant association was found between modelled and measurement-derived  $R_h$  (0.44  $\leq r \leq$  0.77); significant error was found only for the EC-derived  $R_h$  at the Lincolnshire site. Significant association and no significant error were also found for SRF-Scots Pine and perennial grass. For the arable fields, the modelled CO<sub>2</sub>correlated well just with the IRGA-derived  $R_h$  at one site (r = 0.75). No bias in the model was found at any site, regardless of the measurement type used for the model evaluation. Across all land uses, fluxes of CH<sub>4</sub> and N<sub>2</sub>O were shown to represent a small proportion of the total GHG balance; these fluxes have been modelled adequately on a monthly time-step. This study provides confidence in using ECOSSE for predicting the impacts of future land use on GHG balance, at site level as well as at national level.

#### Introduction

The interest in using bioenergy crops as an alternative energy source to fossil fuels, and to reduce greenhouse gas (GHG) emissions, has increased in recent decades (Hastings *et al.*, 2014). The commitment of the European Union is to increase the percentage of energy from renewable sources to 20% of total energy consumption by 2020 (EU, 2009). Under the Climate Change Act 2008 (Great Britain, 2008), the UK government committed to reduce GHG emissions by 80% in 2050 compared to 1990 levels; the use of bioenergy could contribute to this target using dedicated 'second generation' (2G) lignocellulosic crops/plantations, including short rotation coppice (SRC), *Miscanthus* and short rotation forestry (SRF) (Somerville *et al.*, 2010; McKay, 2011; DECC, 2012; Valentine *et al.*, 2012). Consequently, a substantial land-use change (LUC) may occur, and it might have considerable environmental and economic impact (Fargione *et al.*, 2008; Searchinger *et al.*, 2008; Gelfand *et al.*, 2011).

Carbon dioxide  $(CO_2)$  emissions of bioenergy had previously been assumed to be zero (Gustavsson *et al.*, 1995; UK, 2008) on the assumption that emissions during combustion are balanced by the carbon (C) uptake during the growth of these bioenergy plantations, but this fails to take account of GHG emissions following LUC and subsequent crop growth. To this end, it is important to assess the GHG balance of bioenergy crops, particularly during the first years after conversion.

Two approaches have been widely used to monitor  $CO_2$  fluxes: eddy covariance (EC) and the enclosure (or chamber) method. Eddy covariance (McMillen, 1988; Aubinet *et al.*, 2012) is a technique developed to estimate land–atmosphere exchange of gas and energy at ecosystem scale. The measured  $CO_2$  flux, known as net ecosystem exchange (NEE), includes ecosystem respiration ( $R_{eco}$ ) which consists of heterotrophic ( $R_h$ ) and autotrophic ( $R_a$ ) respiration, and gross primary production (GPP) at ecosystem scale. As photosynthesis only occurs during daylight hours, the night time flux is typically used to partition the NEE signal between GPP and  $R_{eco}$ . A flux-partitioning algorithm that defines a short-term temperature sensitivity of  $R_{eco}$  is applied to extrapolate  $CO_2$  fluxes from night to day (Reichstein *et al.*, 2005). In a plant removal experiment (Hardie *et al.*, 2009), the total  $R_h$  from the whole soil profile was found to be approximately between 46 and 59% of the total  $R_{eco}$ . Abdalla *et al.* (2014) used these values to simulate  $R_h$  from selected European peatland sites using a soil process-based model, ECOSSE.

Enclosure methods have been developed to measure  $CO_2$  efflux from soil; these methods involve covering an area of soil surface with a chamber and the soil  $CO_2$  efflux can be determined using two main

modes: dynamic (closed or open) and closed static. In the former mode, a steady stream of air is pumped directly in to the chamber (Christensen, 1983; Skiba *et al.*, 1992). The latter mode simply involves closing the chamber for approximately 20–60 min and taking gas samples at intervals for analysis (Hutchinson & Mosier, 1981), or circulating the chamber air through a nondestructive infrared gas analyser (IRGA) for approximately 2 min (Norman *et al.*, 1992; Smith & Mullins, 2000). Several studies have used the closed chamber method combined with root-exclusion methods, tree grilling or stable isotopes to understand the relative contribution of  $R_h$  and  $R_a$  to total soil respiration ( $R_{tot}$ ) under different land uses.

Byrne & Kiely (2006) demonstrated that  $R_a$  under grassland soil in Ireland accounted for approximately 50% of  $R_{tot}$  during the summer months and 38% during the rest of the year. Pacaldo *et al.* (2013) reported a contribution of  $R_a$  of about 18–33% of  $R_{tot}$  under SRC-Willow at three different development stages in the USA. In a study on commercial farms located across the UK, Koerber *et al.* (2010) reported a contribution of  $R_h$  on  $R_{tot}$  for wheat of approximately 32% from January to May, 79% from June to September and 67% from October to December. A meta-analysis of soil respiration partitioning studies reported values for the ratio  $R_h/R_{tot}$  for forest soils as ranging from 0.03 to 1.0 (Subke *et al.*, 2006). Overall, the ratio was higher for boreal coniferous forests than temperate sites. In temperate, mixed deciduous forests ranges for  $R_h/R_{tot}$  of 0.3–0.6 were reported (Gaudinski *et al.*, 2000; Borken *et al.*, 2006; Millard *et al.*, 2010; Heinemeyer *et al.*, 2012). Several studies have also shown that bioenergy plantations have low nitrous oxide (N<sub>2</sub>O) emissions compared to agricultural crops because of their lower nutrient requirements, thus reducing the fertilizer requirements, and more efficient nutrient uptake, thus increasing competition with microbial organisms of N<sub>2</sub>O production (Flessa *et al.*, 1998; Hellebrand *et al.*, 2010; Drewer *et al.*, 2012).

