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PHD DISSERTATION

**CARBON DYNAMICS IN
FORESTS AND CROPLANDS:
INFLUENCE OF MANAGEMENT
AND CLIMATE EXTREMES**

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Summary

Management and climate change had and continue to have a strong impact on carbon (C) dynamics in terrestrial ecosystems. In this context, the possible effects due to changes in climate extreme frequency are far to be fully understood as well as mitigation strategies related to forest and agricultural management are still strongly debated.

The main objective of this thesis is to bring some additional knowledge on the above mentioned topics through manipulation experiments, medium-term micrometeorological observations and long-term field measurements (chronosequences) in forests and croplands.

In particular, after an introduction to the main topic of research (Chapter 1) and a brief summary of the adopted methods (Chapter 2), the thesis is divided into three research papers (one already published, one submitted and one in preparation):

- the first paper (Chapter 3) examines, through two forest chronosequences and model simulations, the C pools dynamics in a beech and a spruce forest in Italian Alps. The main objective of the work was to determine the influence of stand age and applied management on the C cycle in these forests. Results showed that shelterwood management had no influence on soil C stock, while model simulations confirmed that the applied management allows a rapid recovery of the ecosystem after disturbance (i.e. final cut);
- the second paper (Chapter 4) investigates the effects of conservation agriculture (i.e. minimum tillage + cover crops) on ecosystem C dynamics and crop productivity in a 5-year crop rotation. Continuous micrometeorological measurements on paired plots were used to quantify all C fluxes of the ecosystem. Minimum tillage resulted in a higher net ecosystem productivity than control (conventional tillage), but in a decrease of the overall C budget (net ecosystem C budget, NECB);
- the third paper (Chapter 5) focuses on temperature extreme event (i.e. cold and hot waves) impacts on heterotrophic respiration, yield and C budget in a soybean crop. Results showed that both extreme events did not alter the C budget even though an acceleration in decomposition was found in warm treatment.

Tesi di dottorato di Giuseppe De Simon, discussa presso l'Università degli Studi di Udine

General introduction

1.1 Carbon Cycle and Climate Change

Carbon (C) flows between four main compartments: the atmosphere, the hydrosphere, the terrestrial biosphere (which is composed by terrestrial biota and soil carbon) and the lithosphere. These four compartments are pools that store and exchange C at different rates and quantities, and with different lifetimes. The largest amount of C is stored in the intermediate and deep ocean, but also vegetation, soils, surface ocean and the atmosphere are considerable C pools (Figure 1.1). Moreover, these last compartments present higher turnover rates than intermediate and deep ocean, which is supposed to be quite neutral for the C cycle in coming decades (Bolin *et al* 2000, Gruber *et al* 2004).

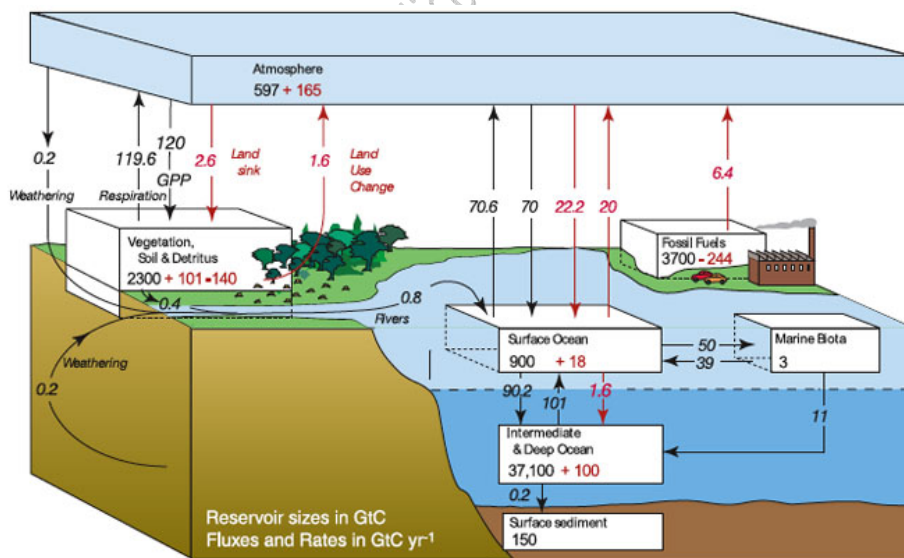


Figure 1.1: The global carbon cycle for the 1990s, showing the main annual fluxes in GtC yr⁻¹: pre-industrial 'natural' fluxes in black and 'anthropogenic' fluxes in red (modified from Sarmiento and Gruber 2006, with changes in pool sizes from Sabine *et al* 2004). Source: Denman *et al* 2007.

The higher turnover rates of terrestrial biosphere, surface ocean and atmosphere indicate that these pools play a determining role in the C cycle at time scales ranging from months/seasons to

centuries. Other pools, especially the lithosphere, present very small fluxes with a C turnover time many orders of magnitude longer than the other reservoirs (Holmén 1992).

The anthropogenic emissions are relatively small compared to the main C fluxes. The emissions from fossil fuel burning and cement production rose from $5.4 \pm 0.3 \text{ GtC yr}^{-1}$ in the 1980s to $6.4 \pm 0.4 \text{ GtC yr}^{-1}$ in the 1990s (Marland *et al* 2010), and they have continued to increase between the 1990s and 2000 to 2005, climbing to $7.2 \pm 0.3 \text{ GtC yr}^{-1}$ (\pm standard deviation; Denman *et al* 2007). However, these emissions are quite relevant as the release over the last three centuries of about 300 PgC by fossil fuel burning and cement production (Marland *et al* 2010) and of 140-160 PgC by land-use change (Houghton 2003) led to a perturbation of the C cycle which has been at a relatively steady state for millennia. These perturbations are thought to be the dominant driver of climate change because of their persistent effect on the atmosphere (Denman *et al* 2007).

In fact, in the 10,000 years before 1750, the atmospheric carbon dioxide (CO_2) concentration had been relatively stable between 260 and 280 ppm (Denman *et al* 2007; Figure 1.2); after 1750, anthropogenic activities – primarily the burning of fossil fuels, industrial processes, deforestation and land-use changes – have increased the CO_2 concentration in the atmosphere to the actual (2011) value of 391.57 ppm, with a mean annual increase for the 2002-2011 decade of 2.07 ppm yr^{-1} (data from NOAA-ESRL, Mauna Loa Observatory, Hawaii).

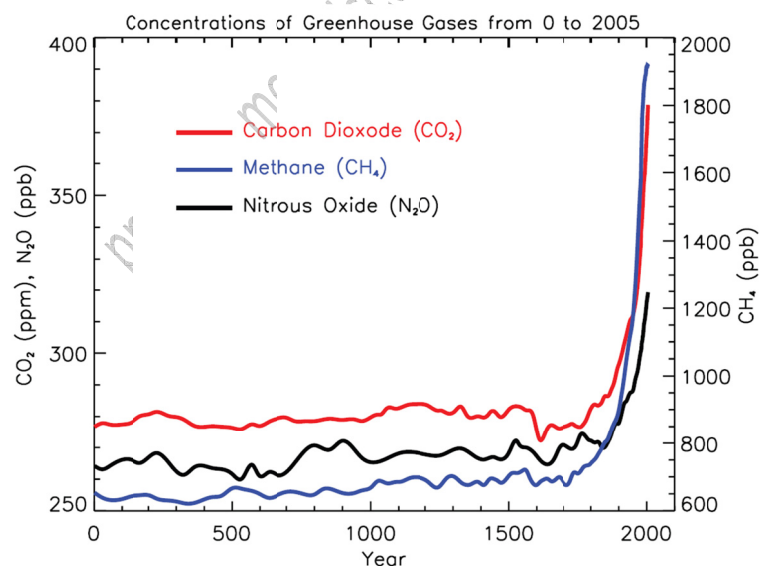


Figure 1.2: Atmospheric concentrations of important long-lived greenhouse gases over the last 2,000 years. Increases since about 1750 are attributed to human activities in the industrial era. Source: Forster *et al* 2007.

The increase in CO_2 and other greenhouse gases (GHGs) atmospheric concentration, modified the radiative climate forcing (i.e. the rate of energy change per unit area of the globe as measured at the top of the atmosphere; Rockström *et al* 2009) leading to changes in the balance between the incoming and outgoing energy to the earth's surface, thus altering the climate on Earth. In

particular, the Intergovernmental Panel on Climate Change (IPCC) reports that the global mean surface temperature has risen by $0.74 \pm 0.18^\circ$ over the last 100 years (1906–2005), precipitation has generally increased over land at latitude higher than 30°N over the period 1900 to 2005 while downward trends have been dominating the tropics since the 1970s, substantial increases in heavy precipitation events frequency have been found, droughts have become more common especially in the tropics and subtropics since the 1970s and a widespread reduction in the number of frost days at mid-latitude has been registered, an increase in the number of warm extremes and a reduction in the number of daily cold extremes have been observed in 70 to 75% of the land regions where data were available (Trenberth *et al* 2007). However, assuming a general increase in the mean global temperature, the regional and local effects of climate change are still a matter of debate. As an example, Sheffield *et al* (2012) found out that the previously reported increase in global drought is overestimated due to the simplicity of the indexes used in large-scale drought assessments.

Since temperature affects almost all aspects of terrestrial C processes, increasing Earth's surface temperature likely enhances ecosystem C fluxes, potentially feeding back to a build-up of atmospheric CO_2 concentration (Luo 2007). In fact, coupled climate-carbon cycle models predict a substantial decrease in the efficiency of the Earth system (land and ocean) to absorb anthropogenic CO_2 in consequence of climate warming (Heimann and Reichstein 2008). However, the rather wide range of results from the different model simulations show that there is still an high uncertainty in the estimation of the climate-C cycle feedback.

Actually, model projections predict an increase by another 1.1-6.4 $^\circ\text{C}$ of the global mean surface temperature by the end of this century, more intense, more frequent and longer lasting heat waves, an increase in daily minimum temperatures, a decrease in frost days, an increase in precipitations in the tropics and at northern latitudes, a decrease in precipitations in the subtropics, more frequent and intense precipitation events and droughts, a reduction in ice and snow cover, ocean acidification and sea level rise by 0.18-0.59 m (Meehl *et al* 2007).

Since these climate modifications are supposed to influence strongly the biosphere, it is very important to monitor and study the C dynamics across all C cycle compartments. In the last decades there has been a considerable increase in C cycle research in order to study all the processes and the interactions that build up the global C cycle that led to many advances, especially considering the effects of climate change on C dynamics (e.g. Knorr *et al* 2005, Davidson and Janssens 2006, Frank *et al* 2010, Beer *et al* 2010, Mahecha *et al* 2010), however we are not yet able to fully understand the complexity of the C cycle and to foresee its behaviour in a changing world. This is the main challenge for the next years.

1.2 Climate extremes

In addition to a general increase in temperature, it has been predicted a change in the geographical distribution, intensity and frequency of extreme events (Mitchell *et al* 2006). There is not a single definition of climate extremes, however there are three criteria often used to classify an event as extreme (Beniston *et al* 2007): *rarity* (low frequency/rate occurrence), *intensity* (large magnitude deviations from the normality) and *severity* (events that result in large socio-economic losses).

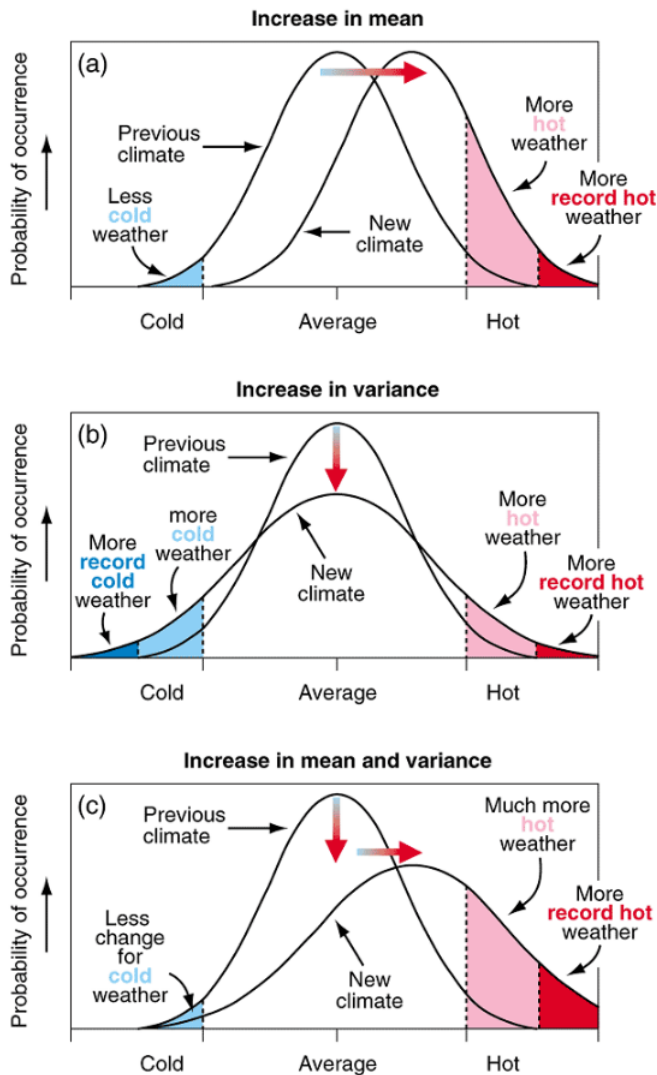


Figure 1.3: Schematic showing the effect on extreme temperatures when (a) the mean temperature increases, (b) the variance increases, and (c) when both the mean and variance increase for a normal distribution of temperature. Source Folland *et al* 2001.

Finally, the mean, the variance and even the symmetry of the frequency distribution may change at the same time (Figure 1.3c) leading to an alteration in the occurrence of extreme events in several different ways (Meehl *et al* 2000).

Looking at climate extremes from a statistical point of view, if there is a simple shift in the distribution of a climate variable (e.g. increase in the mean temperature, Figure 1.3a), there will be an increase in the probability of occurrence of the extreme events at one end of the distribution and a decrease at the other. Moreover, the frequency of climate extremes changes non-linearly with the change in the mean of a distribution, hence even a small variation in the mean can result in a large change in the frequency of extremes (Mearns *et al* 1984).

An increase in variability without a change in the mean implies an increase in the probability of extremes at both ends of the frequency distribution (Figure 1.3b). Consequently, a change in the variance will have a larger effect on the occurrence of extremes than a change in the mean (Katz and Brown 1992).

Extreme climatic events affect our economies and human health, as well as the productivity and C balance of terrestrial ecosystems, and different key parameters of our climate system are already moving beyond the patterns of natural variability (Soussana *et al* 2010). Current climate models consistently project increases in several climate extremes, such as an increase in the frequency and magnitude of hot extremes on the whole globe, an increase in the frequency of heavy precipitation events, as well as an increase in drought occurrence in some regions. These events have been shown to influence the biosphere's C balance, which in turn may accelerate climate change

1.3 The role of terrestrial ecosystems

The terrestrial biosphere interacts strongly with the climate, providing both positive and negative feedbacks due to biogeophysical and biogeochemical processes (Denman *et al* 2007). Terrestrial ecosystems influence the climate mainly through CO₂ exchange between the atmosphere and terrestrial biota (Figure 1.4), which is one of the prime links in the global C cycle (Holmén 1992). However, terrestrial ecosystems could also interact with climate through other ways (e.g. changing the surface albedo or interacting with the water cycle).

Various processes influence the C flow between terrestrial biota and the atmosphere, but the most important is the CO₂ uptake by plants through photosynthesis. Globally, this flow amounts to 120 PgC yr⁻¹. The amount of C fixed through photosynthesis is called Gross Primary Production (GPP). As around 50% of this C is returned back to the atmosphere through respiration (Ciais *et al* 1997), the net C uptake by plants is the Net Primary Production (NPP), which represents the storage of organic matter in excess of autotrophic plant respiration. This last flux can be divided into above-ground (R_a) and below-ground plant respiration (R_b):

$$NPP = GPP - (R_a + R_b)$$

Global terrestrial NPP has been estimated to be around 56-60 PgC yr⁻¹ (Field *et al* 1998, Saugier *et al* 2001). Also most of the C allocated in new plant tissues returns back to the atmosphere after their death as CO₂ through heterotrophic respiration (R_h). This is due to the decomposition of the dead organic matter (litter) by bacteria and fungi, which forms several soil C pools with different C content, chemical and biological composition and lifetime. The difference between NPP and R_h is called Net Ecosystem Productivity (NEP), and represents the net exchange of C between the atmosphere and the ecosystem. The NEP is by convention equal to the Net Ecosystem Exchange (NEE) but opposite in sign, since NEP is a measurement of the net gain (or loss) of C in an ecosystem over a period of time, while NEE is an instantaneous measurement of the inward and

outward flows of C within an ecosystem and, as a micro-meteorological variable, is considered positive when there is a C gain to the atmosphere:

$$NEP = NPP - R_h = -NEE$$

Global annual NEP has been quantified between 3 and 10 PgC (Watson *et al* 2000, Cramer *et al* 2001, Prentice *et al* 2001, Grace 2004, Schaphoff *et al* 2006), even though is characterized by an high uncertainty, due particularly to soil processes (Grace 2004), and by considerable inter-annual variability (Valentini *et al* 2000). When also accounting for C losses due to fires, land-use change, harvest, and erosion (i.e. disturbance; D), the total C flux is called Net Biome Productivity (NBP):

$$NBP = NEP - D$$

NBP represents the amount of C subtracted from the atmosphere and effectively stored in terrestrial ecosystems over long time periods. Recent estimates quantified this flux at 0.9 ± 0.6 PgC yr⁻¹ for the years 2000-2005 (Denman *et al* 2007). Since NBP has been positive for many decades, although fluctuating (Houghton 2003, Grace 2004, Van Minnen 2008), we can conclude that terrestrial ecosystems are acting as a net C sink. Climate variability and disturbances may alter NEP or NBP influencing directly GPP, R_a, R_b, NPP and R_h. Moreover, the C balance of terrestrial ecosystems could also be altered by climate change induced modifications, such as increased length in growing season, CO₂ and N fertilization, or by changes in land use and management.

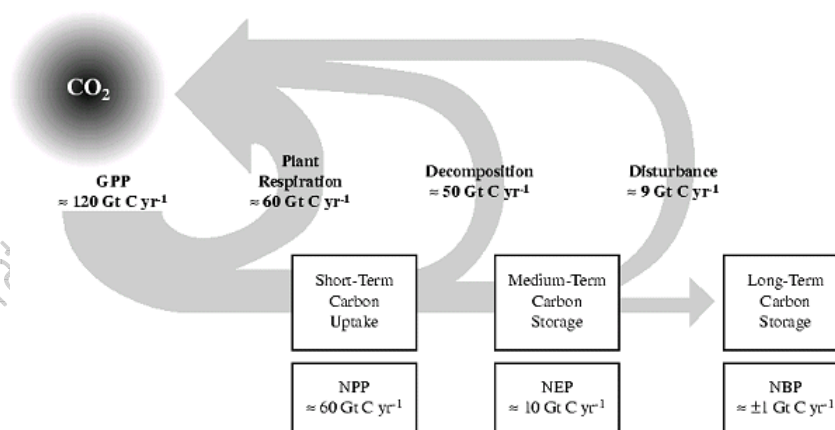


Figure 1.4: Schematic representation of the global terrestrial C uptake. Source: Watson *et al* 2000.

Considering that terrestrial biosphere stores about 2200-2500 PgC, and that only 466-654 PgC of which are in plant tissues, we can conclude that the most considerable C pool of terrestrial biosphere is represented by soil (1567-2011 PgC for the top 1 m; Prentice *et al* 2001). Carbon in soil is in the form of organic and inorganic carbon (i.e. carbonates). Organic carbon enters the soil

through litterfall, that is part of the NPP (leaves, twigs, small branches and roots, exudates), and together with the microbial biomass and dead microbes forms the soil organic matter (SOM), which has been estimated to be between 1220 and 1576 PgC for the 0-100 cm soil layer (Tarnocai *et al* 2009). Conceptually, several soil carbon pools can be distinguished: detritus and microbial biomass have a short turnover time (<10 yr), while modified soil organic carbon has decadal to centennial turnover time. Moreover, a small fraction of SOM is converted into inert forms (stable or recalcitrant; Prentice *et al* 2001). SOM is a key component of any terrestrial ecosystem, and any variation in its abundance and composition has important effects on many of the processes that occur within the ecosystems (Batjes 1996). In fact, litter is decomposed by soil microorganisms who release CO₂ back to the atmosphere through their respiration. This microbial respiration (R_h) represents the heterotrophic component of soil respiration (R_s = R_b + R_h), which accounts for 30-80% of total ecosystem respiration (R_e = R_a + R_s) in forests (Luo and Zhou 2006), while root/ rhizosphere respiration can account for as little as 10% to greater than 90% of total in situ soil respiration depending on vegetation type and season of the year (Hanson *et al* 2000). Thus, since R_s is the second largest carbon flux after GPP in most ecosystems (Davidson *et al* 2002), it accounts for a large portion of total biosphere respiration, and at the global scale, it releases C at a rate that is more than one order of magnitude larger than anthropogenic emissions (Luo and Zhou 2006). Thus, soil organic carbon and its related fluxes play a determining role in the regulation of the C cycle and consequently of the climate system.

The drivers that affect both the above- and below-ground processes of the C cycle in terrestrial ecosystems can be classified as (Denman *et al* 2007): (1) direct climate effects (such as changes in precipitation, temperature and radiation regime); (2) atmospheric composition effects (CO₂ fertilization, nutrient deposition, damage by pollution); and (3) land use change effects and management (deforestation, afforestation, agricultural practices, and their legacies over time). Changes in the environmental conditions could alter the C balance of terrestrial ecosystems, either increasing the C uptake in the terrestrial biosphere or decreasing it. The net result depends on the combination and intensity of the individual environmental conditions, however it is often difficult to quantify the final net result for the entire C cycle (van Minnen 2008).

1.3.1 Forests

The total forest area has been estimated at about 4.16×10^9 ha, which is equivalent to about 30% of total world land (Fischlin *et al* 2007). However, world forest area is declining, mainly due to deforestation. In particular, the total net loss of forest area in the period 2000-2010 has been quantified at 5.2×10^6 ha yr⁻¹, but the current annual net loss is decreasing in comparison to 1990s as a result of both a decrease in the deforestation rate and an increase in the area of new forest

established through planting or seeding and natural expansion of existing forests, especially at Northern latitudes (FAO 2010).

It has been estimated that deforestation and degradation of forest ecosystems in tropical and sub-tropical regions cause about 25% of anthropogenic CO₂ emissions (Houghton, 2003). Nevertheless, according to Dixon *et al* (1994), forest ecosystems store up to 80% of all above-ground and ~40% of all below-ground terrestrial C. In detail, forest contain 359 PgC in tree biomass and 787 PgC in forest soils, and more recent estimates are even higher (1,640 PgC above- + below-ground; Sabine *et al* 2004). Luysaert *et al* (2010) evaluated all the principal C fluxes of European forests, estimating a GPP equal to 899–1199 gC m⁻² yr⁻¹, a NPP at 439–574 gC m⁻² yr⁻¹, an R_h at 287–527 gC m⁻² yr⁻¹, a NEP at 19–200 gC m⁻² yr⁻¹ and a NBP at 63–89 gC m⁻² yr⁻¹ (different estimates depending on the used measuring method). For these reasons, forests are one of the main determinants of the global C cycle and are major contributors to the terrestrial C sink (Canadell and Raupach 2008, FAO 2010, McMahon *et al* 2010) with an estimated net sink of 1.1 ± 0.8 Pg C yr⁻¹ (Pan *et al* 2011). Therefore changes in such a sink may have a considerable impact on atmospheric CO₂ concentrations (WBGU 1998).

Generally, forest ecosystems are expected to be a sink for atmospheric CO₂, however high spatial heterogeneity and temporal variability of terrestrial C pools and fluxes, as well as natural and anthropogenic disturbances and environmental changes, lead to large uncertainties in estimates (Mund 2004). In particular, even if in the last decades some effects of the climate change (CO₂ and N fertilization, lengthening of the growing season, warmer temperatures) combined with changes in management may have raised the C uptake capacity of forest ecosystems in some biomes (Ciais *et al* 2008), other effects (such as warmer soil temperatures, more frequent droughts and heat waves, and changes in disturbance regimes and fires) may have led to a decrease in the CO₂ storage capacity of forests.

In addition, forests have proved to be susceptible to all types of climate extremes, especially droughts (Ciais *et al* 2005), which may decrease transpiration, gross photosynthesis and respiration of trees (Granier *et al* 2007) and have been identified as a major contributing factor in the recently accelerated rates of tree decline and mortality in European forests (Bréda *et al* 2006). Other types of climate extremes could potentially influence forest ecosystems by causing damages, increasing tree mortality or leading to indirect effects (e.g. increase the probability of fires or of pathogens outbreaks).

1.3.2 Croplands

World agricultural land, defined as land occupied by arable land, permanent crops and pastures occupies about 34% of total world area (5.02×10^9 ha; Smith *et al* 2007). During the last four

decades, agricultural land has gained almost 500×10^6 ha from other land uses, largely in the developing world. Because of its scale and intensity, agriculture has had a significant impact on the atmospheric GHG content (Salinger 2007) and it presently accounts for about 25% of the CO_2 , 50% of the CH_4 and 70% of the N_2O anthropogenic emissions (Hutchinson *et al* 2007).

Regarding the role of croplands in the global C cycle, there are contrasting opinions, even though agricultural ecosystems have been generally considered as C sources (Jenssens *et al* 2003, Loubet *et al* 2011). However, according to a recent synthesis by Ciais *et al* (2010), croplands may act either as a small C sink or small C source. In particular, cropland C fluxes has been quantified at $879\text{--}1360 \text{ gC m}^{-2} \text{ yr}^{-1}$ (GPP), $482\text{--}846 \text{ gC m}^{-2} \text{ yr}^{-1}$ (NPP), $299 \text{ gC m}^{-2} \text{ yr}^{-1}$ (R_h) and $34\text{--}193 \text{ gC m}^{-2} \text{ yr}^{-1}$ (NEP) (different estimates depending on the used method). The mean cropland NBP has been estimated ranging from -8.3 ± 13 to $-13 \pm 33 \text{ gC m}^{-2} \text{ yr}^{-1}$ (mean of the models and inventories data, respectively). Thus, the carbon sequestration potential of croplands should be considered as modest but non-negligible (Hutchinson *et al* 2007).

Similarly to forests, also for croplands the warmer climate and the fertilization effect due to the increased atmospheric CO_2 concentration would theoretically lead to an increase in crop productivity and yields, but the technological improvements in breeding, pest and disease control, fertilization and mechanization, also make identifying climate-change signals difficult (Hafner 2003). Other climate change aspects led to detectable effects on agriculture, such as the lengthening of the growing season, the increase in soil evaporation rates and the phenological effects (i.e. the advancing of the germinations and flowering dates), that have an impact on management practices (e.g. the advancing of the sowing and harvesting date; increased need for irrigation). Moreover, a change in climatic conditions could have an effect also on the occurrence of pest diseases and on the emergence of weeds.

Also extreme events can have a deep impact on croplands, both via negative impacts on plant physiological processes and direct physical damage, as well as by affecting the timing and conditions of field operations. Similarly to forests, also for croplands drought is the most severe extreme event affecting plant growth and productivity, especially if it occurs in connection with high temperatures. The effect of extreme high temperature is still uncertain, while frost, heavy rains and storm events could affect the agro-ecosystem carbon balance by damaging crops.

1.4 The influence of management

1.4.1 Forests

Management can influence the carbon balance of forest ecosystems (Thornley and Cannell 2000, Eggers *et al* 2007). The conversion of primary forests or old-growth forests to plantations or managed forests leads to a significant reduction of C pools in the living tree biomass and in the

dead aboveground biomass. On the contrary, the impact of forest management on the soil C pool depends on many site-specific factors and is often overridden by the high spatial variability of SOC pools in forest soils. Consequently, general effects on SOC are evident only with the most intensive practices (e.g. clear cuttings; Mund 2004).

Thus, since management could have a deep impact on forest C dynamics, changes in forest use and management can have a considerable impact also on atmospheric CO₂ concentrations. Hence, good forest management practices could play a key role in climate change mitigation, as acknowledged in the Articles 3.3 and 3.4 of the Kyoto Protocol, under which countries have to account for the loss or gain of carbon due to deforestation, afforestation and reforestation practices and may choose to include forest management activities to enhance the forest C sink strength and account it within the emission reduction target.

According to Lindner and Karjalainen (2007) the mitigation strategies in forest management could be grouped in 3 categories:

1. *Conservation management*: aims to protect the existing forest carbon pool to prevent emissions (e.g. reducing deforestation and disturbance);
2. *Sequestration management*: aims to increase the sequestration and storage of carbon in forest ecosystems or in wood products (e.g. increase forest productivity, afforestation);
3. *Substitution management*: aims to substitute carbon emissions from fossil fuels (e.g. utilization of woody biomass fuels).

The most effective forest mitigation strategy in the short-term is the conservation management, but also enhancing C sequestration may have important effects in the mid-term. Substitution management is the only C mitigation strategy, which offers also long-term mitigation potentials because each time, when fossil fuels are substituted by the use of renewable woody biomass, less carbon is emitted to the atmosphere compared to the present baseline.

However, several factors determine the C sequestration potential of forest ecosystems by means of forest management, such as soil carbon stock, soil characteristics, species composition, disturbance regime, rotation length and management operations. Since these factors differ between forests, specific local management options have to be found, even if, at least for European forests, a drastic change in the current sink should not be expected (Nabuurs *et al* 2008).

1.4.2 Croplands

Croplands have a very high biomass turnover rate and most of the C fixed by plants (i.e. NPP) is converted into food. Thus, SOC is the principal C pool of agro-ecosystems. According to the meta analysis by Guo and Gifford (2002), the soil C stock declines by 59% after land use change from pasture to cropland and by 42% from native forest to cropland. Similarly, VandenBygaart *et al* (2003) estimated that there is an average loss of $24 \pm 6\%$ of SOC when a native soil is converted to agricultural land. Consequently, it has been estimated that one fifth of CO₂ increase in the last 150 year was due to land use change from forest-grassland to cropland (Lal *et al* 1998). Furthermore, in the last decades agro-ecosystems have been losing most of their soil C stock due to farming practices intensification (Kucharik *et al* 2001).

Changes in agricultural management can potentially increase the accumulation rate of SOC, thereby sequestering CO₂ from the atmosphere. In particular, a study from West and Post (2002) found that a change from conventional tillage to no-till management can, on average, sequester $57 \pm 14 \text{ g C m}^{-2} \text{ yr}^{-1}$, while enhancing rotation complexity can sequester on average $20 \pm 12 \text{ g C m}^{-2} \text{ yr}^{-1}$. Moreover, other factors concerning crop management have an influence on the C dynamics of agro-ecosystems, such as irrigation, organic or inorganic fertilizers, fallow periods length, the residue management. As an example, Lal and Bruce (1999) estimated that the C sequestration potential through judicious management of world cropland includes: $0.08 \pm 0.12 \text{ PgC yr}^{-1}$ by erosion control, $0.02 \pm 0.03 \text{ PgC yr}^{-1}$ by restoration of severely degraded soils, $0.02 \pm 0.04 \text{ PgC yr}^{-1}$ by reclamation of salt-affected soils, $0.15 \pm 0.175 \text{ PgC yr}^{-1}$ by adoption of conservation tillage and crop residue management, $0.18 \pm 0.24 \text{ PgC yr}^{-1}$ by adoption of improved cropping system.

Other recent issues suggest that an effective positive contribution to global C balance by new farming practices is possible (Freibauer *et al* 2004, Smith 2004) and, as for forests, also in croplands there are the same three possible ways for climate change mitigation through management (ECCP 2003):

1. *Conservation management*: aims to reduce the soil disturbance in order to decrease soil C losses and to protect the existing soil C stock (e.g. no-tillage, reduced-tillage, set-aside land, growth of perennial crops);
2. *Sequestration management*: aims to increase the C inputs to agricultural soils (e.g. better use of organic amendments, irrigation and fertilization, improved crop rotations, livestock management);
3. *Substitution management*: aims to switch from conventional arable agriculture to other land-uses with higher C inputs or reduced disturbance (e.g. bioenergy crop production, conversion of arable land to grassland or woodland, natural regeneration, extensification, organic farming and integrated farm management).

Each of these practices has an estimated contribution to climate change mitigation up to almost 2 tC ha⁻¹ yr⁻¹ leading to an overall biological potential for C storage in European (EU15) cropland of 90–120 MtC yr⁻¹ (Smith 2004).

1.5 Thesis outline

After a brief description of the main measurement techniques used to quantify C fluxes in terrestrial ecosystems (Chapter 2), the thesis is composed of three research papers concerning the effects of management and climate extremes on C dynamics in forests and croplands.

The first paper (Chapter 3) examines C dynamics of the two main forest categories of the Friuli Venezia-Giulia Region (i.e. beech and spruce forests) using a chronosequence approach coupled with model simulations. For each chronosequence, the influence of stand age and applied management on the C pools and fluxes was assessed. This work was inspired by the lack of knowledge regarding the C stocks and fluxes of long-term managed forests in the Italian Alps, despite mountain environments are considered to be the most susceptible ecosystem to climate change effects. Moreover, there is still a debate whether forest management (i.e. thinnings, harvest) has an impact on soil C stock or if this pool remains unchanged despite the disturbance. Since it is an hard task to quantify precisely forest soil C fluxes, we used a model approach to investigate the biomass and NEP trends immediately after the final harvesting in order to determine if and for how long a forest becomes a C source due to human disturbance.

The second paper (Chapter 4) investigates the effects of crop management on the C dynamics in croplands. During the last decades, the intensification of farming practices increased crop productivity, however this has led to a general decrease of soil C stocks. Hence, there is a need to apply conservative farming practices with a positive effect on the C budget of croplands, such as conservation agriculture (CA). In this context, we performed a field experiment aimed at investigate the effects of CA (minimum tillage + cover crops) on the C dynamics of a 5-year crop rotation.

The third paper (Chapter 5) focuses on the soil respiration in croplands. Since soil respiration is a critical parameter in the regulation of the global C balance, even a small change in R_s can seriously alter the global C balance. In particular, an increase in R_s due to global warming might bring to a rise in CO₂ concentration that could enhance the greenhouse effect. In particular, an increase in R_s could bring to a positive feedback loop to reinforce climate warming, and this led to a growing concern about the effects of global change on soil respiration. Our work in this field of research regarded the response of the heterotrophic component of soil respiration to warm and cold extreme events during late winter-early spring in a soybean crop. Our choice was motivated by the fact that usually manipulation experiments are conducted over natural ecosystems and little

is known about the consequences of climate change on the carbon balance of croplands, despite they represent a huge part of the world area.

Finally, the overall conclusions are reported in Chapter 6.

Tesi di dottorato di Giuseppe De Simon, discussa presso l'Università degli Studi di Udine

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2

Methods

Summary

In order to assess all C fluxes in a terrestrial ecosystem, two different approaches can be used: i) direct quantification of C stock changes with time through repeated inventories, long term experiment (i.e. permanent plots) or through space-for-time substitutions (i.e. chronosequences); ii) micrometeorological techniques (i.e. eddy covariance and continuous soil respiration monitoring) to directly measure in- and out- ecosystem C fluxes. As the three chapters that follow are based on already published or submitted papers, a deeper description of the different methods adopted in the present thesis are reported in this section. In particular, advantages and limits of the chronosequence approach to study C dynamics in forest ecosystems are analyzed and a full description of measurement principles and calculation is reported for eddy covariance and soil respiration.

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2.1 The chronosequence approach

Chronosequences are the most used indirect approach to study long-term vegetation dynamics, assuming that spatial and temporal variations are equivalent (space-for-time substitution; Pickett 1989, Fukami and Wardle 2005). The chronosequence approach has also provided significant insight into the patterns and mechanisms of many aspects of the terrestrial C cycle in different ecosystems (Thuille and Schulze 2006, Alberti *et al* 2008, Smittenberg *et al* 2012), even though it has some limitations that involve the use of time as a surrogate for the past environment and prior system status (Pickett 1989) and the possible presence of other confounding factors.

Walker *et al* (2010) defined a chronosequence as a set of sites formed from the same parent material or substrate that differs in the time since they were formed. In forest sciences, a chronosequence is a set of forested sites that share similar attributes but are of different ages (Johnson and Miyanishi 2007) and can be considered a time sequence (Salisbury 1952, Pickett 1989). The key assumptions of chronosequences are that each of the sites representing different developmental stages had the same initial conditions and has traced the same sequence of changes. As mentioned before, the validity of this assumption is highly unlikely (Johnson and Miyamishi 2007). However, when there are demonstrable linkages between stages (i.e. the successional trajectory is predictable), chronosequences provide a useful approach to study temporal changes with time frames of 1–100 years (Walker *et al* 2010).

Concerning with C cycle, the chronosequence approach, if applicable, allows to estimate ecosystem C fluxes, such as NPP and, with proper soil sampling, changes in soil C content. However, in order to derive the NEP, an estimation of R_h is required. Thus, if R_h measurements are not provided, it is necessary to assume that there is an equilibrium between ecosystem inputs and outputs (Raich and Nadelhoffer 1989, Van Tuyl *et al* 2005). This assumption can be made if no changes are detected in soil carbon stocks across the chronosequence.

In the present thesis, a chronosequence approach has been used to investigate the effect of age and disturbance (i.e. seed cut) on C dynamics (Chapter 3), as schematically represented in Figure 2.1. For this study, we assumed that differences in ecosystem properties across sites were solely due to differences in time after disturbance because soil and climate condition were very similar among sites. Thus, the long-term effects of age and management were estimated through changes in the ecosystem C pools over time.

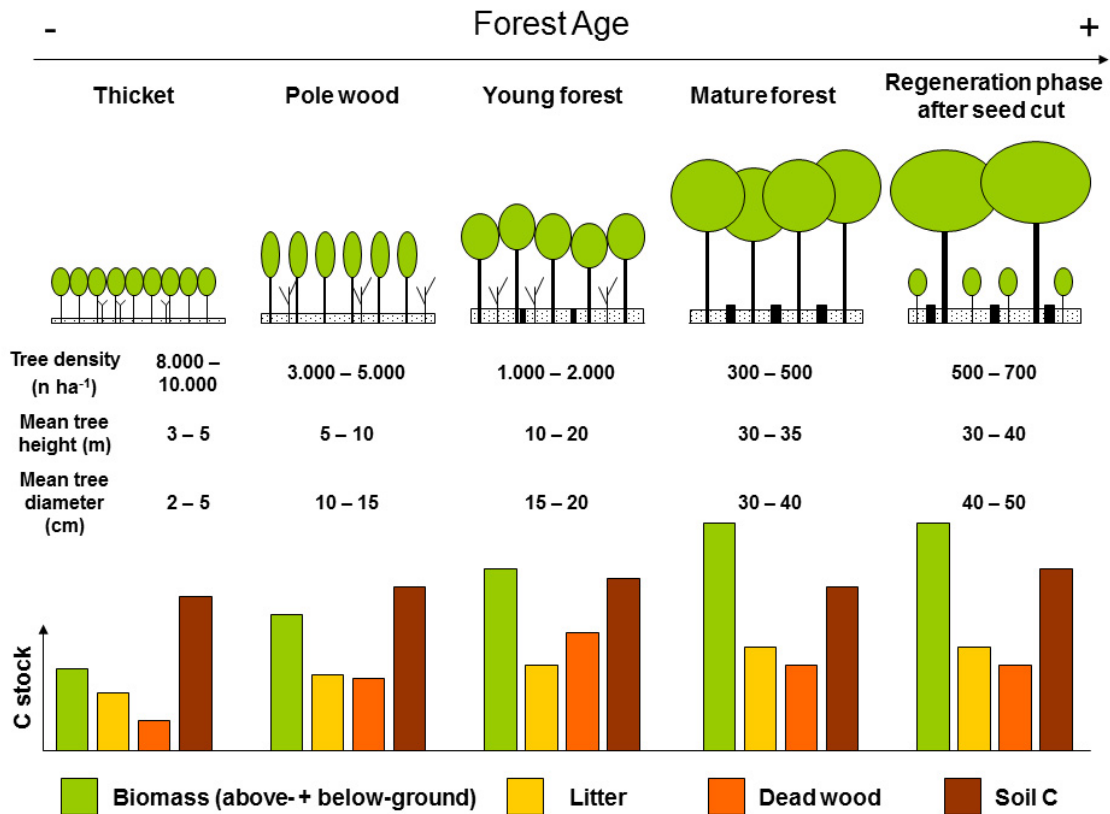


Figure 2.1: Conceptual scheme of a chronosequence for a managed forest with data about the main tree characteristics and forest C stocks.

2.2 Flux measurements

2.2.1 Eddy Covariance

2.2.1.1 Eddy Covariance principles

The Eddy Covariance technique (EC) is one of the most direct and defensible ways to measure heat, water and CO₂ fluxes (Burba and Anderson 2010). Since the atmosphere contains turbulent motions of upward and downward moving air, EC samples these turbulent motions to determine the net difference of material moving across the canopy-atmosphere interface (Baldocchi 2003).

In physical terms, "eddy flux" is computed by measuring how many molecules, moles, or milligrams of gas went up with upward wind movement at one moment and how many went down with downward wind movement in the next moment. In mathematical terms, "eddy flux" (F) is computed as the covariance between the instantaneous deviation in vertical wind speed (w') from the mean value (\bar{w}), the instantaneous deviation in gas mixing ratio (s') from its mean value (\bar{s}) and the instantaneous deviation in air density (ρ_a') from its mean value ($\bar{\rho}_a$):

$$F = \overline{(\rho_a + \rho_a')(\bar{w} + w')(\bar{s} + s')}$$

When EC is used over flat and vast spaces air density fluctuations can be assumed negligible as well as mean vertical flow, leading to:

$$F \approx \overline{\rho_a w' s'}$$

Thus, the eddy flux is approximately equal to mean air density multiplied by the mean covariance between deviations in instantaneous vertical wind speed and mixing ratio. By analogy, sensible heat flux is equal to the air density (ρ_a) multiplied by the specific heat of air (C_p) and by the covariance between deviations in instantaneous vertical wind speed and temperature (T'), while latent heat flux is computed in a similar manner using the latent heat of vaporization of water (λ) and the instantaneous deviation in water mixing ratio (e'):

$$H = \rho_a C_p \overline{w' T'}$$

$$LE = \lambda \frac{M_w / M_a}{P} \rho_a \overline{w' e'}$$

Since instruments usually do not measure gas mixing ratio (s), there is yet another assumption in the practical formulas (Burba and Anderson 2010) that brings to a CO₂ flux (F_c) defined as the mean covariance between deviations in instantaneous vertical wind speed and density of CO₂ in the air (ρ_c'):

$$F_c = \overline{w' \rho_c'}$$

In addition, there are other important assumptions in the EC method:

- Measurements at a point are assumed to represent an upwind area;
- Measurements are assumed to be done inside the boundary layer of interest, and inside the constant flux layer;
- Fetch and footprint are assumed adequate, so flux is measured only from the area of interest;
- Flux is fully turbulent.

Moreover, since instruments record flux at certain measurement height, concentration could build up or decrease below this measurement height. Thus, in order to evaluate total CO₂ flux, the *storage* term ($F_{storage}$) must be calculated from the temporal changes in CO₂ concentration profile (Aubinet *et al* 2001):

$$F_{storage} = \frac{P_a}{RT_a} \int_0^{h_m} \frac{dc(z)}{dt} dz$$

where P_a is the atmospheric pressure, R the molar gas constant, T_a the air temperature, h_m is the maximum measurement height above-ground level (a.g.l.), c is the CO₂ concentration, t is time and z is height a.g.l.. This term has to be added to F_c to come up with final net ecosystem exchange (NEE), especially in case of low wind, stable stratification, high canopies, or when air mixing is significantly reduced. With respect to CO₂, the storage term is small over short crops while it is an important quantity over taller forests. The storage term value is greatest around sunrise and sunset when there is a transition between respiration and photosynthesis and between the stable nocturnal boundary layer and daytime convective turbulence. However, summed over 24 hours, the storage term is assumed to be zero.

2.2.1.2 Eddy Covariance implementation

The EC is, to a large extent, a site-specific method. In fact, the entire process of experimental design, implementation and data processing should be tailor-made for a specific purpose at specific site (Burba and Anderson 2010).

The application of EC method involves issues related to site selection, instrument placement, sampling duration and frequency, calibration and post-processing. In particular, the typical EC workflow is divided into three consecutive parts: design of the experiment, implementation and data processing. The key elements of the design portion of EC experiments are: setting the purpose and variables for the study, deciding on the instruments and hardware to be used, creating new or adjusting existing software to collect and process data, establishing appropriate experiment location and a feasible maintenance plan to avoid data gaps. The major elements of the implementation portion are: placing the tower, placing the instruments on the tower, testing data collection and retrieval, collecting data, and keeping up the maintenance schedule. The processing portion includes: processing the real time, “instant” data, processing averaged data, quality control, and long-term integration and analysis. The main elements of data processing include: converting voltages into units, de-spiking, applying calibrations, rotating the coordinates, correcting for time delay, de-trending if needed, averaging, applying corrections, quality control, filling in the gaps, integrating, and finally, data analysis and publication (Burba and Anderson 2010).