Methane (CH<sub>4</sub>) is another important GHG that may be a substantial component of the GHG balance from several terrestrial ecosystems (van den Pol-van Dasselaar *et al.*, 1999). In agricultural systems, soil is typically a small net source or sink for CH<sub>4</sub> (Boeckx & Van Cleemput, 2001). Bioenergy crops usually present either a small CH<sub>4</sub> sink (Hellebrand *et al.*, 2003; Kern *et al.*, 2012) or a small CH<sub>4</sub> source (Gelfand *et al.*, 2011). The magnitude of the CH<sub>4</sub> flux is typically much smaller than CO<sub>2</sub> and N<sub>2</sub>O, in both agricultural soils (Boeckx & Van Cleemput, 2001) and bioenergy crops (Hellebrand *et al.*, 2003). However, very few studies (Hellebrand *et al.*, 2003; Gelfand *et al.*, 2011; Kern *et al.*, 2012) have reported on the contribution of CH<sub>4</sub> emission from bioenergy systems, increasing uncertainty in the direction of this small flux (Zona *et al.*, 2013).

Several factors control the GHG emissions of both bioenergy and conventional crops, such as site management, for example fertilization (Crutzen *et al.*, 2008; Hellebrand *et al.*, 2008, 2010), previous land use (Smith & Conen, 2004) and climatic conditions (Flessa *et al.*, 1998; Hellebrand*et al.*, 2003). Despite the high variability of the GHG fluxes, to our knowledge, only one study in the UK (Drewer *et al.*, 2012) has reported on all three GHG fluxes (CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub>) from soils under bioenergy crops (*Miscanthus* and SRC-Willow) and, in particular, after transition from former conventional systems. To fill this gap, soil models are a useful tool to predict GHG fluxes when site measurements are not available, especially when studying the effects of the change in land use over time and under different climatic conditions over large areas.

However, soil models need to be extensively tested under a range of climates and soils before being applied under conditions different from those used to parameterize and calibrate the model itself. In fact, model evaluation involves running a model using input values that have not been used during the calibration process, demonstrating that it is capable of making accurate simulations under a wide range of conditions (Moriasi *et al.*, 2007). A model can only be properly evaluated against independent data and a useful model should be able to simulate those data with some degree of accuracy (Smith & Smith, 2007).

Although several soil models have been developed for conventional agricultural and forest systems, most of them have not been fully parameterized and effectively tested for application on 2G bioenergy crops, such as *Miscanthus*, SRF and SRC (Dimitriou *et al.*, 2012; Borzęcka-Walker *et al.*, 2013; Robertson *et al.*, 2015). Here, we focus on the applicability of the process-based model ECOSSE to predict soil CO<sub>2</sub> (heterotrophic respiration), N<sub>2</sub>O and CH<sub>4</sub> after transition from conventional to bioenergy crops.

The ECOSSE model was developed mainly to simulate the C and nitrogen (N) cycles using minimal input data on both mineral and organic soils (Smith et al., 2010a,b). The ECOSSE model has been previously С evaluated across the UK to simulate the effect on soil of LUC to SRF (Dondini et al., 2015a), Miscanthus and SRC-Willow (Dondini et al., 2015b), to simulate soil N<sub>2</sub>O emissions in cropland sites in Europe (Smith et al., 2010b; Bell et al., 2012) and CO<sub>2</sub> emissions from peatlands (Abdalla et al., 2014).

This article evaluates the suitability of ECOSSE for estimating soil GHG fluxes from SRC-Willow, SRF-Scots Pine and *Miscanthus* soils in the UK after LUC from conventional systems (grassland and arable). Based on previously published recommendations, a combination of graphical techniques and error statistics has been used for model evaluation (Moriasi *et al.*, 2007). Model testing is often limited by the lack of field data to which the simulations can be compared (Desjardins *et al.*, 2010). In this study, the model is evaluated against 2 years of observations at four locations in the UK, comprising one transition to SRF-Scots Pine, three transitions to SRC-Willow and two transitions to *Miscanthus*. Modelled GHG fluxes from conventional systems have also been evaluated against field measurements (three grassland and two arable fields).

## Materials and methods

#### ECOSSE model

The ECOSSE model includes five pools of soil organic matter, each decomposing with a specific rate constant except for the inert organic matter (IOM) which is not affected by decomposition. Decomposition is sensitive to temperature, soil moisture and vegetation cover; soil texture (sand, silt and clay), pH and bulk density of the soil along with monthly climate and land-use data are the inputs to the model (Coleman & Jenkinson, 1996; Smith *et al.*, 1997). The ECOSSE model is able to simulate C and N cycle for six land-use categories of vegetation: arable, grassland, forestry, seminatural, *Miscanthus* and short rotation coppice willow (SRC-Willow).

The vegetation input to the soil (SI) is estimated by a subroutine in the ECOSSE model which uses a modification of the Miami model (Lieth, 1972), a simple model that links the climatic net primary production of biomass (NPP) to annual mean temperature and total precipitation (Grieser *et al.*, 2006). For a full description of the ECOSSE model and the plant input, estimates refer to Smith *et al.* (2010a) and Dondini *et al.* (2015b).

The minimum ECOSSE input requirements for site-specific simulations are as follows:

Climate/atmospheric data:

- 30-year average monthly rainfall, potential evapotranspiration (PET) and temperature,
- Monthly rainfall, temperature and PET.

Soil data:

- Initial soil C content (kg ha<sup>-1</sup>),
- Soil sand, silt and clay content (%),

- Soil bulk density  $(g \text{ cm}^{-3})$ ,
- Soil pH and
- Soil depth (cm).

Land-use data:

• Land use for each simulation year.

The initialization of the model is based on the assumption that the soil column is at steady state under the initial land use at the start of the simulation. Previous work has used soil organic carbon (SOC) measured at steady state to determine the plant inputs that would be required to achieve an equivalent simulated value (e.g. Smith *et al.*, 2010a). This approach iteratively adjusts plant inputs until measured and simulated values of SOC converge. In the absence of additional measurements, estimated plant inputs were calculated from a feature built in the ECOSSE model which combine the NPP model Miami (Lieth, 1972, 1973), land-management practices of the initial land use and measured above-ground biomass (details are given in Dondini *et al.*, 2015b).