An EC flux system (usually named as “tower”) is composed of a sonic anemometer to measure 3D wind components at high frequency, an infrared gas analyzer (IRGA) to measure CO₂ and water vapour concentrations, and a device for real time data acquisition and processing. There are

two IRGA designs: closed-path and open-path. In a closed-path system, air from the vicinity of the sonic anemometer is drawn through a sampling tube to the IRGA, where the CO₂ and H₂O concentration of the sampled air is measured. In an open-path system, instead, air can pass freely between the IR source and detector of the IRGA. Thus, in order to measure the gas concentration fluctuations, it is essential that the open-path IRGA is located close to the anemometer. The choice of an open-path versus a closed-path sensor is largely a function of power availability and frequency of precipitation events (Burba and Anderson 2010). Generally, EC systems are coupled with auxiliary sensors that measure environmental parameters, such as short-wave and long-wave radiation (incoming and outgoing), net radiation, photosynthetic active radiation, soil heat flux and soil respiration measurements.

Ideally, the field site should be flat, with an extensive footprint of uniform vegetation. The height of the sensors depends on the height of the vegetation, the extent of footprint, the range of wind velocity and the frequency response of the instruments, which usually are set between 10 and 20 Hz in order to ensure complete sampling of the high frequency portion of the flux co-spectrum. The footprint is the area upwind from the tower, such that fluxes generated in this area are registered by the tower instruments, while the term fetch usually refers to the distance from the tower when describing the footprint.

There are different models to evaluate footprint contribution from the given distance (Burba and Anderson 2010). For near neutral conditions, one of the reasonably simple yet descriptive models is the one by Schuepp *et al* (1990). This model calculates the Cumulative Normalized contribution to Flux measurement (CNF) from analytical solutions of the diffusion equation for near-neutral conditions:

$$CNF(x_L) = - \int_0^{x_L} \frac{U(z-d)}{u^* kx^2} e^{-\frac{U(z-d)}{u^* kx}} dx = e^{-\frac{U(z-d)}{u^* kx_L}}$$

where x_L is the distance from the station (m), U is the integrated wind speed (m s⁻¹) z is the measurement height (m), u^* is the friction velocity (m s⁻¹), d is zero plain displacement (m) and k is von Karman constant (0.4). From these variables the model computes how much of the measured flux comes from what distance.

2.2.1.3 Eddy Covariance data processing

After instantaneous data have been collected, they must be processed in order to get the flux calculations as close as possible to the reality of what's actually happening in the field (Burba and Anderson 2010). This data processing include: converting signals from voltages to physical units;

de-spiking (remove data spikes due to both electronic and physical noise); applying calibration coefficients if needed; rotating coordinates (correct for the possible non perpendicular w axis of the anemometer to the mean flow/wind streamlines); correcting for time delays (compensation for time delay in signal acquisition from different instruments); de-trending if needed and finally instantaneous data averaging. The sampling duration must be long enough to capture low frequency contributions to flux covariances, but not too long to be affected by diurnal changes in temperature, humidity and CO₂. Adequate sampling duration and averaging period usually vary between 30 and 60 minutes.

After instantaneous data have been averaged, the mean fluxes must be corrected. According to Webb *et al* (1980), the Webb-Pearman-Leuning (WPL) term is used to compensate for the fluctuations of temperature and water vapour that affect the measured fluctuations in CO₂ and H₂O. The WPL term is usually relatively small during the growing season and relatively large during the off-season, when it could reach values several times the actual flux (Burba and Anderson 2010). The WPL corrections for CO₂ and H₂O fluxes measured with open-path analyzers are:

$$F_c = F_{c_0} + \mu \frac{E}{\rho_a} \frac{q_c}{1 + \mu \frac{\rho_v}{\rho_a}} + \frac{H}{\rho C_p} \frac{q_c}{T_a}$$

$$E = (1 + \mu \frac{\rho_v}{\rho_a}) (E_0 + \frac{H}{\rho C_p} \frac{q_c}{T_a})$$

where F_c is the WPL corrected CO₂ flux, F_{c_0} is the uncorrected CO₂ flux, E is the WPL corrected H₂O flux, E_0 is the uncorrected H₂O flux, H is sensible heat flux, q_c is the mean CO₂ density, ρ_a is dry air density, ρ_v is water vapour density, ρ is total air density, C_p is the specific heat of air, T_a is air temperature and μ is the ratio of molar masses of air to water. For closed path instruments, the last portion of the equation (sensible heat flux portion) is usually not used, because temperature fluctuations are assumed to be attenuated in the sampling tube (if the tube is longer than 50-100 cm depending on the internal diameter; Burba and Anderson 2010).

Then, for open-path analyzers, the instrument surface heating correction according to Burba *et al* (2006) is applied. This correction is necessary, especially in cold environments, since the surface of an open-path instrument becomes substantially warmer than ambient due to electronics and radiation load during daytime. This effect often leads open-path CO₂ analyzers to measure an apparent ecosystem CO₂ uptake during off-season periods, especially in cold climates. The correction assumes some resultant expansion of the air by a sensible heat flux created by

instrument surface heating (H_I). Such a flux can be estimated from the temperature gradient using resistance approach (Monteith 1963):

$$H_I = \frac{\rho C_p (T_s - T_a)}{r_a}$$

where T_s is the instrument surface temperature and r_a is the aerodynamic resistance. For an instrument oriented near vertically, only a fraction of H_I would affect the measurement because the wind will remove much of the warmed air from the optical path, carrying away most of the sensible heat without affecting the measurements. However, a portion of the warmed and expanded air will remain in the boundary layer, reducing the measured air density and influencing the measurement. The fraction (fr) of H_I retained in the optical path (H_P) can be expressed as follows:

$$H_P = fr H_I$$

This term has to be added to sensible heat (H) in the WPL correction, leading to a new formulation of the corrected flux (F_{c-new}):

$$F_{c-new} = F_c + fr \frac{(T_s - T_a) q_c}{r_a T_a} \left(1 + \mu \frac{\rho_v}{\rho_a}\right)$$

After the application of these corrections, data are controlled in order to remove bad data due to instrument malfunctions, processing/mathematical artefacts, ambient conditions not satisfying EC method, winds not from the footprint of interest or heavy precipitation. Generally, algorithms and protocols are implemented in order to get to a quality assessment and quality check analysis (QA/QC). Usually, the protocols used by different EC groups/networks present these features:

- ranges of tolerance established for each variable;
- data outside tolerance ranges removed or flagged;
- precipitation events flagged;
- U, u^* , and/or stationarity tests conducted;
- non-stationary periods removed.

However, quality control is a site- and instrument-specific activity, thus a specific protocol for each site and/or instrument must be implemented.

Once collected data are verified and corrected, measured fluxes must be validated. One way to validate EC fluxes is to construct an energy budget for the study site. The general equation used for the energy budget is (Burba and Anderson 2010):

$$R_n + H + LE + G \approx 0$$

where R_n is the net radiation, H is sensible heat flux, LE is latent heat flux and G is the sum of soil heat flux and soil heat storage. These key components must sum to zero as required by conservation of energy. However, after wide application of the EC method including all corrections, it became obvious that the energy balance at the earth's surface could not be closed with experimental data (Foken and Oncley 1995). Generally, for many field experiments and also for the CO₂ flux networks, a closure of the energy balance of approximately 80% was found. This unclosed energy balance is not related to errors in the EC technique but to atmospheric phenomena which cannot be measured with this technique (Foken *et al* 2012).

After data validation, a data gap inventory and a data gap-filling procedure have to be applied to obtain daily fluxes. Currently, there are several gap-filling techniques based on a wide range of approaches including interpolation, probabilistic filling, look-up tables, non-linear regression, artificial neural networks, and process-based models in a data-assimilation mode (Moffat *et al* 2007). One of the most recent methods to gap-fill eddy covariance and meteorological data has been proposed by Reichstein *et al* (2005). This method identifies three different conditions: 1) only the data of direct interest are missing, but all meteorological data are available; 2) also air temperature or vapour pressure deficit (VPD) is missing, but radiation is available, and 3) also radiation data is missing. In the first case, the missing value is replaced by the average value under similar meteorological conditions within a time-window of ± 7 days. Similar meteorological conditions are present when global radiation (R_g), air temperature (T_a) and VPD do not deviate by more than 50 W m^{-2} , $2.5 \text{ }^\circ\text{C}$, and 5.0 hPa , respectively. If no similar meteorological conditions are present within the time window, the averaging window is increased to ± 14 days. In the second case, the same approach is taken, but similar meteorological conditions can only be defined via R_g deviation less than 50 W m^{-2} and the window size is first not further increased. In the third case, the missing value is replaced by the average value at the same time of the day ($\pm 1 \text{ h}$), i.e. by the mean diurnal course. In this case, the window size starts with ± 0.5 days (i.e. similar to a linear interpolation from available data at adjacent hours). If after these steps the values could not be filled, the procedure is repeated with increased window sizes until the value can be filled.

2.2.1.4 Eddy Covariance flux partitioning

Despite the increasing popularity of the EC technique to assess ecosystem C exchange (Baldocchi 2003), micrometeorological techniques are only able to obtain the total net CO₂ efflux (NEE) and cannot partition total efflux into its individual sources (Buchmann 2002). For this reason, several

flux-partitioning algorithms that aim to separate the observed NEE into gross primary production (GPP) and total ecosystem respiration (R_e) have been developed (Hollinger *et al* 1994, Falge *et al* 2002, Law *et al* 2002, Papale and Valentini 2002, Reichstein *et al* 2002, Gilmanov *et al* 2003, Rambal *et al* 2003, Reichstein *et al* 2005). These algorithms could be classified into 2 categories: i) those that use only night-time data for the estimation of R_e and ii) those that use daytime data or both day- and night-time data using light response curves. Also in this case, one of the newest algorithms has been proposed by Reichstein *et al* (2005), which derives a short-term temperature sensitivity of R_e from eddy covariance data and applies this to the extrapolation from night to daytime. This short-term temperature sensitivity parameter (E_0) was estimated with the exponential regression model by Lloyd and Taylor (1994):

$$R_e = R_{ref} e^{E_0(1/(T_{ref}-T_0)-1/(T-T_0))}$$

where the regression parameter T_0 is kept constant at -46.02 °C, the reference temperature (T_{ref}) is set at 10 °C. and T could be either air or soil temperature. For the estimation of E_0 , the data set is divided into short sub-periods (windows) of 15 days and the regression has to be performed for each window. The $x+1$ window is shifted 5 days with respect to window x . The window size of 15 days has been selected to be sufficiently short to avoid strong confounding seasonal effects and to be long enough to provide enough data and an adequate temperature range for the regression. Then, a E_0 parameter representative for the whole year could be estimated either as the average of the three E_0 estimates that yielded the lowest standard errors or as the average of all E_0 estimates with the inverse of the standard error as weighing factor.

After the temperature sensitivity has been estimated, the temperature independent level of respiration (R_{ref}), has to be quantified. Since this parameter varies with time in an ecosystem, it has to be estimated for consecutive 4-day periods by nonlinear regression using the same Lloyd and Taylor (1994) model, fixing all parameters except R_{ref} . After these parameters are estimated for each period, they are linearly interpolated between the estimates. This procedure results in a dense time series of the Lloyd & Taylor (1994) parameters R_{ref} and E_0 , the latter being constant. Consequently, for each point in time (t), an estimate of R_e can be provided according to:

$$R_e(t) = R_{ref}(t) e^{E_0(1/(T_{ref}-T_0)-1/(T(t)-T_0))}$$

Then, for each period, GPP is derived by the difference between NEE and R_e :

$$GPP = NEE - R_e$$

2.2.2 Soil respiration

As said before, the EC method is only able to partition NEE into GPP and R_e . In order to know which is the contribution of soil compartment to R_e , it is necessary to measure R_s , which may be also partitioned into its autotrophic (R_b) and heterotrophic (R_h) component (Figure 2.2).

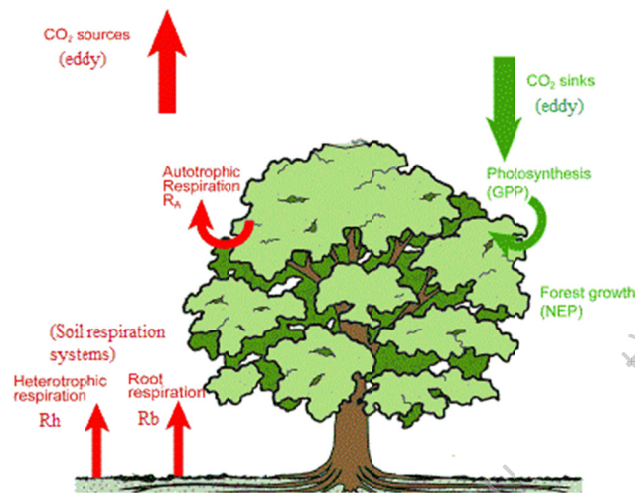


Figure 2.2: Main fluxes of terrestrial ecosystems and the measurement techniques associated.

Soil CO₂ efflux can be measured with several different chamber techniques. According to the classification by Livingston and Hutchinson (1995), there are three major chamber techniques used to measure R_s : i) non-steady-state non-through-flow chamber (also known as closed static chamber); ii) non-steady-state through-flow chamber (closed dynamic chamber) and iii) steady-state through-flow chamber (open dynamic chamber). In non-steady-state chambers, of both the through-flow and non-through-flow types, the R_s is determined from the rate of concentration increase in an isolated chamber, that has been placed on the soil surface for a known period of time. In steady-state chambers, instead, R_s is calculated from the difference between CO₂ concentration at the inlet and the outlet of the chamber (Pumpanen *et al* 2004).

No single method has been established as a standard, however the most common systems use closed dynamic chambers (Figure 2.3), such as the Li-Cor 8100 Automated Soil CO₂ Flux System (Li-Cor, Lincoln, NE, USA) and the soil respiration system described by Delle Vedove *et al* (2007).

When using this type of chambers, it is necessary to avoid the wind induced pressure difference between inside and outside the chamber through a pressure vent. This vent should allow static pressure changes inside the chamber to follow whatever static pressure changes occur in the surrounding air outside the chamber, both in calm and windy conditions, while remaining insensitive to wind direction (Xu *et al* 2006).

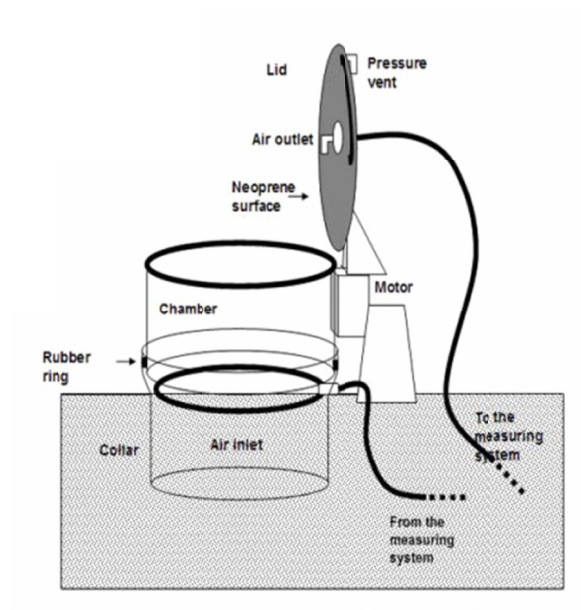


Figure 2.3: A drawing of a closed dynamic chamber shown in the open position. Source: Delle Vedove *et al* (2007)

The measuring principle of the closed dynamic uses the rate of increase of CO₂ within the chamber to estimate the rate at which CO₂ diffuses into free air outside the chamber (Figure 2.4).

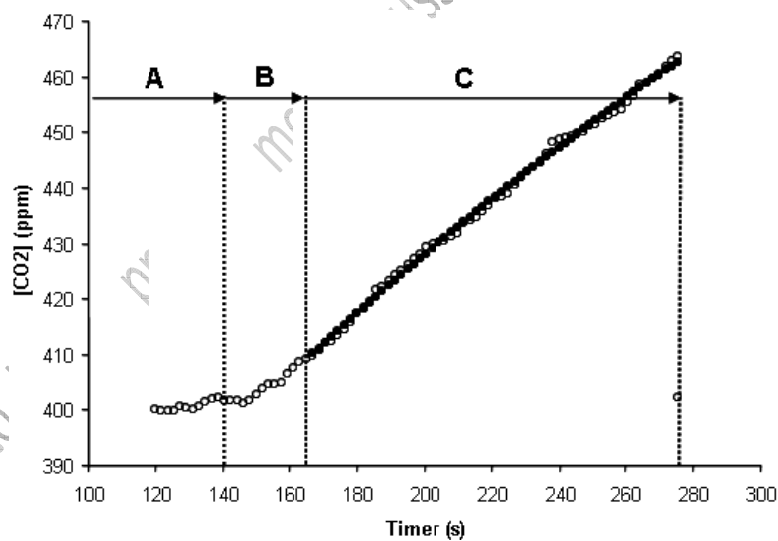


Figure 2.4: Example of closed dynamic chambers measuring principle: chamber closure and measure of the CO₂ concentration target (A); mixing (B); non-linear regression computation (C); final efflux computation at the end of the process. Open symbols are measured CO₂ concentrations; closed symbols are derived values from the computed non-linear regression.

To minimize the underestimation of the efflux due to the alteration of the diffusion gradient, a non-linear curve fitting (Davidson *et al* 2002; LI-COR 2004) can be used. In this case, when a

steady chamber mixing is established after closure, a non-linear regression between CO₂ concentration and time is performed according to the equation:

$$C(t) = C_x - (C_x - C_0) e^{-a(t-t_0)}$$

where $C(t)$ is the CO₂ concentration corrected by the water mole fraction, and C_x , t_0 and a are estimated regression's parameters, C_0 is the initial concentration at chamber closure, C_x is the asymptote, and a is the parameter which defines the curvature (positive if $C_x > C_0$ or negative otherwise), t_0 represents the time when $C(t)$ is equal to C_0 . The regression coefficients C_x , t_0 and a are calculated using a non-linear regression by a modified Gauss Newton iterative method. The initial rate of change in CO₂ (dC/dt) at C_0 and $t=t_0$ is computed using the equation:

$$dC/dt = a (C_x - C_0) e^{-a(t-t_0)}$$

assuming $t=t_0$:

$$dC/dt = a (C_x - C_0)$$

Then, soil CO₂ efflux ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) is computed using the following equation:

$$SR = \frac{V}{S} \cdot \frac{dC}{dt} \cdot \frac{P_0}{R \cdot (T_0 + 273.15)}$$

where P_0 and T_0 are the air pressure and air temperature measured as average of the last ten measure before lid closure; R is the universal gas constant ($8.31 \text{ J mol}^{-1} \text{ K}^{-1}$), V is system volume (cm^3) and S is chamber basal area (cm^2).

2.2.2.1 Soil respiration partitioning

As stated in paragraph 1.3, R_s is the sum of R_b and R_h . Thus, not all the CO₂ coming from soil is produced by SOM decomposition, and vegetation may contribute strongly to the total CO₂ efflux by root (R_r) and rhizomicrobial respiration (R_{mr}). Hence, the contribution of vegetation to R_s strongly limits the use of total CO₂ efflux to determine whether soil is a net source or a net sink of CO₂ and to estimate C balance in various ecosystems (Kuzyakov 2006).

For this reason, various methods allowing the separation of autotrophic and heterotrophic soil respiration have been developed over the last few decades. These methods have been recently reviewed by Kuzyakov (2006), who separated them into two main categories: isotopic and non-isotopic techniques. The principles and the main advantages and disadvantages of each method are reported in Table 2.1a and 2.1b.

One of the simplest and economic partitioning methods is a non-isotopic technique defined as “root exclusion”. This method consists in a comparison between planted and unplanted soil, assuming the root-derived CO₂ ($R_r + R_{mr}$) as the difference between soil respiration measurements in root-affected and root-free soils. Existing root exclusion techniques may be categorized into three broadly defined areas (Hanson *et al* 2000): i) root removal – roots are removed, soil is placed back in reverse order of removal, and further root growth is prevented by barriers; ii) trenching – existing roots are severed by trenching at a plot boundary but not removed, and a barrier is installed to inhibit future root growth and iii) gap analysis – aboveground vegetation is removed from relatively large areas and R_s measurements in the gap are compared to R_s data for a vegetated area.

The root-exclusion approach has been criticized since the water regime and the temperature balance in planted and unplanted soil could be very different (Fisher and Gosz 1986, Ross *et al* 2001). Moreover, the presence of vegetation affects also the cycling of nutrients (such as N which interacts with the C cycle) and the decomposition of SOM and plant residues, which is dependent on direct biological effects due to the presence of living roots (Kuzyakov 2006). In particular, the CO₂ evolved as a consequence of the rhizosphere priming effect may be 2–3 times larger than that of basal respiration (Cheng *et al* 2003). Thus, the review by Kuzyakov (2006) concludes that this approach cannot be accepted as suitable for the estimation of SOM-derived CO₂ from vegetated soils.

However, as an example, this method is still successfully adopted in cropland experiments. In this kind of environment trenching could be used as it causes low soil disturbance and there are not differences between trenched and non-trenched plots in water regime during the growing season (at least in irrigated croplands where soil is kept near field capacity by irrigation). Moreover, outside the growing season, trenched plots are removed as all soil respiration is represented by R_h .