#### Data

In 2011–2013, four sites were sampled in Britain using a paired site comparison approach (Keith *et al.*, 2015; Rowe *et al.*, 2015). The sites and the relative measurements contribute to the ELUM (Ecosystem Land Use Modelling & Soil Carbon GHG Flux Trial) project (Harris *et al.*, 2014). Each site consisted of one reference field (arable or grassland, depending on the previous land use of the bioenergy fields) and one or more adjacent bioenergy fields (*Miscanthus*, SRC-Willow, SRF-Scots Pine), for a total of six transitions to bioenergy at four site across UK (Table 1). A full description of the sites can be found in Drewer *et al.* (2012, 2015); J. McCalmont, N. McNamara, I. Donnison and J. Clifton-Brown (in preparation); and Z. M. Harris, G. Alberti, J. R. Jenkins, E. Clark, R. Marshall, R. Rowe, N. McNamara and G. Taylor (in preparation).

**Table 1.** Details of soil C, soil bulk density and soil pH to 1 m soil depth, as well as information on the land-use history at the study fields. Soil texture to 1 m soil depth was extracted from the soil database (1 km resolution) described in Bradley *et al.* (2005).

Sito	l and use	Latitude,	Establishment	Carbon (%)	Nitrogen	Bulk density
one		longitude	year		(%)	(g cm⁻³)
West	Short rotation coppice	50 9 -0 4	2008	0.63	0.17	15
Sussex	(SRC)-Willow	50.5, 0. <del>4</del>	2000	0.03	0.17	1.5
	Grassland	50.9, -0.4	2000	0.53	0.17	1.55
East	Short rotation forestry	560 26	2000	0.05	0.19	1 47
Grange	(SRF)-Scots Pine	56.0, -5.6	2009	0.95	0.16	1.47
	Grassland	56.0, -3.6	2009	1.3	0.17	1.49
	SRC-Willow	56.0, -3.6	2009	1.57	0.17	1.38
	Arable	56.0, -3.6	Pre-1990	1.37	0.18	1.57
Lincolnshire	SRC-Willow	53.1, -0.3	2006	1.26	0.11	1.41
	Miscanthus	53.1, -0.4	2006	1.3	0.13	1.53
	Arable	53.1, -0.5	Pre-1990	1.47	0.13	1.37
Aberystwyth	Miscanthus	52.4, -4.0	2012	0.98	0.25	1.21
	Grassland	52.4, -4.0	Pre-2007	1.16	0.26	1.45

At each bioenergy and reference field, the NEE data were obtained from continuous EC measurements (McMillen, 1988; Aubinet *et al.*, 2012) using open path IRGAs (LI-7500) and sonic anemometers. All details regarding the EC data corrections, quality control, footprint and gap filling procedures can be found in Aubinet *et al.* (2003). The night time fluxes were used to partition the NEE flux measurements into GPP and  $R_{eco}$  (Reichstein *et al.*, 2005).

Soil GHG fluxes were measured on a monthly basis at eight points randomly distributed within each field. Soil CO<sub>2</sub> fluxes were measured using an IRGA connected to an SRC-1 soil respiration chamber (PP Systems, Amesbury, MA, USA). Measurements of soil CH<sub>4</sub> and N<sub>2</sub>O fluxes were made using a static chamber method (approx. 30 l) with the addition of a vent to compensate for pressure changes within the chamber during times of sampling. Gas samples were analysed by gas chromatograph. All details regarding the chamber data can be found in Drewer *et al.* (2012), Yamulki *et al.* (2013) and Case *et al.* (2014).

Measurements of soil C, soil bulk density and soil pH to 1 m soil depth, as well as information on the land-use history, were collected for each field (Keith *et al.*, 2015; Rowe *et al.*, 2015). Soil texture was measured for each site up to a depth of 30 cm; values to 1 m soil depth were extracted from the soil

database (1 km resolution) described in Bradley *et al.* (2005), which is a collated soils data set for England and Wales, Scotland and Northern Ireland. Air temperature and precipitation data at each location were extracted from the E-OBS gridded data set from the EU-FP6 project ENSEMBLES, provided by the ECA&D project (Haylock *et al.*, 2008). This data set is known as E-OBS and is publicly available (http://eca.knmi.nl/). For each location, monthly air temperature and precipitation for the 30 years before measurements started were used to calculate a long-term average (Table 2). At each site, air temperature and precipitation were collected during the entire study period and monthly values were used as input to the model. Monthly PET was estimated using the Thornthwaite method (Thornthwaite, 1948), which has been used in other modelling studies when direct observational data have not been available (e.g. Smith *et al.*, 2005; Dondini *et al.*, 2015a).

**Table 2.** Long-term (30 years) monthly rainfall, temperature, potential evapotranspiration (PET). Monthly rainfall and temperature were extracted from the

 E-OBS data set (Haylock *et al.*, 2008;http://eca.knmi.nl/). Monthly PET was estimated using the Thornthwaite method (Thornthwaite, 1948).

	Aberystwyth			East Grange			Lincoln			West Sussex		
Month	Rain (mm)	Temperatur e (°C)	PET (mm)	Rain (mm)	Temperature (°C)	PET (mm)	Rain (mm)	Temperature (°C)	PET (mm)	Rain (mm)	Temperature (°C)	PET (mm)
January	152	4	15	103	3	11	48	4	13	80	5	16
February	112	4	17	72	3	15	37	4	17	54	5	18
March	124	5	29	74	5	27	41	6	30	55	7	30
April	86	7	45	53	7	47	43	9	48	46	9	48
May	82	10	69	61	10	72	45	12	73	47	12	73
June	93	13	89	60	13	96	56	14	97	48	15	95
July	105	15	101	67	14	105	49	17	112	49	17	110
August	114	14	93	77	14	96	55	17	103	52	17	103
September	121	13	71	84	12	70	49	14	76	60	15	79
October	174	10	46	100	9	43	55	11	46	99	12	51
November	171	7	27	94	5	22	53	7	25	88	8	29
December	168	4	17	91	3	12	51	4	14	86	6	18

#### Model evaluation and statistical analysis

Monthly simulations of soil CO<sub>2</sub>, N<sub>2</sub>O and CH<sub>4</sub> fluxes were evaluated against monthly chamber measurements. In addition, the soil CO<sub>2</sub> predicted by the ECOSSE model was compared to estimates of  $R_{\rm h}$  derived from the NEE measured by the EC.

At each site, the ECOSSE model has been run for the reference field (i.e. no land-use transition) and the bioenergy crop field (i.e. following transition from the reference land cover). The reference fields have been run for the conventional crop (arable, grassland) with no LUC, and the length of the simulations has been defined by the age of the plantation. At the bioenergy sites, the model has been run for the reference fields (conventional crop) with LUC to bioenergy crop; the length of the simulations was based on the time after transition to bioenergy crop. Measured soil characteristics and meteorological data have been used as inputs to drive the model (see above for input details), and the results of the simulations were compared to the GHG fluxes measured at the sites.

We expected a monthly underestimate of the soil CO<sub>2</sub> flux simulations because the ECOSSE model simulates  $R_h$  (from living micro-organisms + decomposition of old C sources, i.e. saprotrophic), while the CO<sub>2</sub> fluxes measured at the sites represent the total CO<sub>2</sub> efflux from the soil profile ( $R_a + R_h$ , chamber measurements) or NEE (EC measurements). To compare the modelled and measured  $R_h$ , we estimated the  $R_h$  as a proportion of the measured CO<sub>2</sub> flux, depending on the measurement type (except EC data), vegetation type and growing season.

The EC measurements of NEE were used to derive  $R_{eco}$ ; to our knowledge, only the study by Abdalla *et al.* (2014) has reported estimates of  $R_{h}$  from  $R_{eco}$ . Abdalla *et al.* (2014) applied the approach proposed by Hardie *et al.* (2009) for peaty soils and reported a contribution of  $R_{h}$  to  $R_{eco}$  of 46–59%.

To represent the variations in  $R_h$  throughout the year, Abdalla *et al.* (2014) assumed that  $R_h$  was at the lowest value of the range (46%  $R_{eco}$ ) during the summer (June–August), the highest value (59%  $R_{eco}$ ) during the winter (December–February) and at the mean value (52.5%  $R_{eco}$ ) during the rest of the year (March–May and September–November). In this study, we used the same approach of Abdalla *et al.* (2014) to derive  $R_h$  from EC measurements from all land-use systems.

Chamber measurements represent the total  $CO_2$  flux from the soil as the sum of  $R_a$  and  $R_h$ , with the exception of grassland where exclusion of full leaves from the chamber is difficult, and therefore, aboveground plant respiration is also included in the measurements. We conducted a literature review to determine the partitioning of  $R_{tot}$  measured by the chambers under different vegetation types. Additional

experiments within the ELUM project were also undertaken to directly quantify  $R_h$  and  $R_a$  at selected the  $R_{\rm h}$  site network sites (data not shown); where available, used data we to estimate  $R_h$  from  $R_{tot}$  measured by the chambers (Lincolnshire – *Miscanthus*, West Sussex – SRC-Willow, Aberystwyth – *Miscanthus*). An overview of the data source and the monthly proportion of  $R_h$  for each vegetation type and at each site are shown in Table 3.