Method	Principle	Advantages	Disadvantages
Non-isotopic methods			
<i>Root exclusion</i>	Difference of CO ₂ from planted and unplanted soil	Simple, cheap, various ecosystems	Different C, N and H ₂ O budget, different microbial activity in planted vs. unplanted soil
<i>Trenching</i>	Difference of CO ₂ from planted soil without and with trenched roots	Simple, cheap, mainly forest and shrubs	Dead root decomposition should be considered; different H ₂ O and nutrient budget, different microbial activity in trenched vs. non-trenched soil
<i>Gap formation</i>	Difference of CO ₂ from soil with vegetation and from the soil under the gap	Natural formation of gaps, mainly for forest	Different C, N and H ₂ O budget, different microbial activity in the soils under gap and trees/different SOM decomposition; differences in above litter decomposition
<i>Shading</i>	Difference of CO ₂ from soil with non-shaded and shaded plants	Simple, cheap, mainly grasslands and crops; mainly natural N uptake	Contribution of decomposition of previous rhizodeposits; R _r and R _{nr} are not completely eliminated; different H ₂ O budget, different microbial activity in shaded vs nonshaded soil/different SOM decomposition
<i>Clipping, clear felling</i>	Difference of CO ₂ from soil with non-clipped and clipped plants	Simple, cheap, various ecosystems	Contribution of decomposition of previous rhizodeposits; dead root decomposition should be considered; R _r and R _{nr} are not completely eliminated; different H ₂ O budget, different microbial activity under clipped vs. non-clipped plants/different SOM decomposition
<i>Tree girdling</i>	Interruption of C flows into roots by phloem girdling, difference of CO ₂ from soil under non-girdled and girdled trees	Suitable for forest ecosystems, natural H ₂ O regime and N uptake	Forest ecosystems only; dying of trees after girdling, contribution of dying roots to CO ₂ ; different C and N budget and microbial activity under girdled and non-girdled trees/different SOM decomposition
<i>Regression technique</i>	Linear relation between root mass and root-derived CO ₂	Simple, cheap, various ecosystems	High number of replications is necessary; overestimation of root-derived CO ₂ when big and old roots are present; frequently poor correlation between root amount and total CO ₂ ; SOM-derived CO ₂ could be biased by far extrapolation of the regression line
<i>Component integration</i>	Manual separation of CO ₂ sources and measuring CO ₂ from each source	Separation of 3 (or 4) CO ₂ sources, cheap, various ecosystems	Strong CO ₂ flush after disturbance and root injuries; laborious; different CO ₂ decrease after the flush/dependence of the results on the CO ₂ trapping period
<i>Excised roots</i>	Measuring of respiration of excised roots separated from soil	Cheap, useful for roots of various plants	Strong CO ₂ flush after the disturbance and root injuries; different CO ₂ decrease after the flush/dependence of the results on the CO ₂ trapping period; extrapolation on field conditions is inexact; SOM-derived CO ₂ is estimated as difference to the total CO ₂ efflux
<i>In situ root respiration</i>	Respiration of living roots placed in a cuvette located in the soil	Respiration of living roots under field conditions	Extrapolation on field conditions is inexact; SOM-derived CO ₂ is estimated as difference to the total CO ₂ efflux; water and nutrient supply in the cuvette is necessary

Table 2.1a: Non-isotopic methods allowing partitioning of total CO₂ efflux from soil into root-derived CO₂ and SOM-derived CO₂ (Source: Kuzyakov 2006).

Method	Principle	Advantages	Disadvantages
Isotopic methods			
<i>Continuous labeling of shoots in $^{14}\text{CO}_2$ or $^{13}\text{CO}_2$ atmosphere</i>	Growing plants in $^{14}\text{CO}_2$ or $^{13}\text{CO}_2$ atmosphere and dilution of root-derived CO_2 by unlabeled CO_2 from SOM	The only one fully correct and exact method	Maintenance of a constant specific ^{14}C activity or $^{13}\text{C}:^{12}\text{C}$ ratio during long time period; temperature and air moisture control in the chamber; only laboratory application, special equipment is necessary; isolation of below-ground atmosphere is necessary
<i>Pulse labeling of shoots in $^{14}\text{CO}_2$ or $^{13}\text{CO}_2$ atmosphere</i>	Short-time labeling of shoots in $^{14}\text{CO}_2$ or $^{13}\text{CO}_2$ atmosphere C tracing of $^{14}\text{CO}_2$ or $^{13}\text{CO}_2$ from the soil	Easier than continuous labeling, recently assimilated C is mainly considered	Uneven distribution of labeled C between plant organs and C fluxes including CO_2 ; calculations consider mainly the recently assimilated C; calculation of root-derived CO_2 is based on many assumptions; isolation of below-ground atmosphere is necessary
<i>^{13}C natural abundance</i>	Growing C4 plants on a C3 soil or vice versa C tracing of $\delta^{13}\text{C}$ in CO_2 from the soil	Reasonable alternative for continuous labeling	Only for some soil-plant combinations applicable; not precise; high variation of $\delta^{13}\text{C}$ in CO_2 ; contamination with air CO_2 should be considered
<i>Free air carbon dioxide enrichment (FACE)</i>	Application of additional CO_2 with changed $\delta^{13}\text{C}$ signature compared to air C tracing of $\delta^{13}\text{C}$ of CO_2 from the soil	Suitable for various ecosystems, wide range of plants	Expensive (additional CO_2); correct only for the first year after FACE start; not precise; high variation of $\delta^{13}\text{C}$ in CO_2
<i>Radiocarbon dating of soil CO_2</i>	Different turnover rates of SOM and recent plant-derived C flows	Suitable for various ecosystems	Weighted age ($\Delta^{14}\text{C}$) of SOM-derived CO_2 is necessary; can be biased by bomb- ^{14}C ; very CO_2 expensive (AMS analyses)
<i>Bomb-^{14}C</i>	Different turnover rates of SOM estimated by 'pulse labeling' of SOM by bomb-derived $^{14}\text{CO}_2$ and of root-derived CO_2	Suitable for various ecosystems	Weighted age ($\Delta^{14}\text{C}$) of SOM-derived CO_2 is necessary, control of bomb- ^{14}C -unaffected SOM is necessary; very expensive (AMS analyses)
<i>^{18}O of CO_2</i>	Different $\delta^{18}\text{O}$ of plant-derived CO_2 and soil water CO_2 and isotopic exchange between CO_2 and H_2O	-	Not applicable until now, many uncertainties

Table 2.1b: Isotopic methods allowing partitioning of total CO_2 efflux from soil into root-derived CO_2 and SOM-derived CO_2 (Source: Kuzyakov 2006).

2.3 Conclusions

As reported before, the most direct way to measure net ecosystem C exchange at the ecosystem level is the EC technique (NEP_{eddy}), however over longer time intervals and under some assumptions, NEP could be estimated also through the ecosystem C pools changes with time (NEP_{bio}). Moreover, it is possible to derive NEP by combining biometric and fluxes methods, i.e. subtracting the measured C losses via heterotrophic respiration from the biometric measurements of NPP ($NEP_{bio+flux}$). All this methods are subject to different error sources and uncertainties irrespective of the methodology applied (Luysaert *et al* 2009). The main characteristics of these three approaches are reported in Table 2.2.

NEP estimate	Costs	Complexity	Measurement uncertainty	Detail level	Minimum time resolution
Biometric	Low	Low	High	Plot	> = year
Biometric + fluxes	Intermediate	High	Intermediate	Plot	> = year
Micrometeorological	High	High	Low	Ecosystem	30 min

Table 2.2: Main characteristics of the different NEP estimation methods.

The main advantages associated with micrometeorological techniques are: i) the method is not destructive; ii) continuous measurements provided; iii) net C exchange data as output; iv) measurements on the entire ecosystem and v) wide datasets available from C fluxes networks. The main disadvantage are: i) the method is expensive; ii) complex implementation; iii) a maintenance plan is needed and iv) measurements need for turbulence and stationary conditions to obtain good data. On the contrary, biometric measurements are economic and simpler in comparison to flux measurements, however this approach could only provide seasonal or annual data and usually causes some disturbance to the measured C pools.

Several studies assessed the resulting differences in NEP estimated through the micrometeorological and the biometric + fluxes method, most of them concluded that there is a general agreement between these techniques (Curtis *et al* 2002, Black *et al* 2007, Gough *et al* 2008, Peichl *et al* 2010). However, these methods were not always complementary, since sometimes chamber-based estimates tend to be higher than EC measurements (Myklebust *et al* 2008). The cause of this bias is not understood, since there are several questions associated with both approaches, such as the general lack of energy balance closure suggests that EC may be underestimating NEE (Wilson *et al* 2002); EC measurements usually underestimate night-time CO_2 effluxes; large uncertainties in the up-scaling of chamber measurements; difficulties in the

determination of root systems biomass increment and long-term observations needed to observe changes in soil carbon content (Belelli Marchesini *et al* 2007).

Tesi di dottorato di Giuseppe De Simon, discussa presso l'Università degli Studi di Udine

Carbon stocks and net ecosystem production changes with time in two Italian forest chronosequences¹

Summary

Forest management influences several ecosystem processes, including carbon exchange between forest ecosystem and atmosphere. The aim of this paper was to study the carbon cycle over different age classes of two managed forests in the Italian Alps through direct measurements and modelling. For this purpose, ecosystem carbon dynamics of a beech forest (*Fagus sylvatica* L.) and of a spruce forest (*Picea abies* (L.) Karst.) were investigated using a chronosequence approach. In both forests, five forest development stages were identified (thicket, pole wood, young forest, mature forest and the regeneration phase) with an age spanning from 42 to 163 years for the beech forest and from 35 to 161 years for the spruce forest. Measured total ecosystem carbon stock increased up to 80-100 years, with a mean of 232 MgC ha⁻¹ in the beech forest and of 299 MgC ha⁻¹ in the spruce forest. Calculated net ecosystem production (NEP) was found to decrease linearly with age and had an average value of 2.2 and 4.4 MgC ha⁻¹ yr⁻¹ for beech and spruce forest, respectively. Model simulations reported an increase in NEP till 50-60 years followed by a decrease thereafter. The model also predicted a negative NEP for a short period (8-11 years) after the seed cut. Aboveground biomass was the main driver of carbon accumulation while soil carbon was not significantly influenced by both age and management system. Moreover, measured data and model showed that the applied shelterwood system allowed for a rapid recovery of the ecosystem after the disturbance (i.e. seed cut), bringing back forest to act as C sink in few years.

Keywords: shelterwood system, carbon dynamics, soil carbon, beech, spruce.

¹ De Simon G, Alberti G, Delle Vedove G, Zerbi G and Peressotti A (2012). Carbon stocks and net ecosystem production changes with time in two Italian forest chronosequences. *European Journal of Forest Research* 131:1297–1311

3.1 Introduction

It is well known that forest ecosystems play an important role in the global carbon cycle (Tans *et al* 1990, Walker and Steffen 1996, Hagedorn *et al* 2001): a large amount of carbon (C) is stored and exchanged by the different forest pools (Dixon *et al* 1994, Amthor *et al* 1998, Dopouey *et al* 1999, Luysaert *et al* 2010), and this means that forests could be very important for the mitigation of the climate change. In fact, there are evidences that terrestrial ecosystems have been acting as a C sink in the last decades (Goodale *et al* 2002, Nabuurs *et al* 2003): European forests are believed to offset 7-12% of continental anthropogenic emissions (Janssens *et al* 2003) and recent estimates assess that the net biome production (NBP) of EU-25 forests amounts to $0.75 \pm 0.20 \text{ MgC ha}^{-1} \text{ yr}^{-1}$ (Luysaert *et al* 2010). Furthermore, net absorption of CO₂ in European forests has been enhanced in the last fifty years (Ciais *et al* 2008). This increase in net primary production (NPP) could be due to several factors such as forest re-growth after abandonment of agricultural land (Hooker and Compton 2003, Alberti *et al* 2008), increase of atmospheric CO₂ concentrations, nitrogen depositions and changes in forest management (De Vries *et al* 2006, Magnani *et al* 2007, Ciais *et al* 2008, McMahon *et al* 2010).

It has also been observed that sustainable forest management may enhance carbon sequestration in comparison to unmanaged forests (De Vries *et al* 2006, Nunery and Keeton 2010, Thurig and Kaufmann 2010) and this has been acknowledged by article 3.4 of the Kyoto Protocol (UNFCCC 1997), which specifies that the effects of Forest Management (FM) may be accounted for in the national GHG emissions budget as an additional human-induced activity. This interest in mitigation potential of terrestrial ecosystems has led to some studies about carbon dynamics in different types of forests under different types of managing systems (i.e. Lindner and Karjalainen 2007, Jandl *et al* 2007). Previous papers have also identified some possible actions to maximize forest carbon pools (Nabuurs *et al* 2008) as countries are required to increase or at least maintain their forest C pools. However, there are still uncertainties in estimates because of spatial heterogeneity and temporal variability of carbon stocks and fluxes (WBGU 1998, IPCC 2000). In particular, the most important source of uncertainty in forest ecosystems carbon budget is linked to the dynamics of soil carbon, which generally represents a large part of the total ecosystem carbon stock (Schimel 1995). Previous studies did not find a unique response of soil carbon to forest age and disturbance: Covington (1981), for example, measured a decline in forest floor organic matter after clear cutting and attributed it to an increase in decomposition rates and to a decrease in litter inputs. Yanai *et al* (2003) observed that this trend could be driven also by other factors that normally are not taken into account, such as mixing of forest floor organic matter in the mineral soil during harvest operations and changes in logging treatment over time.

Observing carbon dynamics of a forest could be very challenging in the long term, but estimates are needed in order to achieve forest carbon budgets at any scale. Therefore, several researchers

tried to obtain information about temporal trends using chronosequences (space for time substitution). Even though this approach has been criticized because chronosequences are subject to errors if sites differ in respects other than their age (Yanai *et al* 2003), there is not another way to collect field data about forest ecosystem development in the short term.

In the Italian Alps there have been only a few researches based on chronosequences to study carbon dynamics in forest ecosystems, even though mountain and boreal environments are considered to be the most susceptible to climate change effects (Grabherr *et al* 1994, Beniston *et al* 1997, Dirnböck *et al* 2003). Most of these studies are about the influence of land use change (i.e. afforestation) on carbon stocks (Thuille *et al* 2000, Thuille and Schulze 2006, Alberti *et al* 2008), but there is still a lack of investigations aimed to quantify the carbon stocks and fluxes of long-term managed forests. For this reason, the principal aim of the present paper was to investigate the carbon dynamics in the two main forest categories of Friuli Venezia-Giulia Region: beech forests (*Fagus sylvatica* L.) and spruce forests (*Picea abies* (L.) Karst.). In particular, the specific objectives of this paper for both forests were: (i) to quantify carbon stocks and net ecosystem production (NEP), (ii) to quantify the influence of stand age and applied management on such stocks and fluxes, (iii) to model long term biomass and NEP trends improving a numerical model based on the carbon content variation of each ecosystem compartment with respect to spatial resolution and the environmental conditions of moderately managed Alpine forests. The main hypothesis of the present study is that shelterwood system is not influencing soil carbon stock in the medium-long term and that the disturbance represented by the regeneration cuts causes a short term release of carbon to the atmosphere (i.e. negative NEP).

3.2 Materials and Methods

3.2.1 Study sites and plots selection

The two selected study sites are located in Friuli Venezia-Giulia (North-East Italy). In this region, forests cover about 41% of the territory (323.832 ha; INFC 2006) and are mainly distributed in the northern part of the region, which is characterized by alpine and sub-alpine environments.

The first study site (beech chronosequence) is located in the south-eastern part of the Cansiglio plateau (46° 02' 30''N, 12° 24' 45''E). The elevation is about 1200 m a.s.l., mean annual temperature is 5.7 °C (1972-2001) with an absolute maximum of 33°C (August 1992) and an absolute minimum of -22°C (January 1985). Mean annual rainfall is 1800 mm with a maximum during autumn. Parent material is represented by limestone and soil pH is slightly basic. The most important forest type is represented by pure beech (*Fagus sylvatica* L.) forest with a usual rotation period of 140 years. Silver fir (*Abies alba* Mill.) is also sporadically present.

The second study site (spruce chronosequence) is located in the municipality of Forni Avoltri (46° 35' 13''N, 12° 48' 35''E). The elevation is about 1400 m a.s.l., mean annual temperature is 6.8 °C (1961-1990) and mean annual rainfall is 1400 mm. The bedrock is represented by silicate-flyschoid rocks and soil pH is slightly acid. Forest type is composed of spruce (*Picea abies* (L.) Karst.) and silver fir (*Abies alba* Mill.), while beech (*Fagus sylvatica* L.) is quite sporadic. The usual rotation period is 100-120 years.

Both forests are property of the Public Administration of Friuli Venezia-Giulia and are managed by the regional forest department according to a shelterwood system (Mund and Schulze 2006) characterized by two successive steps of cutting: a seed cut, which removes most of the biomass in order to provide seeds and proper light conditions for natural forest regeneration, and a final cut, which removes the remaining old trees. These harvests are performed over small areas (from thousands square meters to a few hectares) in order to not disturb and alter the climatic conditions of large portions of the forest and to promote a rapid ecosystem recovery. Within the rotation period, the applied management forecasts several thinnings that lead to even-aged stands with homogenous vertical and horizontal structure. According to this shelterwood system, it is possible to identify five forest development stages: (i) thicket, (ii) pole wood, (iii) young forest, (iv) mature forest, (v) regeneration phase after the seed cut.

In each forest category, these five development stages were recognized in order to build up a chronosequence. All the stands within each chronosequence had similar elevation, slope, aspect and soil characteristics (Table 3.1), but different age. In each stand, three circular 531 m² plots were set up for the further analysis according to the Italian National Forest Inventory methodology (INFC 2006). Although the plots were relatively small, their size was appropriate to capture each forest stand characteristics (i.e. species composition, diameter distribution, height) because of the internal stand homogeneity. The plots were selected and surveyed on July-August 2008 and on May 2009 for the beech and spruce forest, respectively. The main characteristics for each stage are reported in Table 3.2.

BEECH FOREST SOIL			SPRUCE FOREST SOIL		
Soil type		Leptosol	Soil type		Cambisol
Slope		33%	Slope		40%
Horizon	Depth (cm)	Description	Horizon	Depth (cm)	Description
Oe	0-15	Black (10YR2.5/1); organic material partially decomposed; abundant gravels with flat shape; common fine roots; non-calcareous, acid pH; abrupt boundary	Oi	0	Leaves not decomposed
A	15-25	Black (10YR2/2); sandy-loam; abundant gravels with flat shape; granular structure, weak and highly friable; common fine roots; non-calcareous; acid pH; abrupt boundary	Oe	0-2	Black (10YR2/2.5); organic residues little decomposed; abundant gravels; highly acid
R	25+	Calcareous rock; unknown boundary	A	2-10	Brown (7.5YR4/3); loam; abundant rounded gravels; subangular poliedric structure, thin, little distinguishable; abundant medium sized roots; acid; abrupt boundary
			Bh	10-24	Brown (7.5YR3/2); silty loam; abundant rounded gravels; angular poliedric structure, thin, partially distinguishable; abundant medium sized roots; acid; abrupt boundary
			Bt	24-54	Brown (7.5YR4/4); loam; abundant rounded gravels; angular poliedric structure, partially distinguishable; little medium roots; acid; clear smooth boundary
			2BC	54-120	Brown (7.5R4/4); sandy-loam; abundant rounded gravels; little fine roots; acid; unknown limit

Table 3.1: Soil properties of the studied chronosequences (beech and spruce forest). Soil type refers to the ISSS-ISRIC-FAO classification (1998).

BEECH	Stand development stage				
	Thicket	Pole wood	Young forest	Mature forest	Regeneration phase
Average age (years)	42 ± 5	61 ± 1	74 ± 7	143 ± 3	163 ± 0
Density of trees (stems ha ⁻¹)	8298 ± 1304	3751 ± 1161	1881 ± 359	370 ± 27	483 ± 138
Average height (m)	8.5 ± 2.6	9.1 ± 1.6	12.1 ± 0.5	30.6 ± 1.6	25.0 ± 4.7
Average diameter (cm)	5.2 ± 0.7	12.7 ± 2.9	20.7 ± 0.6	35.7 ± 1.3	22.6 ± 8.2
Standing C stock (MgC ha ⁻¹)	52.8 ± 12.3	93.6 ± 4.9	121 ± 2.2	172 ± 11.6	137.4 ± 20.2
Soil C stock (MgC ha ⁻¹)	82.0 ± 19.4	97.3 ± 11.1	114.4 ± 28.3	102.4 ± 10.1	129.5 ± 19.2

SPRUCE	Stand development stage				
	Thicket	Pole wood	Young forest	Mature forest	Regeneration phase
Average age (years)	35 ± 4	62 ± 7	87 ± 1	119 ± 8	161 ± 14
Density of trees (stems ha ⁻¹)	6499 ± 770	1255 ± 60	502 ± 66	389 ± 82	734 ± 136
Average height (m)	11.1 ± 0.7	22.5 ± 0.6	25.1 ± 6.7	22.6 ± 5.9	32.9 ± 1.2
Average diameter (cm)	11.0 ± 1.2	22.3 ± 0.7	20.6 ± 7.0	28.3 ± 10.2	37.4 ± 4.2
Standing C stock (MgC ha ⁻¹)	140.7 ± 9.3	227.2 ± 23.4	272.1 ± 5	287.3 ± 49.4	131.6 ± 82.6
Soil C stock (MgC ha ⁻¹)	47.7 ± 16.9	103.9 ± 11.8	60.7 ± 6.6	85.1 ± 11.4	69.2 ± 14.9

Table 3.2: Main characteristics of the studied stands (\pm standard error, $n = 3$).

3.2.2 Dendrometric measurements

3.2.2.1 Aboveground and belowground biomass

In each plot, all standing trees were recognized, numbered and their diameters at breast height (DBH, 1.30 m) were measured. Trees were surveyed within a circular plot of 13 m radius or 4 m radius depending on their DBH (DBH > 17.5 cm and DBH < 17.5 cm, respectively) (INFC 2006). A vitality index was assigned to each surveyed tree according to INFC methodology. Total height was measured on a subsample of ten trees and one wood increment core was taken with a Pressler borer up-slope from each of these trees (INFC 2006). In the field, all the cores were fixed to wooden supports and in the laboratory they were cleaned, using a razor blade, until annual rings could be clearly seen. In order to measure mean diameter increment (Δd ; cm yr⁻¹) of the last five tree rings and to build plot-specific regressions that predict Δd on current tree diameter, a DENDROTAB 2003 table (Walesch Electronic GmbH, CH) with a binocular microscope was

used. Data were collected and stored using T-Tools software (Walesch Electronic GmbH, CH). Once Δd was estimated for each plant, annual growth of bole, branch, and bark was calculated for each tree (NPP_{tree}) by difference between current and previous biomass using the allometric relationship described below.

Aboveground biomass of each tree was calculated using the following allometric relation based on DBH (Zar 1999):

$$\ln(M) = \ln(a) + b \times \ln(D)$$

in which M is the biomass (kg), D is the DBH (cm) and a and b are coefficients depending on wood density (species) and DBH according to the methodology proposed by Anfodillo *et al* (2006). We previously applied this methodology to National Inventory plots and compared calculated volumes with volumes derived from specific yield table available for spruce and beech in Friuli Venezia-Giulia. This comparison showed an overall underestimation of 6 and 5% for beech and spruce, respectively (data not shown).

Coarse root biomass (diameter > 5 mm) was calculated applying a coefficient of 0.50 and 0.26 to the aboveground biomass for the conifers and for the broad-leaved tree, respectively (Anderle *et al* 2002). Fine roots biomass (diameter < 5 mm) was considered equal to 2% of the aboveground biomass (Perruchoud *et al* 1999).

3.2.2.2 Litter and Leaf Area Index (LAI)

In order to quantify the litter carbon stock, the survey was divided in two steps: i) standing litter assessment and ii) annual leaf production ($NPP_{foliage}$).

Standing litter, defined as litter on the soil surface (Lf-horizon), was quantified collecting all the litter and twigs (diameter < 1 cm) within a randomly selected 1 m² subplot in each plot.

Leaf production in the beech chronosequence is equal to litterfall ($NPP_{foliage} = \text{litterfall}$) and was quantified using a circular litter trap with a diameter of 45 cm which was positioned in the centre of each plot and periodically emptied. All the collected material was dried at 70°C for 24 hour and then weighted. Beech Leaf Area Index (LAI) was estimated multiplying dry biomass by a Specific Leaf Area (SLA) equal to 21 m² kg⁻¹ (Bartelink 1997, Bouriaud *et al* 2003).

In the spruce chronosequence, LAI was calculated using the indirect method proposed by Jarvis and Leverenz (1983) and Law *et al* (2008) based on Beer's law using a coefficient of attenuation equal to 0.28. Below-canopy Photosynthetic Active Radiation (PAR) was measured using a ceptometer (Delta Devices – England) every meter along a East-West plot diameter (26 m). Incoming PAR was measured outside the forest every meter along a 26 m East-West transect and

was corrected for solar inclination angle. LAI was then converted to dry biomass assuming a SLA of $5 \text{ m}^2 \text{ kg}^{-1}$ (Hager and Sterba 1985).

Tree foliage ($\text{NPP}_{\text{foliage}}$) production was calculated as total overstory foliage biomass multiplied by the fraction of total foliage mass that grew that year, which was determined from the separation of needle cohorts on three branches collected on each plot.

Dry material was then ground to a fine powder and analyzed for total C and N in a CHN Elemental Analyzer (NA1500 Series 2, Carlo Erba Instruments, Rodano, Italy).

3.2.2.3 *Dead wood*

Standing dead trees were labelled during stand surveys. DBH and total height were recorded in order to calculate volume, which was then converted to biomass using the density values determined for the first woody debris decay class (see below). C and N concentrations were also assumed to be the same of this decay class.

In each 13 m radius plot, stumps volume was calculated by measuring average diameter and total up-slope height of each stump. A decay class was assigned to each stump according to the INFC methodology (INFC 2006).

Downed dead wood volume was quantified using the line-intercept method proposed by Harmon and Sexton (1996). Within each plot, linear transect was set up along one randomly selected diameter (26 m). The decay class and diameter of each deadwood fragment (diameter > 1 cm) crossing the line were registered. To convert volume to biomass, basic density (dry weight/fresh volume) was determined for each decay class collecting one sample for each class and transect. Fresh volume of the materials was measured by immersion in water before drying them at 102°C for 48 hours to measure dry weight. Samples were then reduced to sawdust and sub-samples were collected for C and N analysis. C and N concentrations were then multiplied for dry biomass to obtain total carbon and nitrogen stocks in this compartment.

3.2.2.4 *Soil*

Soil samples were collected around the centre of each plot. Eight soil cores were collected after removal of the litter layer at two depths (0-10 cm and 10-20 cm). Prior to processing, samples were kept at 4°C . Each sample was mixed and sieved through a 2-mm sieve and then dried at 102°C for 48 hours. Dried material was ground to a fine powder, treated with HCl 2:1 to remove carbonates and then analyzed for organic C and N. Average soil bulk density to a depth of 20 cm was estimated in the centre of each plot following the excavation method (Elliott *et al* 1999).

Because of the high density of stones at each site, stone coverage was estimated along a 50 cm wide and 26 m long transect.

Soil carbon content (MgC ha^{-1}) was then calculated using the following equation:

$$C = \frac{d \times 10000 \times \rho \times C_m \times (1 - S)}{1000}$$

where d is the horizon depth (0.20 m), ρ is the soil bulk density of fine soil particles (< 2 mm) (kg m^{-3}), C_m is the mean carbon content along soil profile (%) and S is the stone coverage (%).

3.2.3 Net Ecosystem Production (NEP)

The measured ecosystem carbon pools described above can be combined to estimate aggregate ecosystem fluxes such as ANPP (total aboveground net primary production), BNPP (total belowground net primary production), R_h (total heterotrophic respiration) and net ecosystem production (NEP) assuming that soil respiration is balanced annually with litterfall, gross root carbon allocation and any accumulation (or loss) of carbon in the roots and soil (Raich and Nadelhoffer 1989, Van Tuyl *et al* 2005). This equilibrium between inputs (litterfall) and output (heterotrophic respiration) can be made if no changes are detected in soil carbon stocks across the chronosequence. As such, measurements of soil respiration do not enter the calculations.

Thus, NEP was calculated according to the following equation:

$$NEP = \text{inputs} - \text{outputs} = ANPP + BNPP - Rh + \Delta C_{soil}$$

where:

- ANPP ($\text{MgC ha}^{-1} \text{ yr}^{-1}$) is the above ground net primary production calculated as the sum of net primary productivity of stem, branches and leaves:

$$ANPP = NPP_{tree} + NPP_{foliage}$$

- BNPP ($\text{MgC ha}^{-1} \text{ yr}^{-1}$) is the below ground net primary production calculated as the carbon stored in fine roots ($C_{\text{fine roots}}$; MgC ha^{-1}) multiplied by their turnover (0.4 yr^{-1} ; Thuille and Schulze 2006) plus the carbon annually stored in coarse roots derived by multiplying ANPP for 0.50 and 0.26 for the conifers and for the broad-leaves, respectively:

$$BNPP = (C_{\text{fine roots}} \times \text{turnover}) + \Delta C_{\text{coarse roots}}$$

- R_h is the heterotrophic respiration ($\text{MgC ha}^{-1} \text{ yr}^{-1}$) calculated according to the equation:

$$Rh = \text{litterfall} + a \times C_{\text{stumps}} + b \times C_{\text{woody_debris}} + c \times C_{\text{dead_trees}}$$

where C_{stumps} , $C_{\text{woody_debris}}$ and $C_{\text{dead_trees}}$ are the carbon stocks of stumps, woody debris and dead trees (MgC ha^{-1}), respectively. The decay coefficients (a , b and c ; yr^{-1}) were derived from Matthews (1997) for stumps and dead trees and from De Angelis *et al* (1981) for the woody debris;

- ΔC_{soil} is the variation in soil carbon content.

3.2.4 Modelling net ecosystem production

Net ecosystem production trend across the chronosequence was simulated through a numerical model based on the carbon content variation of each ecosystem compartment (Figure 3.1). The model considers two main compartments: i) aboveground and dead wood ii) soil. Annual ecosystem NPP and biomass are determined using a logistic function while dead wood and litterfall are derived from standing biomass using a mortality factor. Fresh organic matter mineralization is assumed to produce CO_2 and decomposition products which are added to the organic matter already present in the soil compartment. The CO_2 emitted following the fresh organic matter mineralization and the CO_2 emitted following the old organic matter degradation constitute heterotrophic respiration.

The model has a yearly time step and it allows to take into account disturbances (i.e. harvest, thinning) at any time during rotation period.

The model was initialized using standing biomass measured at each stand across each chronosequence and using the constants reported in Table 3.3. The management system applied to model simulations foresaw two harvests for beech forest (seed cut at 160 years and final cut at 170 years) and one for spruce forest (140 years).

A detailed description of model equations is reported in the appendix.

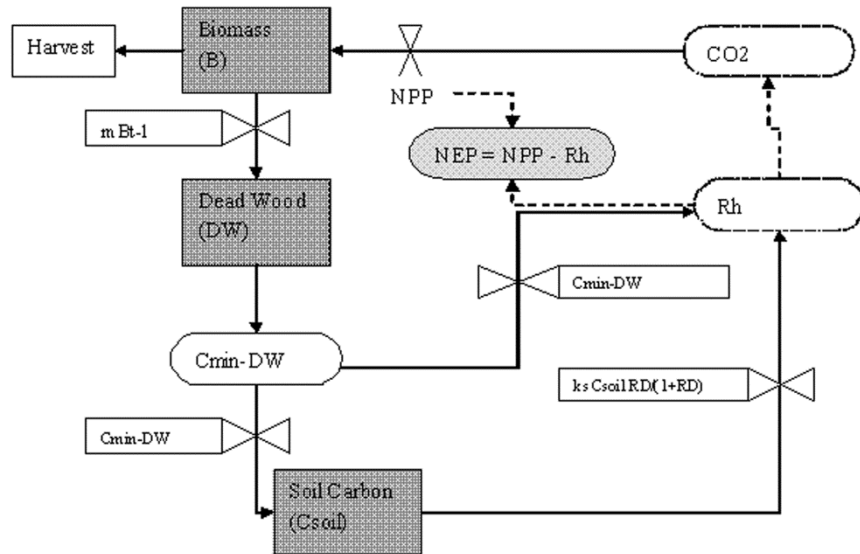


Figure 3.1: Model description.

		BEECH	SPRUCE
CLIMATE DATA			
Annual mean air temperature	°C	6.0	7.0
Annual mean precipitation	mm yr ⁻¹	1800	1400
LIVE BIOMASS			
Initial dry biomass	Mg ha ⁻¹	20	30
Maximum dry biomass	Mg ha ⁻¹	600	700
Initial growth rate (r ₀)	Mg ha ⁻¹ yr ⁻¹	0.25	0.25
Growth rate after harvest	Mg ha ⁻¹ yr ⁻¹	0.2	-
NECROMASS			
Initial necromass	Mg ha ⁻¹	10	10
Mortality ratio	Mg necromass : Mg standing biomass	0.04	0.015
Litter decomposition rate constant	year ⁻¹	0.30	0.36
Biomass C content	%	47%	47%
SOIL			
Clay	%	10%	15%
Carbonates	%	2%	0%
Organic carbon	%	15%	10%
Bulk density	g cm ⁻³	0.40	0.42
Soil depth	cm	20	20

Table 3.3: Constants used to run the model.

3.3 Results

3.3.1 Carbon Pools

3.3.1.1 Aboveground and belowground

The distribution of the ecosystem carbon through the chronosequence is given in Figure 3.2a and 3.2b for beech and spruce forest, respectively. The carbon content of total live biomass increased until mature forest phase and then decreased after the seed cut for both beech and spruce forest. Maximum values for this pool were $172.0 \pm 11.6 \text{ MgC ha}^{-1}$ and $287.3 \pm 49.4 \text{ MgC ha}^{-1}$ (\pm standard error; $n = 3$) for beech and spruce, respectively. Carbon content of standing dead trees (snags) in the beech forest had a maximum in the pole wood phase, when this value reached $16.9 \pm 13.0 \text{ MgC ha}^{-1}$ due to the high mortality caused by competition for light. In the spruce forest, this pool decreased with stand age from $4.7 \pm 0.3 \text{ MgC ha}^{-1}$ (thicket) to zero. In both forests there were not standing dead trees in mature forest stands, while in the regeneration phase stands carbon content of snags amounted at $0.1 \pm 0.1 \text{ MgC ha}^{-1}$ and $0.2 \pm 0.1 \text{ MgC ha}^{-1}$ for beech and spruce, respectively. Thus, the applied forest management system is likely to be the main driver of live biomass carbon stock trend, but has also an effect on standing dead trees due to thinning treatments that usually carry away the snags.

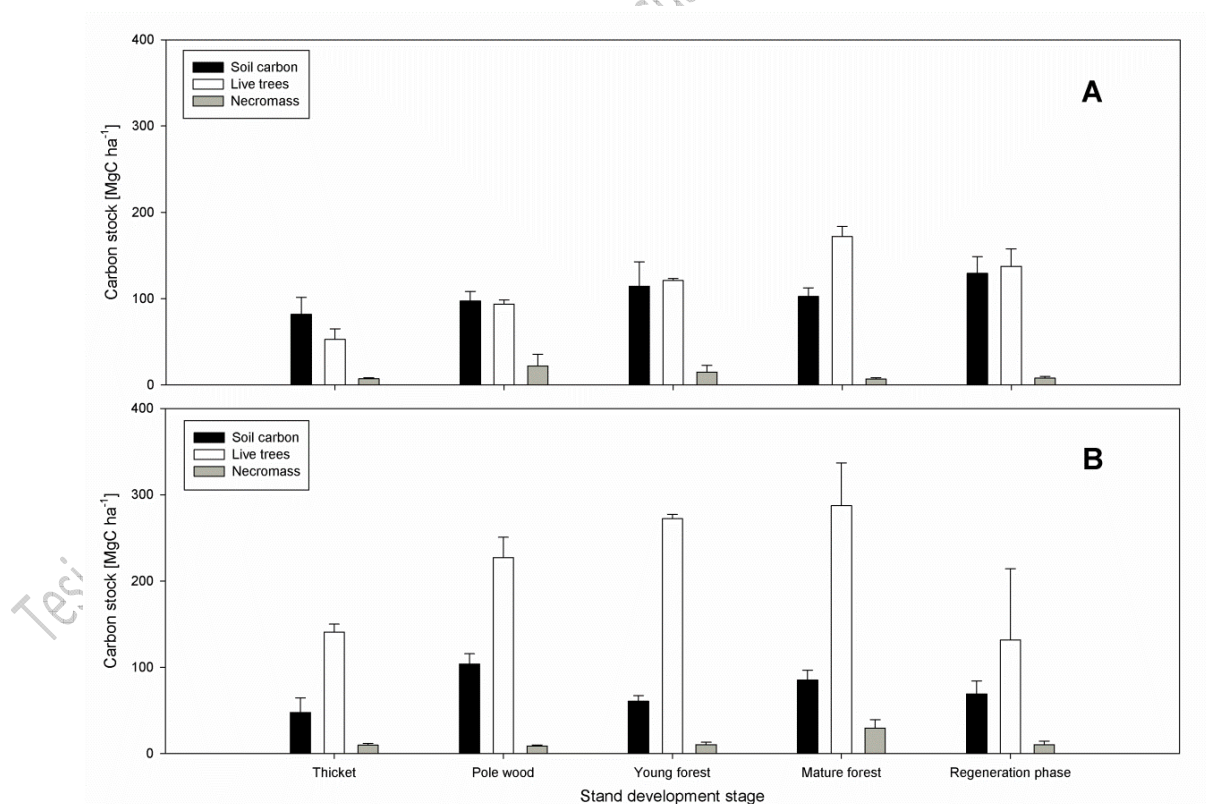


Figure 3.2: Measured total ecosystem carbon stock across the chronosequence (panel A: Beech; panel B: Spruce). Vertical bars represent standard error ($n = 3$).

The trend of total NPP (stem + roots + leaves) differed between the two forests (Figure 3.3a and 3.3b). Beech forest total NPP grew from $3.5 \pm 0.7 \text{ MgC ha}^{-1} \text{ yr}^{-1}$ (thicket) to $4.4 \pm 0.0 \text{ MgC ha}^{-1} \text{ yr}^{-1}$ (young forest) and then slightly decreased in mature forest stands ($3.8 \pm 0.3 \text{ MgC ha}^{-1} \text{ yr}^{-1}$). Afterwards, NPP declined principally due to the effects of the seed cut (regeneration phase). In the spruce forest, total NPP decreased linearly with age ($r^2 = 0.62$; $n=15$; $P < 0.01$), from a maximum of $9.6 \pm 0.1 \text{ MgC ha}^{-1} \text{ yr}^{-1}$ (thicket) to a minimum of $2.9 \pm 1.8 \text{ MgC ha}^{-1} \text{ yr}^{-1}$ (regeneration phase).

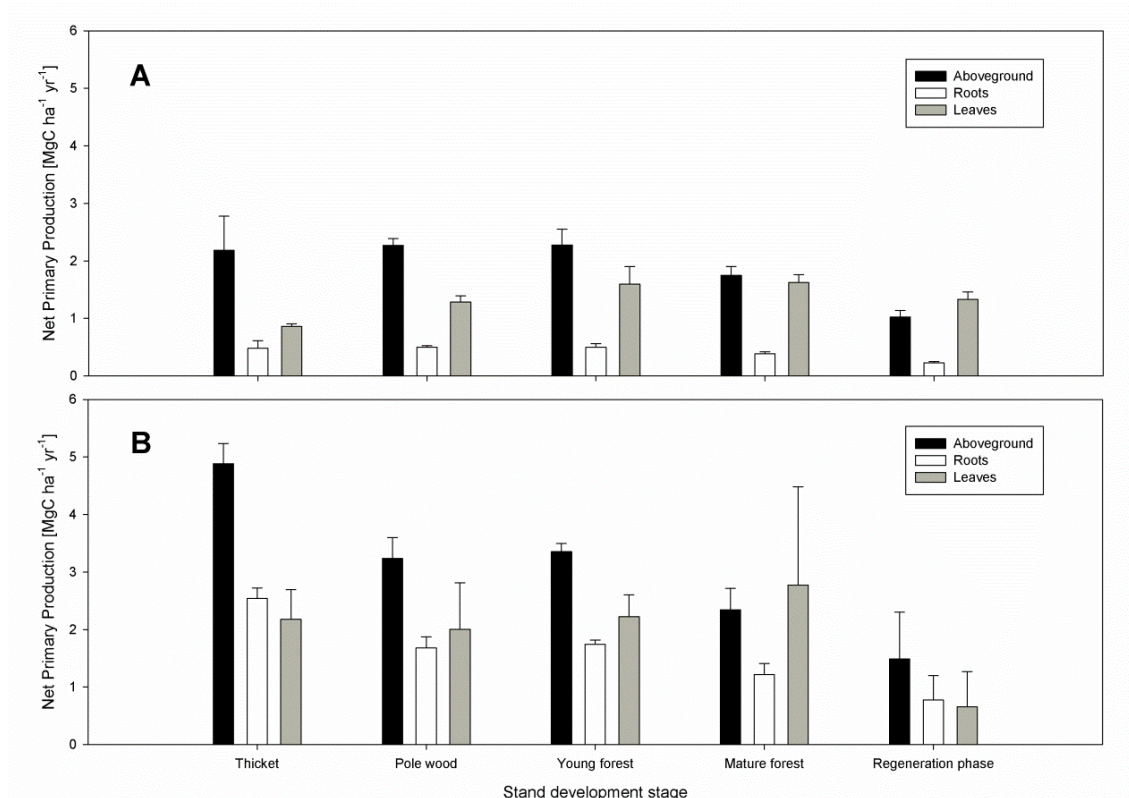


Figure 3.3: Measured net primary productivity (NPP) across the chronosequence (panel A: Beech; panel B: Spruce). Vertical bars represent standard error ($n = 3$).

3.3.1.2 Dead wood and standing litter

The C stock in coarse woody debris (CWD) did not show a clear and significant trend with age along the two chronosequences. However, it was lower in younger stages (thicket and pole wood), had a maximum in young and mature phase (beech and spruce forest, respectively) and then decreased in the regeneration stand. Particularly, in the beech forest, this pool grew from $0.8 \pm 0.1 \text{ MgC ha}^{-1}$ in the thicket up to a maximum value of $4.4 \pm 1.1 \text{ MgC ha}^{-1}$ in young forest, and then decreased to $3.7 \pm 2.0 \text{ MgC ha}^{-1}$ (regeneration phase), where the presence of many post-harvest residues in some plots caused a quite high variability. In the spruce forest this trend was even

more pronounced: C stock in CWD grew from $1.2 \pm 0.5 \text{ MgC ha}^{-1}$ (thicket) to $24.7 \pm 8.2 \text{ MgC ha}^{-1}$ (mature forest) and then went down to $2.2 \pm 0.8 \text{ MgC ha}^{-1}$ after the seed cut. On the contrary, carbon content of stumps tended to decrease with stand age and then to increase as result of thinnings and seed cut. In beech chronosequence, this stock started from $1.1 \pm 0.1 \text{ MgC ha}^{-1}$ in the thicket, decreased to $0.6 \pm 0.1 \text{ MgC ha}^{-1}$ in young forest, and then increased up to $2.2 \pm 0.2 \text{ MgC ha}^{-1}$ after the seed cut. In spruce forest the observations reported a rapid decrease of stump carbon from $3.4 \pm 1.9 \text{ MgC ha}^{-1}$ (thicket) to $1.5 \pm 0.2 \text{ MgC ha}^{-1}$ (pole wood), and an increase to $4.0 \pm 2.0 \text{ MgC ha}^{-1}$ in mature forest as a consequence of thinning treatments, and to $7.8 \pm 3.6 \text{ MgC ha}^{-1}$ in the regeneration phase.

Regarding the litter pool, in the beech forest there was not any significant variation of the standing litter carbon with age ($P > 0.05$) even though an initial reduction of this pool from $2.7 \pm 0.6 \text{ MgC ha}^{-1}$ (thicket) to $1.4 \pm 0.6 \text{ MgC ha}^{-1}$ (young forest) was detected, probably because the degradation of the standing litter was not compensated by new litter input. Following the increase of the aboveground biomass, the C stock of the standing litter rose up to $2.4 \pm 0.2 \text{ MgC ha}^{-1}$ in mature forest and then decreased to $1.9 \pm 0.3 \text{ MgC ha}^{-1}$ after the seed cut. Otherwise, in the spruce forest, this pool increased from $0.5 \pm 0.2 \text{ MgC ha}^{-1}$ (thicket) to $1.5 \pm 0.3 \text{ MgC ha}^{-1}$ (pole wood), then decreased to $0.3 \pm 0.3 \text{ MgC ha}^{-1}$ after the seed cut.

It seems that dead wood carbon stock is more influenced by management system than stand age. In fact, as said above, no significant difference in CWD and stump carbon stock across the two chronosequence has been detected ($P > 0.05$). However, seed cut decreases CWD carbon stock and increases stump carbon stock. Standing litter seems to be influenced only in the first phases after the seed cut when probably there is an increase in decomposition and a decrease in litter input.

3.3.1.3 Soil

None significant trend in soil C stock with stand age was detected in both the chronosequences ($P > 0.05$); soil C stock remained quite constant around a value of $105.1 \pm 8.3 \text{ MgC ha}^{-1}$ and $73.3 \pm 7.1 \text{ MgC ha}^{-1}$ (overall average; $n = 15$) in beech and spruce chronosequence, respectively. Also model simulations did not find any significant variation in soil carbon content in both beech and spruce chronosequences ($P > 0.05$). Hence, we can assert that the applied shelterwood system does not affect soil carbon stock because of the low level disturbance associated with such a silvicultural practice.