Table 3.	Contribution of	of heterotrophic	respiration (A	R <sub>h</sub> ) on total	respiration	(R <sub>tot</sub> ) at the	study sites.
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-		Arable	SRC-Willow		Grassland	SRF-Scots Pine
		<u>Koerber et al.</u> <u>(2010)</u>	<u>Pacaldo et al.</u> <u>(2013)</u>	Miscanthus	Byrne & Kiely (2006)	<u>Millard et al. (2010)</u>
-	January	32% R <sub>tot</sub>	75% R <sub>tot</sub>	41% R <sub>tot</sub> <sup>a</sup>		
	February	32% R <sub>tot</sub>	75% R <sub>tot</sub>	41% R <sub>tot</sub> <sup>a</sup>		
	March	32% R <sub>tot</sub>	75% R <sub>tot</sub>	85% R <sub>tot</sub> <sup>a</sup>		
	April	32% R <sub>tot</sub>	75% R <sub>tot</sub>	85% R <sub>tot</sub> <sup>a</sup>		
ire	May	32% R <sub>tot</sub>	75% R <sub>tot</sub>	85% R <sub>tot</sub> <sup>a</sup>		
nsh	June	79% R <sub>tot</sub>	75% R <sub>tot</sub>	85% R <sub>tot</sub> <sup>a</sup>		
	July	79% R <sub>tot</sub>	75% R <sub>tot</sub>	44% R <sub>tot</sub> <sup>a</sup>		
Li	August	79% R <sub>tot</sub>	75% R <sub>tot</sub>	44% R <sub>tot</sub> <sup>a</sup>		
	September	79% R <sub>tot</sub>	75% R <sub>tot</sub>	44% R <sub>tot</sub> <sup>a</sup>		
	October	67% R <sub>tot</sub>	75% R <sub>tot</sub>	44% R <sub>tot</sub> <sup>a</sup>		
	November	67% R <sub>tot</sub>	75% R <sub>tot</sub>	41% R <sub>tot</sub> <sup>a</sup>		
	December	67% R <sub>tot</sub>	75% R <sub>tot</sub>	41% R <sub>tot</sub> <sup>a</sup>		
	January		82% R <sub>tot</sub> <sup>a</sup>		60% R <sub>tot</sub> <sup>b</sup>	
	February		82% R <sub>tot</sub> <sup>a</sup>		60% R <sub>tot</sub> <sup>b</sup>	
	March		82% R <sub>tot</sub> <sup>a</sup>		60% R <sub>tot</sub> <sup>b</sup>	
	April		82% R <sub>tot</sub> <sup>a</sup>		60% R <sub>tot</sub> <sup>b</sup>	
sex	May		82% R <sub>tot</sub> <sup>a</sup>		60% R <sub>tot</sub> <sup>b</sup>	
Sus	June		82% R <sub>tot</sub> <sup>a</sup>		40% R <sub>tot</sub> <sup>b</sup>	
ist (	July		82% R <sub>tot</sub> <sup>a</sup>		40% R <sub>tot</sub> <sup>b</sup>	
We	August		82% R <sub>tot</sub> <sup>a</sup>		40% R <sub>tot</sub> <sup>b</sup>	
	September		82% R <sub>tot</sub> <sup>a</sup>		60% R <sub>tot</sub> <sup>b</sup>	
	October		82% R <sub>tot</sub> <sup>a</sup>		60% R <sub>tot</sub> <sup>b</sup>	
	November		82% R <sub>tot</sub> <sup>a</sup>		60% R <sub>tot</sub> <sup>b</sup>	
	December		82% R <sub>tot</sub> <sup>a</sup>		60% R <sub>tot</sub> <sup>b</sup>	
	January			62% R <sub>tot</sub> <sup>a</sup>	60% R <sub>tot</sub> <sup>b</sup>	
	February			62% R <sub>tot</sub> <sup>a</sup>	60% R <sub>tot</sub> <sup>b</sup>	
	March			36% R <sub>tot</sub> <sup>a</sup>	60% R <sub>tot</sub> <sup>b</sup>	
	April			36% R <sub>tot</sub> <sup>a</sup>	60% R <sub>tot</sub> <sup>b</sup>	
yth	May			36% R <sub>tot</sub> <sup>a</sup>	60% R <sub>tot</sub> <sup>b</sup>	
stw	June			36% R <sub>tot</sub> <sup>a</sup>	40% R <sub>tot</sub> <sup>b</sup>	
ery	July			36% R <sub>tot</sub> <sup>a</sup>	40% R <sub>tot</sub> <sup>b</sup>	
Ab	August			36% R <sub>tot</sub> <sup>a</sup>	40% R <sub>tot</sub> <sup>b</sup>	
	September			36% R <sub>tot</sub> <sup>a</sup>	60% R <sub>tot</sub> <sup>b</sup>	
	October			36% R <sub>tot</sub> <sup>a</sup>	60% R <sub>tot</sub> <sup>b</sup>	
	November			62% R <sub>tot</sub> <sup>a</sup>	60% R <sub>tot</sub> <sup>b</sup>	
	December			62% R <sub>tot</sub> <sup>a</sup>	60% R <sub>tot</sub> <sup>b</sup>	
	January	32% R <sub>tot</sub>	25% R <sub>tot</sub>		60% R <sub>tot</sub> <sup>b</sup>	61% R <sub>tot</sub>
ast nge	February	32% R <sub>tot</sub>	25% R <sub>tot</sub>		60% R <sub>tot</sub> <sup>b</sup>	61% R <sub>tot</sub>
Gra	March	32% R <sub>tot</sub>	25% R <sub>tot</sub>		60% R <sub>tot</sub> <sup>b</sup>	61% R <sub>tot</sub>
	April	32% R <sub>tot</sub>	25% R <sub>tot</sub>		60% R <sub>tot</sub> <sup>b</sup>	61% R <sub>tot</sub>

Мау	32% R <sub>tot</sub>	25% R <sub>tot</sub>	60% R <sub>tot</sub> <sup>b</sup>	61% R <sub>tot</sub>
June	79% R <sub>tot</sub>	25% R <sub>tot</sub>	40% R <sub>tot</sub> <sup>b</sup>	61% <i>R</i> <sub>tot</sub>
July	79% R <sub>tot</sub>	25% R <sub>tot</sub>	40% R <sub>tot</sub> <sup>b</sup>	61% R <sub>tot</sub>
August	79% R <sub>tot</sub>	25% R <sub>tot</sub>	40% R <sub>tot</sub> <sup>b</sup>	61% R <sub>tot</sub>
September	79% R <sub>tot</sub>	25% R <sub>tot</sub>	60% R <sub>tot</sub> <sup>b</sup>	61% <i>R</i> <sub>tot</sub>
October	67% R <sub>tot</sub>	25% R <sub>tot</sub>	60% R <sub>tot</sub> <sup>b</sup>	61% <i>R</i> <sub>tot</sub>
November	67% R <sub>tot</sub>	25% R <sub>tot</sub>	60% R <sub>tot</sub> <sup>b</sup>	61% R <sub>tot</sub>
December	67% R <sub>tot</sub>	25% R <sub>tot</sub>	60% R <sub>tot</sub> <sup>b</sup>	61% <i>R</i> <sub>tot</sub>

<sup>a</sup> Values derived from direct measurements on root-exclusion plots.

<sup>b</sup> Where  $R_{tot}$  is 60% of measured CO<sub>2</sub> to account for plant respiration.

A quantitative statistical analysis was undertaken to determine the coincidence and association between measured and modelled values, following methods described in Smith *et al.* (1997) and Smith & Smith (2007). The statistical significance of the difference between model outputs and experimental observations can be quantified if the standard error of the measured values is known (Hastings *et al.*, 2010). The standard errors (data not shown) and 95% confidence intervals around the mean measurements were calculated for all field sites.

The degree of association between modelled and measured values was determined using the correlation coefficient (*r*). Values for *r* range from -1 to +1. Values close to -1 indicate a negative correlation between simulations and measurements, values of 0 indicate no correlation and values close to +1 indicate a positive correlation (Smith & Smith, 2007). The significance of the association between simulations and measurements was assigned using a Student's *t*-test as outlined in Smith & Smith (2007).

Analysis of coincidence was undertaken to establish how different the measured and modelled values were. The degree of coincidence between the modelled and measured values was determined using the lack of fit statistic (*LOFIT*), and its significance was assessed using an *F*-test (Whitmore, 1991) indicating whether the difference in the paired values of the two data sets is significant. The EC measurements were not replicated, so the coincidence between measured and modelled values was determined using the mean difference (*M*), calculated as the sum of the differences between measured and modelled values and divided by the total number of measurements (Smith *et al.*, 1997). The variation across the different measurements was then used to calculate the value of Student's *t*-test and compared to the *t* distributions (two-tailed test) to obtain the probability that the mean difference is statistically significant. All statistical results were considered to be statistically significant at P < 0.05.

## Results

The ECOSSE model was evaluated by comparing the outputs to the EC-derived and IRGA-derived  $R_h$  fluxes from eleven fields over four sites, representing the following land-use systems: grassland (permanent), arable (barley), *Miscanthus*, SRC-Willow and SRF-Scots Pine.

Soil CO<sub>2</sub> fluxes under *Miscanthus* were measured at two sites, Lincolnshire and Aberystwyth. At both sites, the modelled  $R_h$  followed the same seasonal pattern of measured data (Fig. 1). At the Lincolnshire site, a statistically significant association between modelled and EC-derived  $R_h(r = 0.54)$  was found, but a small significant bias in the model simulations when tested against the EC-derived  $R_h$  was also found (Table 4). On the other hand, the IRGA-derived  $R_h$  did not correlate well with the modelled outputs (r = 0.29), but no bias was found in the model simulations (Table 4).



**Figure 1.** Eddy covariance derived (dotted line with diamond markers), IRGA derived (filled triangle) and modelled (solid line with circle markers) monthly heterotrophic  $CO_2(R_h)$  under *Miscanthus* plantations during the measurement period.

|--|

Land-use system	n Miscanthus						SRC-Willow	,		SRF-Scots Pine			Grass			Arable		
Site	Aber	ystwyth	Lince	olnshire	West	Sussex	East Grange	Linco	olnshire	East	Grange	West	Sussex	Aberystwyth	East Grange	Lince	olnshire	East Grange
Measurement type	EC	IRGA	EC	IRGA	EC	IRGA	IRGA	EC	IRGA	EC	IRGA	EC	IRGA	IRGA	IRGA	EC	IRGA	IRGA
r = Correlation Coeff.	0.7	0.64	0.54	0.29	0.77	0.75	0.73	0.7	0.44	0.7	0.62	0.87	0.48	0.52	0.54	0.5	0.75	0.03
t = Student's $t$ of $r$	4.65	3.92	2.88	1.44	3.99	5.41	3.72	4.32	2.32	4.1	3.6	5.33	2.66	2.85	2.98	1.91	5.31	0.12
<i>t</i> -value at ( <i>P</i> = 0.05)	2.07	2.07	2.09	2.07	2.2	2.07	2.18	2.09	2.07	2.1	2.08	2.26	2.07	2.07	2.08	2.2	2.07	2.16
LOFIT = Lack of Fit																		
F	N/A	0.88	N/A	0.42	N/A	0.51	0.6	N/A	0.55	N/A	0.4	N/A	0.5	1.47	1.14	N/A	0.61	0.27
F (Critical at 5%)	N/A	1.6	N/A	1.58	N/A	1.58	1.84	N/A	1.58	N/A	1.61	N/A	1.58	1.6	1.61	N/A	1.6	1.8
M = Mean Difference (Kg C ha <sup>-1</sup> month <sup>-1</sup> )	13	_	260	_	-3	-3	-	233	_	-10	_	-104	-	-	-	530		-
t = Student's $t$ of $M$	1.89	-	4.8	-	-0.57	-0.57	-	6.14	-	3.6	-	-2.23	-	_	_	5.54		-
<i>t</i> -value (Critical at 2.5% – two-tailed)	2.23	-	2.09	_	2.2	2.2	-	2.09	_	2.1	-	2.26	_	-	-	2.2		-
Number of Values	24	24	22	22	13	25	14	21	22	24	23	11	24	24	23	13	22	14

Comparison of model outputs with eddy covariance (EC)-derived and IRGA-derived  $R_h$ . Association is significant for t > t-value (at P = 0.05). Error between measured and modelled values is not significant for F < F-value (critical at 5%). Mean difference is not significant for t < t-value (Critical at 2.5% – two-tailed).

At the Aberystwyth site, significant association between modelled and measurement-derived  $R_h$  was found, regardless the type of measurement used. A slightly higher correlation coefficient was calculated correlating the modelled  $R_h$  with the EC-derived  $R_h$  (r = 0.70) compared to the one arising from the correlation with the IRGA-derived  $R_h$  (r = 0.64). No significant error between simulated and IRGA-derived  $R_h$  was found for this site, but a bias in the model was found when it was tested against the EC-derived  $R_h$  (Table 4).

The model performance to simulate soil CO<sub>2</sub> fluxes under SRC-Willow was tested against measurements taken at three sites: Lincolnshire, West Sussex and East Grange (Fig. 2). At all sites, a good agreement was found between simulations and measurement-derived  $R_h$  with r values ranging from 0.44 to 0.77. Also, no significant error between simulated and measurement-derived  $R_h$  was found, with the exception of the EC-derived  $R_h$  at the Lincolnshire site (Table 4).



**Figure 2.** Eddy covariance derived (dotted line with diamond markers), IRGA derived (filled triangle) and modelled (solid line with circle markers) monthly heterotrophic  $CO_2(R_h)$  under SRC-Willow plantations during the measurement period.

Model performance to simulate soil CO<sub>2</sub> fluxes under SRF-Scots Pine has been evaluated against data collected at the East Grange site (Fig. 3). The modelled outputs followed the same pattern of the measured values, and the statistical analysis showed good correlation with both IRGA- and EC-derived  $R_h$ . Moreover, we found no statistically significant error between modelled and measured values as well as no bias in the model (Table 4).



**Figure 3.** Eddy covariance derived (dotted line with diamond markers), IRGA derived (filled triangle) and modelled (solid line with circle markers) monthly heterotrophic  $CO_2(R_h)$  under short rotation forestry-Scots Pine plantation during the measurement period.

Model simulations of soil  $R_h$  have also been evaluated for conventional crops (arable and grassland). Overall, the simulated CO<sub>2</sub> follows the same pattern as the measured values at all sites (Figs 4 and 5). The statistics highlighted a significant correlation (ranging between 0.48 and 0.87 across all sites and measurements types) and no significant error between modelled and measured values as well as no model bias under perennial grass (Table 4). For the arable fields, the modelled CO<sub>2</sub> was significantly correlated to the measured value just for the IRGA-derived  $R_h$  at the Lincolnshire site (r = 0.75); however, no bias in the model was found at any site, regardless of the measurement types used for the model evaluation (Table 4).