3.3.2 Modelled biomass and NEP

Modelled beech total biomass grew asymptotically with age till 350 Mg ha⁻¹ at 160 years, when the seed cut caused a decrease to 207.1 Mg ha⁻¹ (Figure 3.4a). Thereafter, biomass restarted to grow according to the secondary growth rate and reached 223.5 Mg ha⁻¹ at 170 years when the final cut occurred. A good correlation between modelled and measured biomass was found ($r^2 = 0.77$; slope = 0.71; intercept = 59.22; $P < 0.001$). Simulated NEP (Figure 3.4b) reached a maximum value of 3.0 MgC ha⁻¹ yr⁻¹ at 66 years and then slowly decreased to 1.5 MgC ha⁻¹ yr⁻¹ at 159 years. After final cut, a negative NEP was simulated for 8 years, then it started to increase again due to regeneration and to reduction in competition. Even though a good agreement between calculated and modelled NEP values has been found across the chronosequence ($r^2 = 0.57$; slope = 0.81; intercept = 0.24; $P < 0.001$; the intercept is not significantly different from zero [$P = 0.61$]), no measured data in younger forest stages were available and thus there was no possibility to compare this modelled decrease with field data.

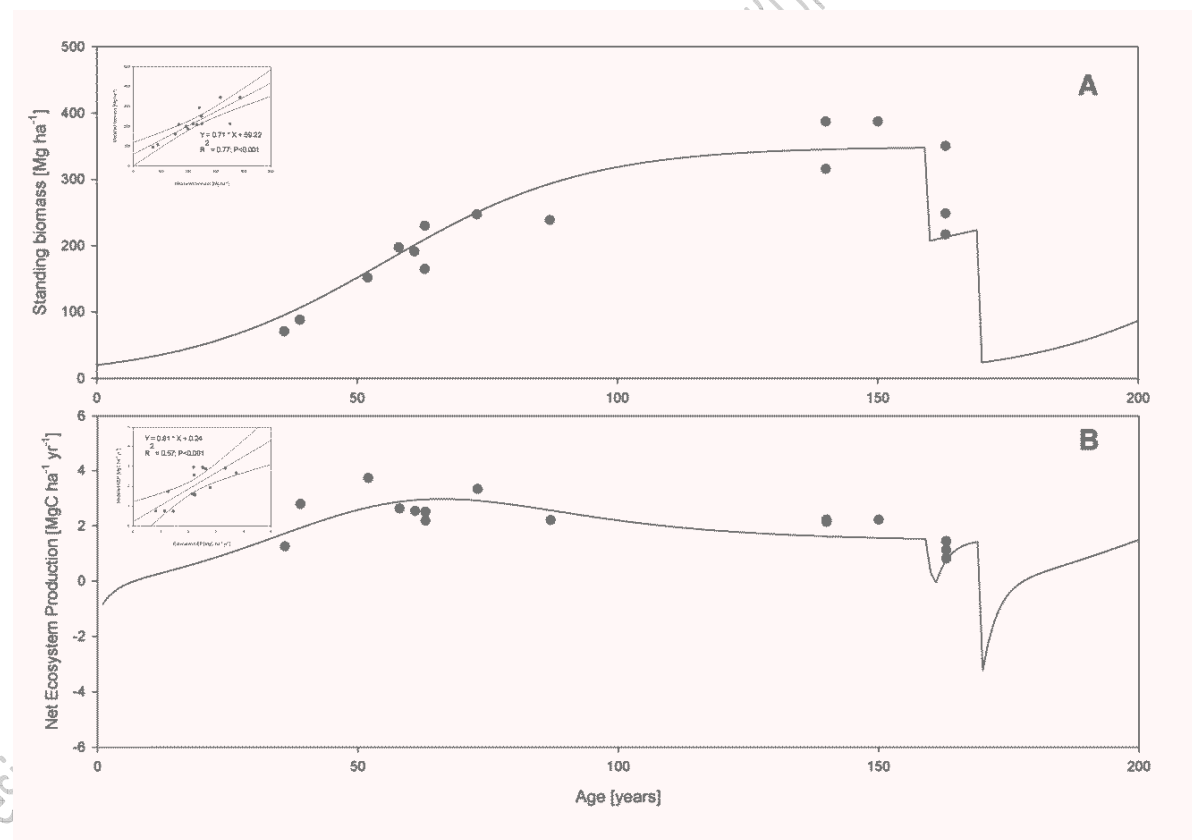


Figure 3.4: Modelled biomass (A) and Net Ecosystem Production (B) across the beech chronosequence as a function of stand age. Dots represent measured biomass or calculated NEP, continuous line represents modelled data. Correlation between modelled and measured data are reported in a top left panel with 95%-confidence intervals (95%).

Modelled spruce total biomass increased with age up to 601.5 Mg ha⁻¹ (140 years) when the seed cut occurred (Figure 3.5a). Also in this case, significant correlations between modelled and measured biomass ($r^2 = 0.87$; slope 0.98; intercept 27.17; $P < 0.001$) and between modelled and calculated NEP ($r^2 = 0.35$; slope 0.39; intercept 0.90; $P < 0.05$; the intercept is not significantly different from zero [$P = 0.23$]) have been found. Modelled NEP (Figure 3.5b) increased till 44 years (4.8 MgC ha⁻¹ yr⁻¹) and then decreased to 1.1 MgC ha⁻¹ yr⁻¹ just before the seed cut. Similarly to beech, simulated NEP was negative after harvest for a period of 11 years.

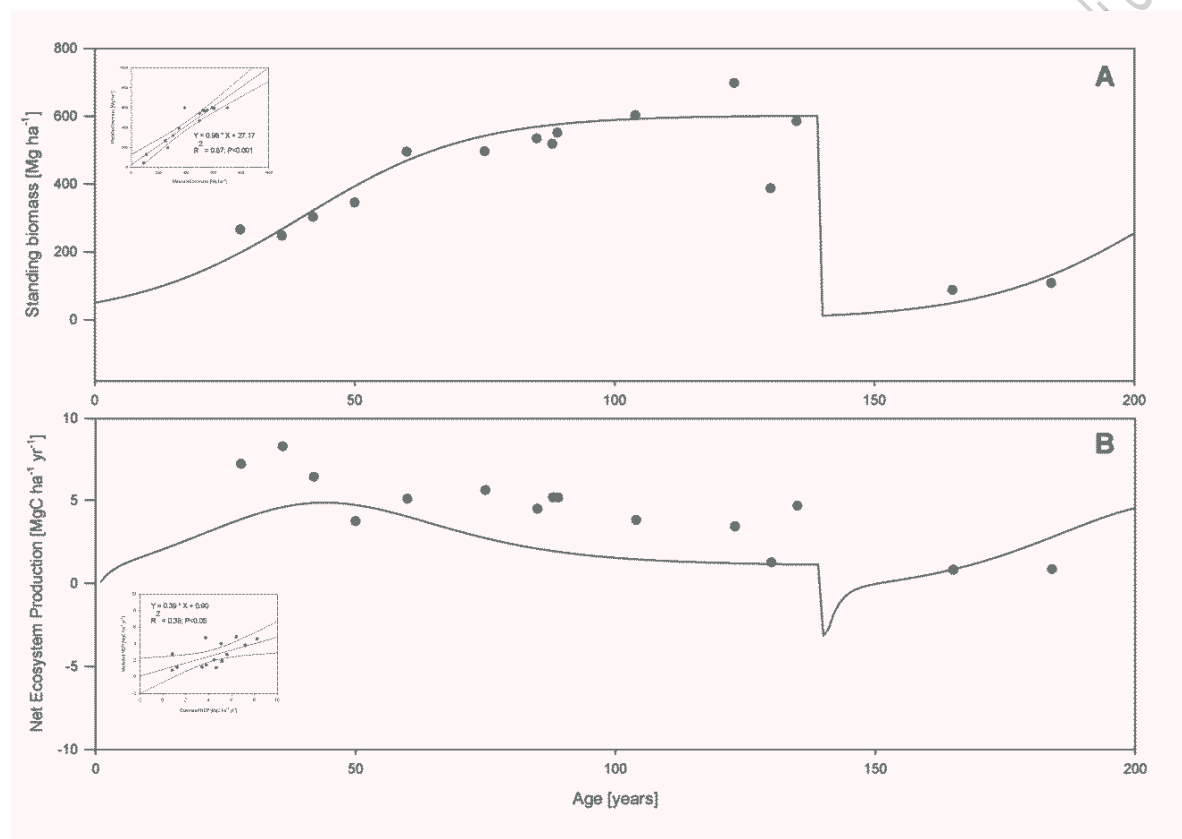


Figure 3.5: Modelled biomass (A) and Net Ecosystem Production (B) across the spruce chronosequence as a function of stand age. Dots represent measured biomass or calculated NEP, continuous line represents modelled data. Correlation between modelled and measured data are reported in a top left panel with 95%-confidence intervals (95%).

3.4 Discussion

3.4.1 Carbon Pools

Total ecosystem C stock in the beech chronosequence ranged from 125 to 305 MgC ha⁻¹ (overall average 232 MgC ha⁻¹). This value is close to those reported by Mund and Schulze (2006) and Wutzler *et al* (2007) for similar managed beech forests in Germany (246 and 225 MgC ha⁻¹, respectively). Moreover, Merino *et al* (2007) reported a total carbon stock of around 243 and 370

MgC ha⁻¹ for partially cut and unmanaged beech forests in Spain, respectively. The difference between our stands and Merino *et al* stands is mainly due to the higher tree biomass in their unmanaged forest. Similarly, our mean value is lower than values reported by Lecointe *et al* (2006) for a mixed managed beech forest in France (283 to 323 MgC ha⁻¹). This difference may be due to the lower elevation of their site (200 m) in comparison to the sites described in the present study (1200 m). As far as spruce is concerned, total ecosystem C ranged between 110 and 458 MgC ha⁻¹ (overall average 299 MgC ha⁻¹). Our average is higher than the mean value of 218 MgC ha⁻¹ reported by Wutzler *et al* (2007), likely because of different forest management (small-strip clear cuts). However, our values are comparable to the range of 75-350 MgC ha⁻¹ observed by Thuille and Schulze (2006) in a secondary spruce forest in the Alps, considering that the lower value is referred to the early stages of the forest succession and root biomass was not calculated. In both the chronosequences, we registered an increment in total C with age which was mainly driven by biomass growth. In fact, while C in standing biomass always increased along the chronosequences, no significant variation in the other pools was measured. This trend is similar to that reported by other studies based on chronosequence approach (Mund and Schulze 2006, Thuille and Schulze 2006).

Concerning with dead wood and litter, comparisons with previous studies are difficult because of the different sampling designs. However, Mund (2004) reported a range between 3 and 9 MgC ha⁻¹ for dead pool (snags + logs + CWD) in an unmanaged beech forest, while Mund and Schulze (2006) quantified a C stock lower than 5 MgC ha⁻¹ in the dead pool of managed beech forests. The mean dead wood (snags + CWD + stumps) carbon stock reported in the present study for beech (9.7 MgC ha⁻¹) is in accordance with these data even though it seems to be higher than expected for a managed forest, probably because of differences in cleaning operations after thinnings or regeneration cuts. On the contrary, our mean volume of dead wood (61.3 m³ ha⁻¹) is lower than the average value of 130 m³ ha⁻¹ reported by Christensen *et al* (2005) reviewing 86 beech forest reserves in Europe. In fact, in managed stands, such as ours, dead wood normally consists only of small twigs and branches and short stumps, with few large logs or snags found (Kruys *et al* 1999). In the spruce chronosequence the mean dead wood carbon was 12.9 MgC ha⁻¹, a value that is comparable to the average of 19.7 MgC ha⁻¹ found by Aakala (2010) between three unmanaged late-successional spruce forests in northern Europe. Instead, CWD stock is lower in comparison to that found by Svoboda *et al* (2010) in an old-growth spruce forest of central Europe because this forest has been developed naturally for many decades.

None significant variation in soil C with age was found for both the chronosequences. Soil C stock of beech forest varied around an average of 105 MgC ha⁻¹ which is quite similar to values reported by Lecointe *et al* (2006) for the 0-30 cm layer (99 MgC ha⁻¹). However, the observed data are higher than values reported by Mund (2004) for the 0-15 cm layer of two beech forest

chronosequences (unmanaged: 53 MgC ha⁻¹; shelterwood system: 42 MgC ha⁻¹). This difference could be due to the fact that these last forests were intensively managed in the past and this could have caused a decrease in C inputs and increase in C outputs. Similarly, our values are higher than those found by Hedde *et al* (2008) for a managed beech forest chronosequence in France (C stock referred to a soil mass equivalent to a soil depth of 0-30 cm; 62-68 MgC ha⁻¹). Contrariwise, our mean is lower than the 110-220 MgC ha⁻¹ found by Meier and Leuschner (2010) in the 0-20 cm layer as these last authors investigated beech stands characterized by lower elevation, lower slope and higher mean temperature. Spruce chronosequence showed an average soil C stock of 73 MgC ha⁻¹, which is comparable to the 65 MgC ha⁻¹ measured by Thiulle *et al* (2000) for the organic layer and mineral soil in an undisturbed 130 years old spruce forest. Our data are also in accordance with the soil carbon content measured by Vesterdal *et al* (2002) in the 0-25 cm layer of a 30 years spruce chronosequence (50-70 MgC ha⁻¹) and with the observations of Thuille and Schulze (2006), which found that in a secondary spruce forest the sum of the organic layer carbon and the mineral soil carbon (0-50 cm) was practically constant around 80 MgC ha⁻¹. Instead, the average soil C stock reported in the present paper is higher than the 53 MgC ha⁻¹ found by Finer *et al* (2003) for an old growth Norway spruce forest in Finland (0-60 cm depth), likely due to the differences in climate and soil type. Conversely, our mean was lower than the 92 MgC ha⁻¹ reported by Stendahl *et al* (2010) for spruce forests in Sweden, but this study derived the 0-100 cm depth soil carbon stock using data from all Swedish spruce plots.

Similarly to literature data (Dupouey *et al* 1999, Lecoite *et al* 2006, Meier and Leuschner 2010), beech forest ecosystem carbon is on average equally distributed between biomass (48%) and soil (46%), while in the spruce chronosequence soil contributed for only 27% to total ecosystem C stock. This last value is well in agreement with Finer *et al* (2003) (62% living vegetation, 30% soil).

3.4.2 NEP

Average NEP measured across the beech chronosequence (2.2 ± 0.2 MgC ha⁻¹ yr⁻¹) is close to NEP measured by eddy covariance technique in a young beech forest in France (2.4 MgC ha⁻¹ yr⁻¹) (Granier *et al* 2000) and in a 80 years old beech forest in Denmark (2.2 MgC ha⁻¹ yr⁻¹) (Pilegaard *et al* 2001). However, our results are lower than those calculated by Mund (2004) (managed forest: 4.6 MgC ha⁻¹ yr⁻¹; unmanaged forest: 4.7 MgC ha⁻¹ yr⁻¹) and measured by Anthoni *et al* (2004) using eddy covariance technique in the same forests (5.0 MgC ha⁻¹ yr⁻¹ and 4.8 MgC ha⁻¹ yr⁻¹, respectively). Regarding spruce chronosequence, mean NEP was 4.4 ± 0.6 MgC ha⁻¹ yr⁻¹, which is similar to the rate of ecosystem C sequestration estimated by Vesterdal *et al* (2007) for two spruce forests in Denmark (4.5 and 4.8 MgC ha⁻¹ yr⁻¹). Anthoni *et al* (2004)

measured a NEP of $0.04 \text{ MgC ha}^{-1} \text{ yr}^{-1}$ in a managed spruce stand (50 years), but this value has been influenced by very high respiration, in fact for another spruce forest in Germany Bernhofer *et al* (2003) reported a net CO_2 uptake between 5.3 and $6.1 \text{ MgC ha}^{-1} \text{ yr}^{-1}$.

As far as modelled NEP is concerned, accordingly to previous studies (Law *et al* 2003, Gough *et al* 2007, Mkhabela *et al* 2009, Zha *et al* 2009), it was lower or even negative in the early stages just after the seed cut and then, after a period of 8 and 11 years, it increased to reach the maximum at 66 and 44 years in beech and spruce chronosequence, respectively. Afterwards, modelled NEP declined due to the increase in forest age even though it remained still positive. As stated by Pregitzer and Euskirchen (2004) in their meta-analysis, NEP is generally higher in intermediate-aged forests (30–120 years), while older forests (> 120 years) are generally less productive accordingly to Odum theory (1969). Moreover, forest age is a highly significant source of variability in NEP at the biome scale and is more related to NPP variations. Pregitzer and Euskirchen (2004) reported a mean temperate forest NEP equal to -1.9 , 4.5 , 2.4 , 1.9 and $1.7 \text{ MgC ha}^{-1} \text{ yr}^{-1}$ across five age classes (0–10, 11–30, 31–70, 71–120, 121–200 years, respectively). NEP was negative after disturbance also in their analysis, and mean ecosystem recovery was also quite fast. Obviously, the time needed for this recovery is primarily due to the intensity and type of the disturbance (Chapin *et al* 2002).

3.5 Conclusions

The main outcome of this work is that the aboveground biomass is the main driver of carbon accumulation. In particular, both field measurements and model simulations proved that soil carbon stock is not influenced by the applied management system. The stability of soil C is probably due to the shelterwood system adopted in this area, which has the aim to reduce as much as possible the impact of human disturbances on soil micro-climate.

The numerical model described in this work has been able to outline significantly the trends of biomass and NEP in both beech and spruce chronosequences. NEP reaches negative values only after the seed cut. However, the recovery period after this event is quite rapid for both chronosequences. Hence, this result seems to confirm that moderate harvesting and regeneration regimes allow for a rapid recovery of the ecosystem, bringing back forest to act as C sink in few years.

Further investigations are desirable in the post-harvesting period and in younger stages (< 40 years) in order to better understand the carbon dynamics of these forests during the regeneration phase.

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3.6 Appendix

Total NPP is simulated using the Osnabrück Biosphere Model (OBM), that is based on regression analysis of temperate climax forests (McGuire *et al* 1993).

Aboveground compartment

Aboveground net primary production (NPP_t ; $\text{Mg ha}^{-1} \text{ yr}^{-1}$) is modelled assuming a logistic growth model, which requires the biomass at time t-1 (B_{t-1}), the maximum aboveground biomass (B_{max} ; Mg ha^{-1}) and the relative growth rate (r_t ; yr^{-1}):

$$NPP_t = r_t \times B_{t-1} \times \left(1 - \frac{B_{t-1}}{B_{max}}\right)$$

The growth rate is dependent on environmental conditions:

$$r_t = r_0 \times \min(f_t, f_p)$$

where r_0 is the optimal relative growth rate (yr^{-1}), with no temperature or water stress limiting conditions, corrected by the minimum among mean annual temperature (f_t) and annual rainfall (f_p) functions. These climate related factors are calculated through empirical equations based on mean annual temperature (T ; °C) and total annual precipitation (P ; mm) (Lieth 1975):

$$f_t = \frac{1}{1 + \exp(1 - 315 - 0.119 \times T)}$$

$$f_p = 1 - \exp(-0.000664 \times P)$$

The annual production of dead wood and litter (DW_{in} ; $\text{Mg ha}^{-1} \text{ yr}^{-1}$) derives from the mortality of the biomass at time t-1 (B_{t-1}) and is computed assuming a constant mortality factor (m ; yr^{-1}):

$$DW_{in} = m \times B_{t-1}$$

The biomass at any time (B_t ; Mg ha⁻¹) is then derived as:

$$B_t = B_{t-1} + NPP_t - DW_{in} - H_t$$

where H_t is a possible harvest at time t (Mg ha⁻¹).

The annual change of dead wood and litter stocks (ΔDW ; Mg ha⁻¹ yr⁻¹) is considered equal to the difference between the dead wood and litter produced (DW_{in}) and the amount mineralized during the year (DW_{out}):

$$DW_{out} = DW_{t-1} \times K_{decay}$$

where K_{decay} is the litter decay constant assuming simple exponential decay (yr⁻¹) and is computed according to the following equation:

$$K_{decay} = K_{DW} \times K_{env}$$

where K_{DW} is the decomposition constant of the dead wood and litter (0.5 yr⁻¹) and K_{env} is a decay factor that depends on the mean annual temperature (T ; °C):

$$K_{env} = \frac{47.9}{1 + \exp\left(\frac{106}{T + 18.3}\right)}$$

This temperature sensitivity function is the same used by the Rothamsted Carbon Model (RothC; Coleman and Jenkinson 1999) and it is applied to all SOM pools, included litter decomposition derived material. Due to the high rainfall conditions at both the sites considered in this study, it is assumed that this parameter is not affected by soil moisture content. The overall litter decomposition rate constants (K_{decay}) are then 0.30 for and 0.36 yr⁻¹ for beech and spruce litter, respectively. These values are closed to the decay constants proposed for the same species in Switzerland by Heim and Frey (2004).

The carbon loss resulting from the mineralization of the dead wood and litter (C_{min-DW} ; MgC ha⁻¹ yr⁻¹) is obtained considering the carbon content of these stocks (%C):

$$C_{min-DW_t} = DW_{out} \times \%C$$

The heterotrophic respiration of the dead wood and litter (Rh_{DW} ; MgC ha⁻¹ yr⁻¹) is a fraction of mineralization that is respired as CO₂. The other part is converted in new SOC by decomposition process. For this purpose, we defined the RD parameter as the ratio between the two fractions of

mineralization (carbon respired/new SOC). The RD parameter is a function of clay percentage in the soil ($\%Clay$):

$$RD = \frac{1.67}{1.85 + 1.6 \times \exp(-0.0786 \times \%Clay \times 100)}$$

The respired portion of C_{min-DW} ($MgC\ ha^{-1}\ yr^{-1}$) is then computed as:

$$Rh_{DW_t} = \frac{RD}{1 + RD} \times C_{min-DW}$$

In the same way the annual portion of C_{min-DW} transferred to SOC ($C_{DW-soil}$) is:

$$C_{DW-soil} = \frac{1}{1 + RD} \times C_{min-DW}$$

Soil compartment

The annual change in soil carbon stock (ΔC_{soil} ; $MgC\ ha^{-1}\ yr^{-1}$) is computed as:

$$\Delta C_{soil_t} = C_{in-soil} - C_{out-soil} = dC_{DW_t} - (Rh_{soil_t} - dCs_{dect})$$

where dC_{DW_t} represent the organic products deriving from the decomposition of dead wood and litter (carbon input to the soil), Rh_{soil_t} is the heterotrophic respiration resulting from organic matter decomposition and dCs_{dect} is the amount of new organic matter formed after the decomposition of the organic matter itself.