**Figure 4.** Eddy covariance derived (dotted line with diamond markers), IRGA derived (filled triangle) and modelled (solid line with circle markers) monthly heterotrophic  $CO_2(R_h)$  under arable plantations during the measurement period.



**Figure 5.** Eddy covariance derived (dotted line with diamond markers), IRGA derived (filled triangle) and modelled (solid line with circle markers) monthly heterotrophic  $CO_2(R_h)$  under grassland plantation during the measurement period.

Monthly fluxes of CH<sub>4</sub> and N<sub>2</sub>O were shown to be highly variable, both spatially and temporally, across all land uses, so we present an example of the correlation between modelled and measured soil N<sub>2</sub>O and CH<sub>4</sub> fluxes for each land use. Both N<sub>2</sub>O and CH<sub>4</sub> are very small fluxes and the model outputs were within the errors of the measurements, for both GHGs and at all sites (data not shown). However, low correlation between measured and modelled values has been found for the majority of the sites, ranging from -0.02 to 0.61 for N<sub>2</sub>O and from -0.29 to 0.53 for CH<sub>4</sub>. The high variability of the measured N<sub>2</sub>O and CH<sub>4</sub> fluxes led to a statistically significant error between simulated and measured values at most of the study sites (Tables 5 and 6).

Land-use system	Misca	SR	SRC-Willow					Arable			
Site	Aberystwyth	Lincolnshire	Lincolnshire	East Grange	West Sussex	East Grange	West Sussex	Aberystwyth East Stange		Lincolnshire	East Grange
<i>r</i> = Correlation Coeff.	0.34	-0.15	-0.13	0.12	-0.02	0.19	0.25	0.06	-0.12	-0.20	0.61
t = Student's t of r	1.72	0.64	0.66	0.48	0.08	0.86	1.24	0.3	0.56	0.97	3.25
<i>t</i> -value at ( <i>P</i> = 0.05) LOFIT = Lack of Fit	2.07	2.1	2.06	2.12	2.06	2.08	2.06	2.07	2.08	2.07	2.1
F	0.37	3.34	54.66	22.62	0.37	40.75	0.62	0.68	312.92	0.43	0.25
F (Critical at 5%)	1.63	1.69	1.59	1.74	1.59	1.63	1.59	1.62	1.63	1.6	1.69
Number of values	24	20	26	18	26	23	26	24	23	25	20
Association is sign	nificant for $t > t-v_{i}$	alue (at P = 0.05	). Error between	measured	and mode	elled values	is not signif	icant for F < F-va	lue (critical a	at 5%).	

Table 5. ECOSSE model performance at simulating N<sub>2</sub>O fluxes at the study sites

Land-use system	Miscanthus		SRC-Willow		SRF-Scots Pine			Grass			Arable	
Site	Aberystwyth	Lincolnshire	Lincolnshire	East Grange	West Sussex	East Grange	West Sussex	Aberystwyth	East Grange	Lincolnshire	East Grange	
r = Correlation Coeff.	0.31	0.28	0.18	0.53	0.18	0.53	0.27	0.51	0.41	-0.29	0.05	
t = Student's $t$ of $r$	1.52	1.28	0.88	2.51	0.91	2.68	1.4	2.81	1.91	1.44	0.2	
<i>t</i> -value at ( <i>P</i> = 0.05)	2.07	2.09	2.07	2.12	2.06	2.1	2.06	2.07	2.1	2.07	2.1	
LOFIT = Lack of Fit												
F	0.33	3.61	6.5	0.53	0.61	2.38	0.3	0.34	4.09	0.66	0.76	
F (Critical at 5%)	1.62	1.65	1.6	1.74	1.59	1.63	1.59	1.62	1.63	1.62	1.69	
Number of values	24	22	25	18	26	23	26	24	23	24	20	

Table 6. ECOSSE model performance at simulating CH4 fluxes at the study sitesLand-use systemMiscanthusSRC-Willow

## Discussion

Soil CO<sub>2</sub> emissions under *Miscanthus* have been quantified at two sites (Lincolnshire and Aberystwyth) using two different sampling methods (EC and IRGA methods). At both sites, we found a high correlation between measured and modelled  $R_h$ , ranging from 0.54 to 0.60, except for the IRGA values at Lincolnshire site (r = 0.29, Table 4). The lack of association at this site was mainly due to differences between modelled and IRGA-derived  $R_h$  in the year 2013 (Fig. 1b). In April 2013, the soil was harrowed and disked to break up the rhizomes for improved yield, so the system was out of balance; the farmer also applied waste wood products, which led to high CO<sub>2</sub> emissions, undetected by the model (May–August 2013 in Fig. 1b) as this was not included in the management file. In the ECOSSE model, the patterns of C and N debris return during the growing season follow a standard exponential relationship, as originally derived by Bradbury *et al.* (1993). Any alteration, such as harrowing or waste application, cannot be easily entered by the user. The scope of the present study is to evaluate the model using independent data which has not been used to develop the model. Therefore, we deliberately chose not to apply any modifications to the model to fit the measured data. However, the model was able to simulate independent data derived from two different sources with a good degree of accuracy.

Soil CO<sub>2</sub> emissions under SRC-Willow and SRF-Scots Pine plantations have been quantified using the same sampling methods. At all sites, the modelled  $R_h$  significantly correlated with all types of measurements, showing no significant error between measured and modelled values (Fig. 2).

The model has also been tested against  $CO_2$  fluxes measured under conventional crops. At all three grassland sites (West Sussex, Aberystwyth and East Grange), the measured  $CO_2$  fluxes correlate significantly with the modelled values and the statistical analysis showed no error between measured and modelled values, and no bias in the model (Fig. 5). This is a striking result which underlines the good quality of the data provided for the model evaluation, as well as the good model performance to simulate soil  $CO_2$  fluxes.

Under grassland,  $R_h$  derived from the IRGA measurements does not always show a high correlation with the modelled values, particularly during the summer months (Fig. 5). This lack of correlation is mainly due to the difficulties in the separation of soil respiration from grassland, due to the possible inclusion of vegetation within the chamber. When deriving  $R_h$  from grassland, we estimated that 60% of the measured CO<sub>2</sub> can be attributed to plant (leaf) respiration, as reported by Byrne & Kiely (2006), but this crude estimate does not always reflect the field conditions. For an accurate quantification of the proportion of the CO<sub>2</sub> derived from the plant occluded in the chambers, field experiments would be needed to explicitly quantify plant respiration and biomass.

The analysis of the soil  $R_h$  fluxes from the arable fields reveals reasonable model performance at the Lincolnshire site, while at the East Grange site, correlation between modelled and measured IRGA values was poor (Table 4). This discrepancy between modelled and measurement-derived  $R_h$  appears to be due to the nature of the source data; in fact, the IRGA-derived  $R_h$  is estimated from a single data point which is taken to represent monthly CO<sub>2</sub> fluxes. Therefore, the monthly CO<sub>2</sub> flux might not be properly represented if high flux variation occurred within the month. Another explanation could also be the discontinuity of the IRGA measurements taken at the East Grange site (Fig. 4b). The latter hypothesis is supported by the  $R_h$  results of the arable field at the Lincolnshire site. In fact, the IRGA measurements at the Lincolnshire site have been taken over a 2-year period, and the statistical analysis shows a good correlation against the model output (r = 0.75; Table 4). Therefore, we conclude that the low correlation at the East Grange arable field is mainly due to the variability and quantity of the measurements, and that the model accurately describes the CO<sub>2</sub>emissions from arable crop.

Generally, the model was able to predict seasonal trends in  $R_h$  at most of the sites; however, the model occasionally over/underestimated the flux values during the warm weather in spring and summer. This is particularly evident at the Lincolnshire site, resulting in a high mean difference between modelled and EC-derived  $R_h$  (Table 4). Despite using a generic method to estimate  $R_h$  from  $R_{eco}$ , therefore providing a challenging test for the model, we found no significant mean difference between modelled and EC-derived  $R_h$  at three sites (for a total of four land uses), proving that the model adequately simulates soil processes under different land-use systems and climate/soil conditions.

Low correlation between measurements and model simulations arose predominantly when comparing model outputs against the IRGA-derived data set; this is mainly due to the nature of the measurements (single data point representing total monthly  $CO_2$  flux), an aspect not related to the soil processes described in the model. However, it is to notice that the IRGA-derived  $R_h$  has been estimated from direct measurements of total soil respiration and the degree of correlation between measured and modelled  $R_h$  is also related to the  $R_h : R_{tot}$  ratio adopted. On the other hand, the EC-derived  $R_h$  was estimated from the  $R_{eco}$  during daytime, which is a modelled flux driven by air temperature and other environmental factors. Further model evaluation should be based on comparison of the model output with direct measurements of soil  $R_h$  fluxes, possibly using automatic chambers on soil plots where roots have been excluded. This measurement technique would provide continuous  $R_h$  measurements which would be

directly comparable to the model outputs and therefore would provide a more accurate evaluation of the performance of the model. However, given the very limited input data used to run the model and the number of sites/locations used for the model evaluation, we conclude that the simulations are robust and the model adequately simulate soil  $CO_2$  fluxes under five land-use systems.

Model simulations of N<sub>2</sub>O and CH<sub>4</sub> fluxes resulted in low correlation and association at most of the study sites (Tables 5 and 6), which is expected with such low fluxes, and does not represent a failure of the model. In fact, the measured N<sub>2</sub>O and CH<sub>4</sub> fluxes are pooled from sample data points containing outliers and extreme variation between sample points in each site, which results in a high standard error of the measured values. But the N<sub>2</sub>O and CH<sub>4</sub> flux simulations are within the 95% confidence interval of the measured values, showing that the model cannot be improved to better fit these data and suggesting that the lack of correlation between modelled and measured values is due to the high variation in the measured fluxes, which is a common phenomenon verified in many N<sub>2</sub>O (e.g. Oenema *et al.*, 1997; Skiba *et al.*, 2013; Cowan *et al.*, 2015) and CH<sub>4</sub> flux measurement experiments (Parkin *et al.*, 2012; Savage *et al.*, 2014). Moreover, if the measured values do not show any seasonal trend, a significant correlation with the model outputs cannot be obtained (Smith & Smith, 2007) and low correlation is expected.

Measured fluxes of  $CH_4$  were shown to be negligible across all land uses and their contribution to the total GHG balance, when converted to  $CO_2$  equivalent, was on average <0.2%, except for the *Miscanthus* field at the Aberystwyth site (3% of the total GHG balance). The high mean value recorded for *Miscanthus* in 2012 is driven by one replicate with very high  $CH_4$  production and there was large standard error associated with the measurements. In general,  $CH_4$  production or consumption was negligible also for this field.

Across all land uses, measured fluxes of  $N_2O$  represent a small proportion (<1.5%) of the total GHG balance, with the exception of the arable field at the Lincolnshire site and the *Miscanthus* field at the Aberystwyth site (6% of the total GHG balance over the 2 years measurement period at both fields). Due to technical issues and issues regarding access to sites for sampling, the data set for the arable and SRC-Willow fields at East Grange is missing a substantial number of months, and therefore, it was not possible to determine the annual GHG balance.

Despite the very low values of the  $CH_4$  and  $N_2O$  fluxes, and their small contribution to the total GHG balance at all experimental sites, both fluxes have been modelled adequately on a monthly time-step and no improvements can be made to the model with the available flux data.

In this study, all major GHG fluxes from five land-use systems were reasonably well estimated using the ECOSSE model. The results from this evaluation exercise show that ECOSSE is robust for simulating GHG fluxes from cropland, grassland, SRC-Willow, SRF-Scots Pine and *Miscanthus* (and transitions from the former two land uses to the latter three energy crops). This validation builds confidence that the model can be used to investigate the impacts of land-use transitions spatially in the UK and to investigate the effects of converting large areas to grow bioenergy crops.

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