The organic products (dC_{DW_t}) are quantified by the difference between the carbon resulting from mineralization and the heterotrophic respiration deriving from the decomposition of dead wood and litter:

$$dC_{DW_t} = C_{min-DW_t} - Rh_{DW_t} = \frac{1}{1 + RD} \times C_{min-DW_t}$$

The heterotrophic respiration of soil compartment (Rh_{soil_t} ; $MgC\ ha^{-1}\ yr^{-1}$) is a function of: soil carbon content at time t-1 (C_{soil-t} ; $MgC\ ha^{-1}$), decomposition products (RD), a constant (Ks_t ; yr^{-1}) that depends on K_{env} , $SOM_{turnover}$ (1.2%) and the clay and carbonates contents ($\%Clay$, $\%Carbonates$) (Rémy and Marin-Lafleche 1974):

$$Rh_{soil_t} = \frac{RD}{1 + RD} \times ks_t \times C_{soil_{t-1}}$$

$$Ks_t = K_{env} \times SOM_{turnover} \times \frac{1200}{(\%Clay \times 100 + 20) \times (\%Carbonates \times 100 + 20)}$$

So, the amount of new organic carbon deriving from the decomposition of the SOM itself (dCs_{dect}) is:

$$dCs_{dec_t} = \frac{1}{1 + RD} \times ks_t \times C_{soil_{t-1}}$$

The annual NEP (NEP_t ; MgC ha⁻¹ yr⁻¹) is finally computed as:

$$NEP_t = NPP_t - Rh_{DWt} - \Delta C_{soil_t}$$

Tesi di dottorato di Giuseppe De Simon, discussa presso l'Università degli Studi di Udine

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Conservation agriculture: effects on the carbon budget of a 5-year crop rotation²

Summary

In recent years, there has been a growing interest in farming practices that can enhance carbon (C) stocks in croplands, which are usually considered C sources for the atmosphere. In this paper, soil temperature, soil water content (SWC), heterotrophic (R_h) and total soil (R_s) respiration and eddy fluxes (gross primary production (GPP); total ecosystem respiration (R_e), net ecosystem exchange (NEE)) were continuously monitored in a paired experiment aimed to assess the effects of conservation management (MT; minimum tillage + cover crops) in a 5-year crop rotation. Then, at the end of growing season, crop biomass and grain yield were quantified in order to compute the net ecosystem C budget (NECB). Data were analyzed independently for three periods based on conventional management: I) tilled period; II) cropping period and III) fallow period. Results did not show any strong soil temperature and soil water content change in MT. Conversely, MT presented a lower R_h leading to an overall reduction in both R_h and R_s considering the whole rotation. Eddy data highlighted the differences among treatments during period I (i.e. higher GPP and lower NEE in MT). Growth analysis showed that MT had no effects on the net primary production (NPP), thus the reduction of R_h in MT was found to improve significantly the C budget of the agro-ecosystem in terms of NEP. However, since MT increased significantly the amount of harvested C, at the end of the 5-year rotation the net ecosystem C balance (NECB) was significantly lower in MT.

Keywords: conservation agriculture, net ecosystem carbon balance, crop rotation, crop yield, soil respiration, eddy covariance.

² De Simon G, Alberti G, Delle Vedove G, Zerbi G and Peressotti A (*submitted*). Conservation agriculture: effects on the carbon budget of a 5-year crop rotation. *Agriculture, Ecosystems & Environment*.

4.1 Introduction

World croplands extension has been estimated around 1.53×10^9 ha at the end of the last millennium (Biradar *et al* 2009), but their contribution to the global carbon (C) budget is still under debate. C balance in agro-ecosystems is mainly driven by the agricultural practice and, to a less extent, by the effects of climate change (Gervois *et al* 2008). This last is expected to cause an increase in mean temperatures, in the frequency and length of climate extremes and a change in the spatial and temporal distribution of precipitations causing also frequent droughts (Trenberth *et al* 2007) especially during summer (Seneviratne *et al* 2006) which coincides with the growing season of the principal crops in Europe. These projected changes are going to have a deep impact on the yield and C balance of croplands, especially for rain fed agriculture.

In terms of C balance, croplands are mostly reported to release C in the atmosphere (Jenssens *et al* 2003, Loubet *et al* 2011) causing a consistent contribution to the carbon balance deficit (Smith 2004). However, recent estimates of cropland net biome production (NBP) revealed that in Europe (EU-25) crops may act as a small source or a small sink of atmospheric C (Ciais *et al* 2010). Moreover, there are examples of croplands acting as systematic C sinks (Aubinet *et al* 2009). During the 20th century, the intensification of farming practices (selection and use of new species, increasing use of mineral fertilizers instead of manure, irrigation) has led to a considerable increase in European crop yields (+300-400%; Mazoyer 2002). However, the increase in yield does not seem to have entrained a parallel increase in soil C stocks (Arrouays *et al* 2002), and soil C may have even decreased in many regions of intensive agriculture (Walter *et al* 1995, Fardeau *et al* 1988, Bellamy *et al* 2005). In particular, some estimations indicate that agricultural soils have lost the 30-50% of C because of intensive cultivation (Kucharik *et al* 2001).

For these and other reasons (i.e. degradation of ecosystem services and biodiversity), business-as-usual agriculture is increasingly considered inadequate to deliver a sustainable production (Shaxson 2006). Hence, there is a need for applying farming practices that can have a positive contribution on C balance in croplands. Accordingly, conservation agriculture (CA) is being increasingly promoted as constituting a set of principles and practices that can make a contribution to sustainable production intensification (FAO 2008, Pretty 2008). The three components of CA are (Kassam *et al* 2009): 1) maintaining year-round organic matter cover over the soil also through cover crops and intercrops; 2) minimizing soil disturbance by tillage and keeping soil disturbance from cultural operations as minimum as possible and 3) diversifying crop rotations, sequences and associations, adapted to local environmental conditions, and including appropriate nitrogen-fixing legumes.

Conservation agriculture is supposed to bring many positive effects to crops in comparison to conventional farming, such as reverse the loss of organic matter, improve and maintain soil porosity, reduce weed, insect pest and disease incidence, raise agro-ecological diversity and moreover reduce production costs (Kassam *et al* 2009). Overall, CA is supposed to reduce C emissions, to rise soil organic matter content (SOM; Lal and Bruce 1999, Lal 2003, 2004; Baker and Griffis 2005) and to increase crop-water availability and thus resilience to drought (FAO 2008). The SOC increase resulting from a shift from conventional to CA has been quantified by West and Post (2002) in $57 \pm 14 \text{ gC m}^{-2} \text{ yr}^{-1}$. Considering water, under CA evapotranspiration may be reduced and the soil water content may increase in the upper soil layer, while the presence of crop residues could lead to a greater water storage counteracting the possible negative effect of reduced- or no-tillage practices on water infiltration (Lahmar 2010). In particular, CA has been effective at sustaining crop production in semi-arid rain-fed regions (Lyon *et al* 2004).

Here, we present the results of a field experiment set up to investigate the effects of a CA practice (minimum tillage + cover crops; MT) on the C dynamics of a 5-year crop rotation (2007-2011). In particular, the aims of this paper were to test the effects of MT management on: i) soil physical parameters (temperature and soil water content); ii) ecosystem C fluxes (gross primary production, ecosystem respiration, heterotrophic and total soil respiration, net ecosystem exchange); iii) crop productivity (biomass, grain yield, harvest index) and overall net ecosystem C budget (i.e. net biome production).

4.2 Materials and methods

4.2.1 Site description

The study site is an agricultural field of 13.3 ha located in Beano (Italy; 46°00' N 13°01'E, 65 m a.s.l.). In the last 30 years before starting the experiment, maize (*Zea mays* L.) was cultivated and soil was tilled using a winter plow up to a depth of 0.35 m and a spring soil preparation (5 cm) before sowing. A quite constant grain yield (10–11 Mg ha⁻¹ dry matter) was achieved using a sprinkler irrigation system and adding chemical fertilizers used in accordance with local standard practices (Alberti *et al* 2010).

Annual mean temperature in Beano is 13.3°C (average over the period 1991-2010) with mean monthly temperatures of 3.7°C and 23.1°C in January and July, respectively. Annual mean precipitation is around 1200 mm (1991-2010), out of which around 650 mm falls during the cropping period (April-September).

Soil can be classified as a Chromi-Endoskeletal Cambisol (FAO 2006) with the following characteristics in the 0–30 cm horizon: total SOC = $48.4 \pm 8.5 \text{ tC ha}^{-1}$, total N = $4.2 \pm 1.1 \text{ tN ha}^{-1}$,

soil bulk density = $1.25 \pm 0.15 \text{ g cm}^{-3}$, soil field capacity = 23% v/v, wilting point = 12% v/v and pH = 7.1 ± 0.02 (Alberti *et al* 2010).

4.2.2 Crop rotation, agronomic management and experimental design

At the beginning of the experiment (December 2006) the study area was split into two parts: an eastern field of 8.6 ha and a western field of 4.7 ha. In each field, three plots (10 x 10 m), according to a block design, were established to test the effects of different crop managements on soil C dynamics, crop growth, crop yield and soil respiration.

Since the beginning of the experiment, two managements were applied: the eastern part of the field was tilled up to 30 cm depth during winter and was designated as the control treatment (CT), while the western part of the field was managed using a minimum tillage strategy (MT).

In the CT field, the crop history consisted in a 4-years (2007-2010) of continuous maize (*Zea mays* L.) and one year (2011) of soybean (*Glycine max* (L.) Merr.). In the MT field, the crop rotation consisted in three years (2007-2009) of alfalfa (*Medicago sativa* L.), one year of maize (2010) and one year of soybean (2011). Following the CT management operations, we defined the “growing season” as the period between sowing and harvest, “fallow period” as the period between harvest and tillage and “tillage period” as the period between tillage and sowing. In MT, during the 2011 fallow period, a cover crop (oats, rye and barley) was planted after harvest and sprayed with glyphosate before sowing. Both field were irrigated and fertilized according to best local agricultural practices (Table 4.1).

Year	Date	Conventional tillage (CT)		Minimum tillage (MT)	
		Crop	Operation	Crop	Operation
2006	15/12/2006	-	Plowing	-	Plowing
2007	25/02/2007	maize	Fertilization	alfalfa	-
	02/03/2007		-		Sowing
	03/04/2007		Soil preparation		-
	04/04/2007		Sowing and fertilization		Fertilization
	24/04/2007		Fertilization		-
	04/05/2007		Weed control		-
	17/05/2007		Fertilization		-
	21/05/2007		-		Harvest
	18/06/2007		-		Harvest
	17/07/2007		-		Harvest
	17/08/2007		-		Harvest
	06/09/2007		Harvest		-
	01/10/2007		-		Harvest
	12/12/2007		Plowing		-

2008	03/04/2008	maize	Soil preparation	alfalfa	-
	04/04/2008		Sowing and fertilization		-
	09/04/2008		Weed control		-
	06/05/2008		-		Harvest
	09/05/2008		Fertilization		-
	03/06/2008		Fertilization		-
	19/06/2008		-		Harvest
	15/07/2008		-		Harvest
	06/08/2008		-		Harvest
	08/09/2008		-		Harvest
	25/09/2008		Harvest		-
2009	16/01/2009	maize	Plowing	alfalfa	-
	19/03/2009		Soil preparation		-
	08/04/2009		Sowing and fertilization		-
	30/04/2009		-		Harvest
	13/05/2009		Fertilization		-
	26/05/2009		Fertilization		-
	01/06/2009		-		Harvest
	30/06/2009		-		Harvest
	23/07/2009		-		Harvest
	23/08/2009		-		Harvest
	10/09/2009		Harvest		-
	25/11/2009		Plowing		-
	2010		17/03/2010		maize
08/04/2010		Fertilization	Fertilization		
10/04/2010		Sowing	Sowing		
19/05/2010		Fertilization	Fertilization		
07/06/2010		Fertilization	Fertilization		
20/09/2010		Harvest	Harvest		
04/10/2010		-	Cover crop sowing		
13/12/2010		Plowing	-		
2011	14/02/2011	soybean	Soil preparation	soybean	-
	19/04/2011		Weed control		Weed control
	04/05/2011		Sowing		Sowing
	06/05/2011		Weed control		-
	11/06/2011		Weed control		Weed control
	26/09/2011		Harvest		Harvest
	10/10/2011		-		Cover crop sowing
	23/11/2011		Plowing		-

Table 4.1: Main agricultural operations for conventional tillage (CT) and minimum tillage (MT).

4.2.3 Meteorological measurements

A weather station was placed within each field. Each station was equipped to measure soil temperature at three depths (5, 15 and 25 cm) using type T thermocouples, soil water content (0–20 cm) using time-domain reflectometers (TDR CS616, Campbell Scientific, Logan, UT),

incoming and outgoing short wave radiation (CMP3, Kipp and Zonen), incoming long wave radiation (CG3, Kipp and Zonen), incoming and outgoing photo flux density (PPFD, LI-190SL quantum sensor, LiCor, Lincoln, NE), net radiation (NR-LITE, Kipp and Zonen), air temperature and humidity (HMP45AC, Vaisala), soil heat flux at two depths (5 and 15 cm) using four soil heat flux plates (Marlow) and precipitation.

Heat storage between soil surface and the depth of the plate was added to the measured flux in order to obtain corrected surface values of soil heat flux (Ochsner *et al* 2007, Cava *et al* 2008) according to the equations reported by Alberti *et al* (2010).

In both stations, variables were measured at 0.1 Hz and then collected and averaged half-hourly using a CR10X datalogger (Campbell Scientific Inc.).

4.2.4 Eddy covariance measurements

An eddy covariance flux tower was installed in each part of the field to assess mass, momentum and energy ecosystem exchanges. The eddy covariance systems were made up of a sonic anemometer (81000V, Young, USA) and an open path infrared gas analyzer (LI-7500, Li-Cor, USA) installed at 2.5 m above soil during winter period and at 4.5 m above soil (2 m above crop canopy) during cropping period when maize was planted. The LI-7500 was pointed toward the North by an angle of 20° to minimize solar radiation influence and to facilitate the shedding of water droplets from the sensor lenses after rain events. Data from the sonic anemometer and the open path IRGA were recorded at a frequency of 20 Hz by a hand computer (Matese *et al* 2008). Ecosystem fluxes of CO₂, momentum, sensible (H), and latent heat (LE) were then averaged over 30 min. The applied methodology was based on the EuroFlux protocol (Aubinet *et al* 2000). All the post-processing elaborations and frequency response corrections have been performed using EdiRe Data software (University of Edinburgh), and quality assessment and quality check analysis (QA and QC) have been applied according to Foken and Wichura (1996). Before the QA/QC analysis, a spike analysis algorithm has been applied to accept or discard data from rainy or foggy conditions, or when water condensation occurs on the instrument optical lens. Moreover, in order to eliminate the influence of the nearby CT field, fluxes at MT site were excluded with wind direction in the range 180–360°. To assume comparability of fluxes from both fields, data from 180° to 360° wind directions were also excluded at the CT site (Alberti *et al* 2010).

A gap-filling procedure was applied to obtain daily fluxes (Reichstein *et al* 2005) when the QA/QC criteria were not satisfied and when a lack of turbulent transport was evident from data. Total ecosystem respiration (R_e) was computed according to the short-term temperature response of night-time fluxes, while gross primary production (GPP) was derived as difference between net ecosystem exchange (NEE) and R_e . The gap-filling related NEE measurement uncertainty was

assessed by simulating random gaps and re-applying the gap-filling procedure (Reichstein *et al* 2005). Other sources of NEE measurement uncertainty, such as systematic and random flux measurement errors or the uncertainty related to data processing options, were not considered in this study.

4.2.5 Soil respiration measurements

Continuous soil respiration coupled with continuous soil temperature (2.5 and 5 cm depth) and soil moisture (12 cm depth) measurements were performed every 2 h using three automated closed dynamic soil respiration systems (Delle Vedove *et al* 2007). The system used the rate of increase of CO₂ within the chamber to estimate, by an empirical diffusion model, the gas efflux ($\mu\text{mol m}^{-2} \text{s}^{-1}$) within twelve soil respiration chambers per block. Each chamber consisted of a steel collar (20 cm diameter, 8 cm height) and a DC motor closing a steel lid, that was positioned to end up on the North side of the collar when it was open in vertical position in order to avoid chamber shadowing. During measurements, air was circulated between the soil chamber and the infrared gas analyzer (IRGA, SBA-4, PP-System), at a constant flow rate (0.5 l min^{-1}). Next to the chambers, soil temperature at two depths (2.5 and 5 cm) and soil water content were measured continuously.

In the period 2007-2009, three chambers were placed in each replicate: two chambers were used to estimate total soil respiration (R_s) and one was used to estimate heterotrophic respiration (R_h) on a root exclusion subplot. During crop year 2010, six chambers were placed in each plot: four chambers were used to estimate R_s and two was used to estimate R_h on root exclusion subplots. During crop year 2011, only three chambers per replicate were placed due to the presence of another experiment in the field (De Simon *et al*, under review; Chapter 5): one chamber measured R_s and two chambers measured R_h . The root exclusion subplot consisted in isolating soil below the chamber using a stainless-steel cylinder opened on both ends (32 cm diameter, 40 cm height). The steel cylinders were placed in the field after sowing and removed after final harvest.

Daily soil respiration data was calculated as average across four periods during the day (00:00-6:00; 6:00-12:00; 12:00-18:00; 18:00-24:00). Days with not at least three periods of data were discarded in order to not under- or over-estimate soil respiration as a complete daily trend was not available. Across all the study period (2007-2011), 17% and 13% of daily data for R_h and R_s , respectively, in CT and 35% and 29% in MT were missing or discarded according to the abovementioned quality check. The missing data were then gap-filled using soil temperature at 2.5 cm depth (Pavelka *et al* 2007) according to Van Hoff equation (Lloyd and Taylor 1994).

4.2.6 Growth analysis

The above-, below-ground dry biomass and grain yield were assessed each year at final harvest by destructive sampling.

The maize above- and below-ground dry biomass was determined after oven drying at 70 °C for 48 h each plant component (foliage, stem, roots, cob) of 10 sampled plants. Maize grain yield was determined by hand-harvesting an inner area on each plot at the end of the growing season (20 m²).

Alfalfa aboveground biomass was estimated at each harvest in 32 m² subplots. Belowground biomass was estimated at the end of 2009 growing season by excavating three soil profiles (around 30 x 30 cm) per plot up to a depth of 1 m. Thus, total 2007-2009 NPP for alfalfa was estimated as sum of total above ground production in the 2007-9 period and root biomass estimated in 2009.

For soybean, all plants in a 1.7 m² subplot in each plot were collected and, after oven-drying at 70 °C for 48 h, above- and below-ground dry biomass and dry grain yield were determined. Furthermore, on a subsample of 10 plants, the harvest index (HI) was calculated as the ratio between the grain yield and the above-ground biomass.

Every year, the crop residues after harvest were estimated on three 1 m² subplots for each plot.

The total biomass was computed as the sum of above- and below-ground biomass and crop residues. The difference between total biomass and the grain yield was assumed to be the apparent soil C input.

Carbon and nitrogen contents for each plant component of alfalfa, maize and soybean were determined using a CHN Elemental Analyzer (NA1500 Series 2, Carlo Erba Instruments, Rodano, Italy).

4.2.7 Carbon dynamics

Net Ecosystem Carbon Budget (*NECB*) was calculated according to Smith *et al* (2010):

$$NECB = NEP - H$$

where *NEP* was derived by the annual Net Ecosystem Exchange, measured by eddy covariance technique ($NEP = -NEE$) or as difference between NPP calculated using the growth analysis and R_h , and *H* is the amount of harvested C.

The Carbon Use Efficiency (*CUE*) was calculated as the ratio between NPP and GPP during the growing season (Choudhury 2000), while the Carbon Sequestration Efficiency (*CE*) was

computed for the whole year as the ratio between NECB and the apparent soil C input (Ciais *et al* 2010). The $R_e : GPP$ ratio was calculated in order to test ecosystem carbon sequestration rate in each period. Considering soil respiration, while the $R_h : NPP$ ratio were computed annually in order to assess the C loss from microbial respiration and the potential C gain from net primary productivity (Guzman and Al-Kaisi 2010). Moreover, a comparison between the NEP estimated by growth analysis ($NEP_{GA} = NPP - R_h$) and the NEP derived from eddy covariance ($NEP_{eddy} = -NEE$) was performed. NEP_{GA} was calculated on the assumption that turnover rates of root exudates is of the order of few hours (Kuzyakov 2006) as such exudates mainly consist of low molecular weight organic substances such as carbohydrates, carboxylic acids and amino acids. Thus, on an annual basis, their contribution to total NPP can be considered equal to zero as they are rapidly decomposed by microbes. Finally, annual ratio between R_h and R_s was also calculated to estimate microbial contribution to total soil respiration among different crops and management strategies.

4.2.8 Data analysis

As said before, each solar year was divided in 3 periods according to the timing of the agricultural operations in CT: I) tilled period (after plowing); II) cropping period (from sowing to harvest) and III) fallow period (from harvest to plowing). Data were then analysed independently for each period in each year, for the whole year and finally for the entire study rotation (2007-2011).

The mean daily trend of soil temperature was obtained averaging half-hourly data over each period. The trends of heterotrophic respiration and soil water content over the entire period of study were analysed on weekly basis. Differences between treatments in mean values of the considered periods, in the daily trends and in the weekly means were tested by t-tests. All analyses were performed using SigmaPlot 11 (© Systat Software Inc.). When not differently specified, the level of significance was set at $P = 0.05$. All errors presented in text and graphs are standard error of the mean unless reported otherwise.

4.3 Results

4.3.1 Soil temperature and water content

The average soil temperatures at 2.5 and 5 cm depth ($T_{2.5}$ and T_5 ; Table 4.2) were tested on the basis of the crop history (i.e. average of maize and alfalfa 2007-2009; of maize 2010 and of soybean 2011). Considering 2007-2009, there were no significant differences in both $T_{2.5}$ and T_5 during period I, while $T_{2.5}$ of MT was higher than in CT during period II ($P = 0.045$), probably due to crop senescence in 2009. In the fallow period, instead, T_5 of MT was lower than in CT ($P =$

0.032). $T_{2.5}$ and T_5 were not different among treatments during period I of 2010, while during the cropping period of this year both $T_{2.5}$ and T_5 were higher in MT than CT ($P = 0.015$ and $P = 0.031$). After harvest, again there were no differences in mean soil temperatures. On the contrary, during 2011 both $T_{2.5}$ and T_5 were lower in MT than in CT during the tilled period ($P = 0.027$ and $P = 0.024$), while we did not observe differences among treatments in periods II and III. These differences had an effect also on the annual mean soil temperatures, but not on the 5-year rotation average (Table 4.3).

The differences in the mean daily trend of $T_{2.5}$ (Figure 4.1) were tested only when both treatments were planted with the same crop (2010 and 2011). This analysis showed that during period I of 2010 MT presented a significantly higher $T_{2.5}$ during night and early morning (3.00 – 10.00), while during daytime (11.30 – 17.00) $T_{2.5}$ in MT was significantly lower than in CT, even by more than 1°C in the mid-afternoon. In the same period of 2011 we did not observe any significant difference among treatments during night, however the daytime difference in $T_{2.5}$ between treatments was even more pronounced ($T_{2.5}$ in MT significantly lower than in CT from 11.00 to 20.00, with a maximum alteration of -1.8°C in the mid-afternoon). During the cropping period (II) of 2010, $T_{2.5}$ in MT was significantly higher than in CT during the morning (from 5.30 to 8.30), while in the same period of 2011 the mean $T_{2.5}$ of MT was higher than CT only between 6.30 and 7.00, and, on the contrary, it was significantly lower in MT in the late afternoon (17.00 – 17.30 and 18.30 – 19.00). In the fallow period (III), $T_{2.5}$ in MT was significantly higher than in CT from 2.30 to 7.30 in 2010, while in period III of 2011 we did not detect any significant difference between treatments at any hour.

There were generally no significant differences among treatments in SWC during the considered periods (Table 4.2), except for the tilled period of both 2010 ($P = 0.021$) and 2011 ($P = 0.047$). Also for SWC, these alterations led to a significant difference in the annual average of 2010 ($P = 0.040$; Table 4.3). The mean daily trends of SWC were similar between treatments, even if diurnal fluctuations were narrower in MT (data not shown).

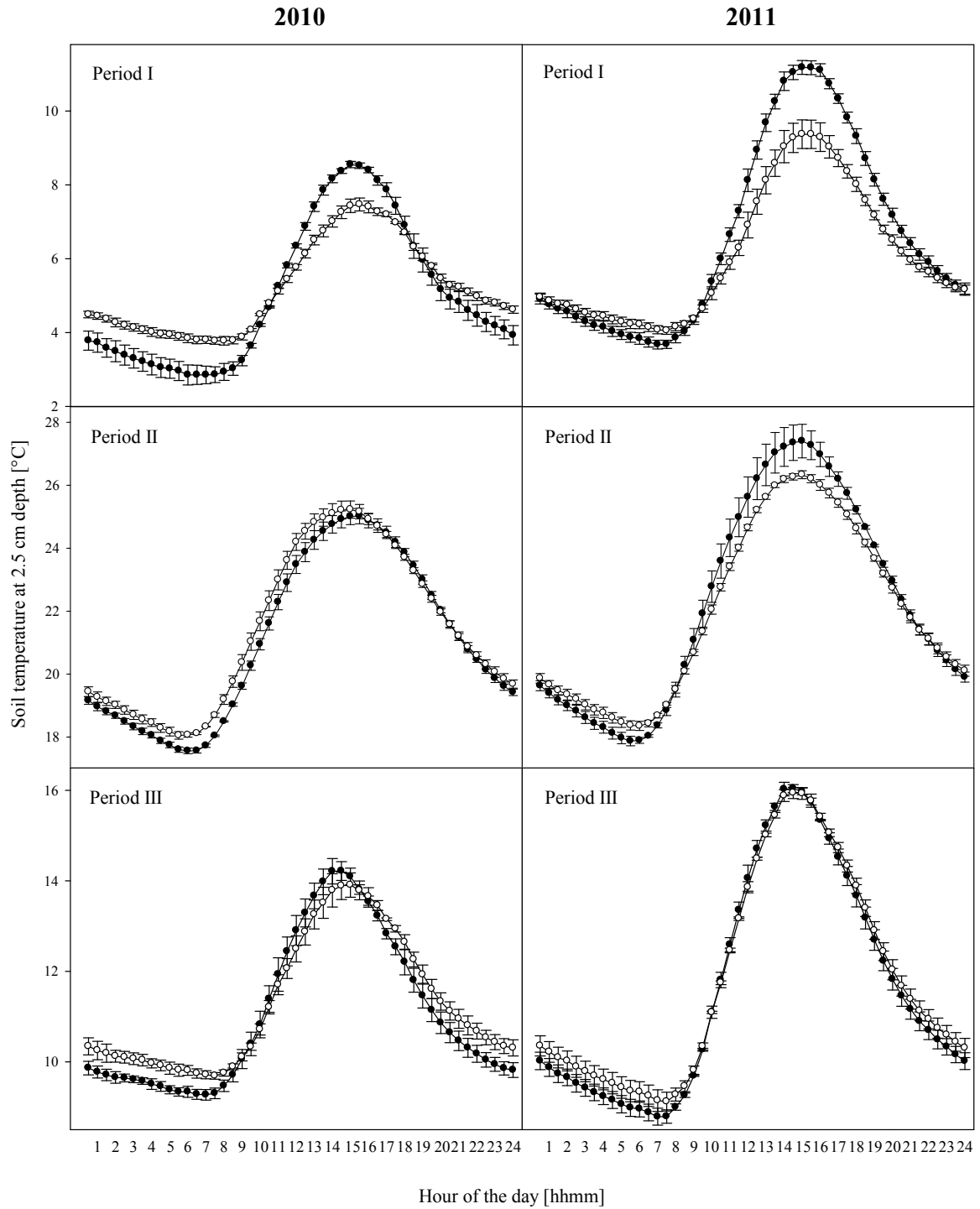


Figure 4.1: Mean daily trends of soil temperature at 2.5 cm depth of conventional tillage (CT; filled circles) and minimum tillage (MT; open circles) for maize (2010) and soybean (2011). Vertical bars represent standard error ($n = 3$).

		Period								
Treat		2007-2009 - I	2007-2009 - II	2007-2009 - III	2010 - I	2010 - II	2010 - III	2011 - I	2011 - II	2011 - III
T _{2.5} (°C)	CT	6.88 ± 0.04 a	21.12 ± 0.06 b	11.16 ± 0.07 a	5.18 ± 0.09 a	21.11 ± 0.07 b	11.11 ± 0.07 a	8.27 ± 0.10 a	22.77 ± 0.12 a	12.57 ± 0.10 a
	MT	6.83 ± 0.07 a	21.48 ± 0.11 a	11.14 ± 0.04 a	5.32 ± 0.03 a	21.45 ± 0.04 a	11.34 ± 0.08 a	7.73 ± 0.13 b	22.70 ± 0.08 a	12.77 ± 0.11 a
T ₅ (°C)	CT	6.96 ± 0.02 a	21.05 ± 0.06 a	11.34 ± 0.04 a	5.28 ± 0.09 a	21.05 ± 0.07 b	11.26 ± 0.09 a	8.31 ± 0.10 a	22.67 ± 0.08 a	12.76 ± 0.13 a
	MT	6.83 ± 0.04 a	21.42 ± 0.12 a	11.15 ± 0.04 b	5.33 ± 0.05 a	21.34 ± 0.06 a	11.49 ± 0.07 a	7.75 ± 0.12 b	22.60 ± 0.06 a	12.96 ± 0.09 a
SWC (%)	CT	0.26 ± 0.01 a	0.28 ± 0.02 a	0.27 ± 0.01 a	0.23 ± 0.02 b	0.22 ± 0.01 a	0.32 ± 0.01 a	0.23 ± 0.02 b	0.27 ± 0.03 a	0.25 ± 0.03 a
	MT	0.29 ± 0.02 a	0.28 ± 0.02 a	0.28 ± 0.02 a	0.32 ± 0.02 a	0.27 ± 0.02 a	0.31 ± 0.02 a	0.28 ± 0.01 a	0.27 ± 0.02 a	0.28 ± 0.01 a
R _h (gC m ⁻²)	CT	359 ± 35 a	793 ± 50 a	419 ± 40 a	134 ± 19 a	295 ± 30 a	112 ± 9 a	243 ± 11 a	298 ± 16 a	74 ± 8 a
	MT	189 ± 11 b	794 ± 47 a	224 ± 18 b	65 ± 14 b	376 ± 21 a	95 ± 15 a	96 ± 12 b	266 ± 66 a	46 ± 4 b
R _s (gC m ⁻²)	CT	437 ± 114 a	2015 ± 126 a	512 ± 86 a	153 ± 33 a	583 ± 79 a	127 ± 19 a	243 ± 11 a	614 ± 55 a	74 ± 8 a
	MT	382 ± 29 a	1652 ± 70 a	507 ± 21 a	126 ± 10 a	537 ± 22 a	139 ± 13 a	149 ± 11 b	426 ± 44 a	67 ± 5 a

Table 4.2: Mean values of soil temperature at 2.5 and 5 cm depth (T_{2.5} and T₅), soil water content (SWC) and cumulated values of soil respiration (R_h and R_s) in conventional tillage (CT) and minimum tillage (MT) for each of the periods considered (± standard error, *n* = 3). Different letters indicate a significant difference between treatments (*P* < 0.05).

	Treat	Period			
		2007-2009	2010	2011	5-years
T _{2.5} (°C)	CT	14.28 ± 0.04 b	13.68 ± 0.04 b	14.71 ± 0.03 a	14.25 ± 0.03 a
	MT	14.43 ± 0.02 a	13.93 ± 0.01 a	14.48 ± 0.08 a	14.34 ± 0.03 a
T ₅ (°C)	CT	14.33 ± 0.03 a	13.72 ± 0.04 b	14.72 ± 0.03 a	14.28 ± 0.03 a
	MT	14.4 ± 0.05 a	13.92 ± 0.03 a	14.48 ± 0.07 b	14.32 ± 0.04 a
SWC (%)	CT	0.27 ± 0.01 a	0.25 ± 0.00 b	0.25 ± 0.02 a	0.26 ± 0.01 a
	MT	0.28 ± 0.02 a	0.30 ± 0.02 a	0.28 ± 0.00 a	0.29 ± 0.01 a
R _h (gC m ⁻²)	CT	1571 ± 71 a	541 ± 17 a	615 ± 14 a	2726 ± 81 a
	MT	1206 ± 46 b	537 ± 41 a	408 ± 81 a	2150 ± 114 b
R _s (gC m ⁻²)	CT	2963 ± 109 a	862 ± 87 a	930 ± 51 a	4757 ± 72 a
	MT	2541 ± 73 b	801 ± 17 a	642 ± 55 b	3985 ± 136 b
GPP (gC m ⁻²)	CT	5986	1922	1212	9120
	MT	5484	1706	1327	8517
Re (gC m ⁻²)	CT	4857	1569	1383	7810
	MT	3855	1372	1294	6520
NEE (gC m ⁻²)	CT	-1128 ± 23	-353 ± 11	173 ± 10	-1307 ± 27
	MT	-1629 ± 24	-335 ± 12	-31 ± 11	-1995 ± 29
Biomass (gC m ⁻²)	CT	3131 ± 58 a	1099 ± 28 a	419 ± 23 a	4649 ± 73 a
	MT	3094 ± 15 a	962 ± 103 a	463 ± 33 a	4519 ± 103 a
Grass/Grain yield (t ha ⁻¹)	CT	30.6 ± 0.6 b	11.1 ± 0.1 a	4.0 ± 0.3 a	45.7 ± 0.9 b
	MT	53.4 ± 0.9 a	9.6 ± 0.4 b	4.4 ± 0.4 a	67.4 ± 1.3 a

Table 4.3: Annual and 5-years averages of soil temperature at 2.5 and 5 cm depth (T_{2.5} and T₅), soil water content (SWC) and cumulated values of soil respiration (R_h and R_s), eddy fluxes (GPP, Re, NEE) and crop productivity (biomass and grain yield) in conventional tillage (CT) and minimum tillage (MT; ± standard error, $n = 3$; NEE ± uncertainty according to Reichstein *et al* 2005). Different letters indicate a significant difference between treatments ($P < 0.05$).

4.3.2 Soil respiration

During the 5-year crop rotation, R_h ranged between 0.11 and 5.22 gC m⁻² d⁻¹ in CT and between 0.03 and 4.39 gC m⁻² d⁻¹ in MT, while R_s was between 0.14 and 8.30 gC m⁻² d⁻¹ in CT and between 0.14 and 6.24 gC m⁻² d⁻¹ in MT.

The cumulated values of R_h and R_s for each of the considered periods are reported in Table 4.2. R_h was significantly lower in MT compared to CT during the tilled period of 2007-2009 ($P = 0.010$), 2010 ($P = 0.043$) and 2011 ($P < 0.001$). However, this did not lead to a significant difference in R_h annual sum in 2010 and 2011, but the cumulative R_h was significantly lower in MT in 2007-

2009 ($P = 0.012$) and in the entire 5-year rotation ($P = 0.014$; Table 4.3). Cumulative R_s , instead, did not show any significant difference among treatments during all considered periods, except for period I of 2011, when R_s of MT was significantly lower compared to CT ($P = 0.004$). However, MT caused a significant reduction of R_s considering the annual sum of years 2007-2009 ($P = 0.032$), 2011 ($P = 0.018$) and of the 5-year rotation ($P = 0.007$).

The R_h : NPP ratio was significantly lower in MT during 2007-2009 (0.50 ± 0.03 in CT and 0.39 ± 0.02 in MT; $P = 0.031$) and 2011 (CT = 1.47 ± 0.08 ; MT = 0.87 ± 0.15 ; $P = 0.023$), while during 2010 this ratio was equal to 0.49 ± 0.02 in CT and to 0.57 ± 0.06 in MT (no difference; $P = 0.296$). Considering the whole rotation, R_h : NPP of MT (0.48 ± 0.01) was significantly lower in comparison to CT (0.59 ± 0.03 ; $P = 0.022$). Considering the R_h : R_s ratio, there were no significant differences among treatments for all considered years. In particular, this ratio was 0.53 ± 0.02 in CT and 0.47 ± 0.005 in MT for years 2007-2009, 0.64 ± 0.09 in CT and 0.67 ± 0.04 in MT for 2010, 0.66 ± 0.03 in CT and 0.62 ± 0.08 in MT for 2011 and finally 0.57 ± 0.02 in CT and 0.54 ± 0.01 in MT considering the whole rotation.

Mean weekly R_h (Figure 4.2) showed that both treatments followed the same annual trend with the highest values during the growing season, a part for 2009, when the highest weekly R_h of CT was measured during the fallow period. Generally, mean weekly R_h was found to be significantly lower in MT in comparison to CT during period I (weeks 1 to 3 and 5 to 8 in 2007; weeks 4 to 9 in 2008; weeks 8, 9, 12 and 48 to 52 in 2009; weeks 2 to 8 in 2010 and weeks 1 to 8, 10 to 18 and 48 to 52 in 2011). On the contrary, during period II, R_h was occasionally higher in MT in the first three years (weeks 29 and 30 in 2007, weeks 15 and 20 in 2008 and weeks 17 and 18 in 2009), while in 2010 R_h was lower in MT on weeks 19, 31 and 34 and finally during 2011 there were not differences in R_h among treatments during the growing season. Considering period III, no differences between CT and MT in R_h were found during 2007, while in 2008 R_h was significantly lower in MT on weeks 39, 41, 44, 45, 48 and in 2009 R_h resulted lower in MT only on week 47. In 2010 and 2011, again there were no differences among treatments in R_h in the fallow period.

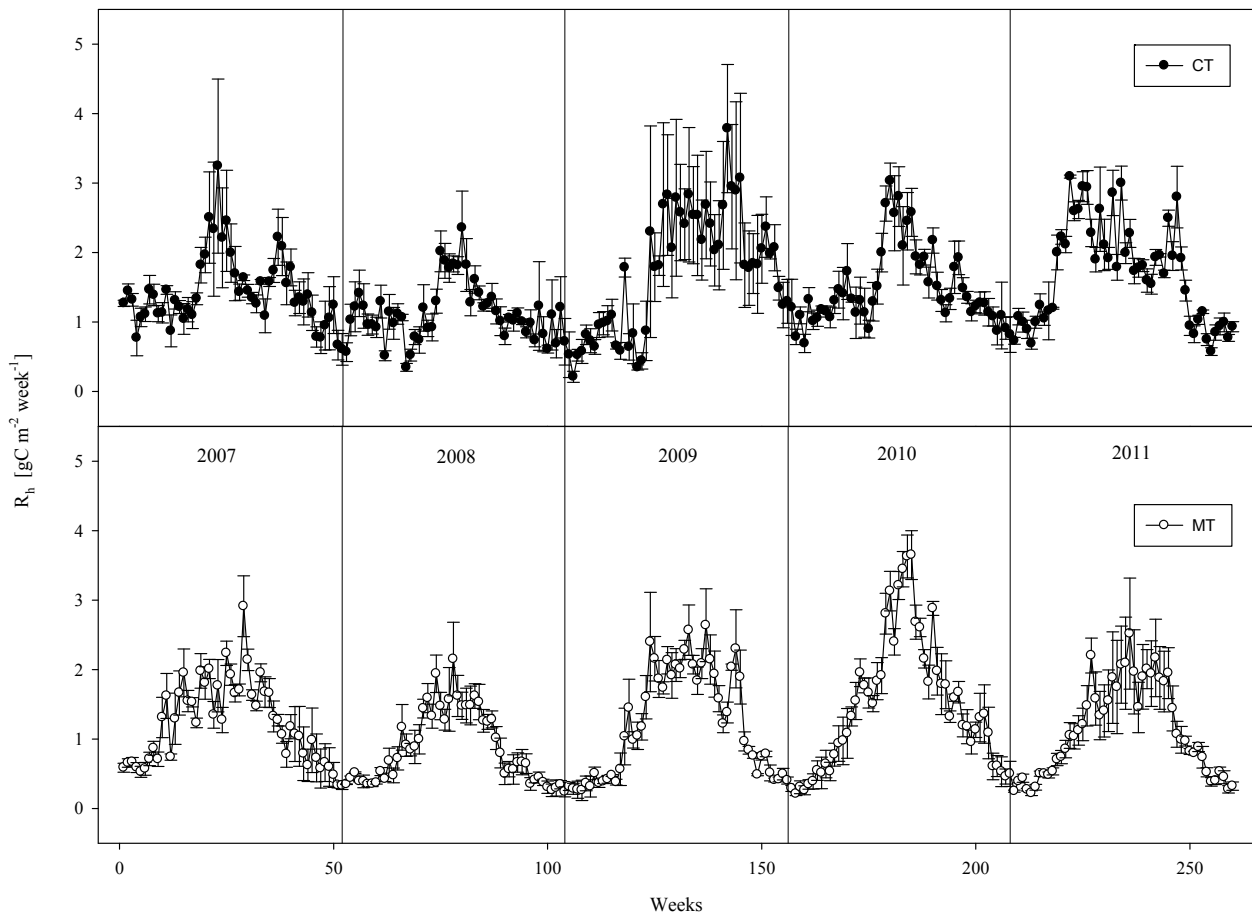


Figure 4.2: Mean weekly soil heterotrophic respiration (R_h) of conventional tillage (CT; filled circles) and minimum tillage (MT; open circles). Vertical bars represent standard error ($n = 3$).

4.3.3 Eddy fluxes

Both eddy stations showed a good agreement between the measured energy fluxes and the energy balance calculated from weather data using net radiation and soil heat fluxes (Figure 4.3). The eddy fluxes of both treatments presented a similar trend during the studied period, except for MT during years 2007-2009, when the periodic harvests of alfalfa reduced GPP leading to ecosystem C losses (i.e. positive NEE; Figure 4.4). Considering the cumulative eddy fluxes of these first three years (Figure 4.5), while there were no differences between treatments during period I of 2007 as alfalfa was not yet planted, in 2008 and 2009 the presence of the crop in MT increased GPP leading to different NEE values in comparison to CT. Accordingly, the $R_e : \text{GPP}$ ratios were equal to 6.00 and 5.01 in 2007, to 4.20 and 0.99 in 2008 and to 3.96 and 0.84 in 2009 for CT and MT, respectively. During the cropping period of 2007-2009, GPP was lower in MT due to the different crop (alfalfa vs. maize). Even though MT R_e was lower than in CT, NEE was still higher

in MT, especially in 2009 due to alfalfa senescence. During this period, R_e : GPP ratios were 0.55 and 0.58 in 2007, 0.68 and 0.65 in 2008 and to 0.61 and 0.67 in 2009 for CT and MT, respectively. Similarly to period I, also in the fallow period the presence of the crop in MT increased GPP leading to a lower NEE in comparison to CT (R_e : GPP of 3.41 and 0.66 in 2007, 3.19 and 0.98 in 2008, 4.10 and 0.78 in 2009 for CT and MT, respectively).

Taking into account 2010 and 2011, when both treatments were planted with the same crop, the major discrepancies among treatments were found during period I, when MT presented a higher GPP in comparison to CT due to the presence of alfalfa in 2010 and of the cover crop in 2011, a lower R_e and thus a lower NEE in both years, even if this trend was more evident in 2011. In particular, the effects of MT during the tilled period were such as to reduce the cumulative NEE by 52% during 2010 and by 63% in 2011. Accordingly, R_e : GPP ratios were equal to 3.57 and 1.81 in 2010 and 4.51 and 1.60 in 2011 for CT and MT, respectively. Taking into account the cropping period, also in 2010 treatment MT led to a lower GPP and R_e in comparison to CT, even though the difference in C emissions by R_e was not enough to compensate the minor GPP leading to a higher NEE in MT. In the same period of 2011 we did not observe the same differences in GPP and R_e between CT and MT, thus leading to a similar NEE. During this period, R_e : GPP ratios were 0.62 and 0.65 in 2010 and 0.78 and 0.76 in 2011 for CT and MT, respectively. In the fallow period of 2010, R_e was lower in MT leading to a lower NEE in comparison to CT, while during period III of 2011 there were not such dissimilarities among treatments in the cumulative eddy fluxes. However, while the R_e : GPP ratio of this period in 2010 is similar for both treatments (CT = 3.41; MT = 3.31), during fallow period of 2011 this ratio was slightly lower in MT (CT = 4.41; MT = 3.60).

Considering annual eddy fluxes (Table 4.3), a part for 2010, both NEE and R_e : GPP ratio were lower in MT, thus indicating a greater propensity to save C by this treatment. This is clearly visible from Figure 4.6. In fact, even though the absolute values of NEE were higher in MT during 2007-2009, the presence of the crop during whole year has meant that the ecosystem acted almost constantly as a C sink (i.e. negative slope of cumulated NEE), while in CT the decrease in GPP after harvest brought the ecosystem to act as a C source (i.e. positive slope). While during 2010 treatments showed a similar trend in NEE, during the first part of 2011 the presence of the cover crop was switching the ecosystem towards a C sink. However, once the cover crop was removed, the ecosystem returned to act as C source (around DOY 100). At the end of the 5-years rotation, CT resulted in a cumulated NEE of -1311 gC m^{-2} while NEE in MT was -1997 gC m^{-2} (-53%). Considering the cumulative fluxes of the whole rotation, the R_e : GPP ratio was 0.86 in CT and 0.77 in MT.

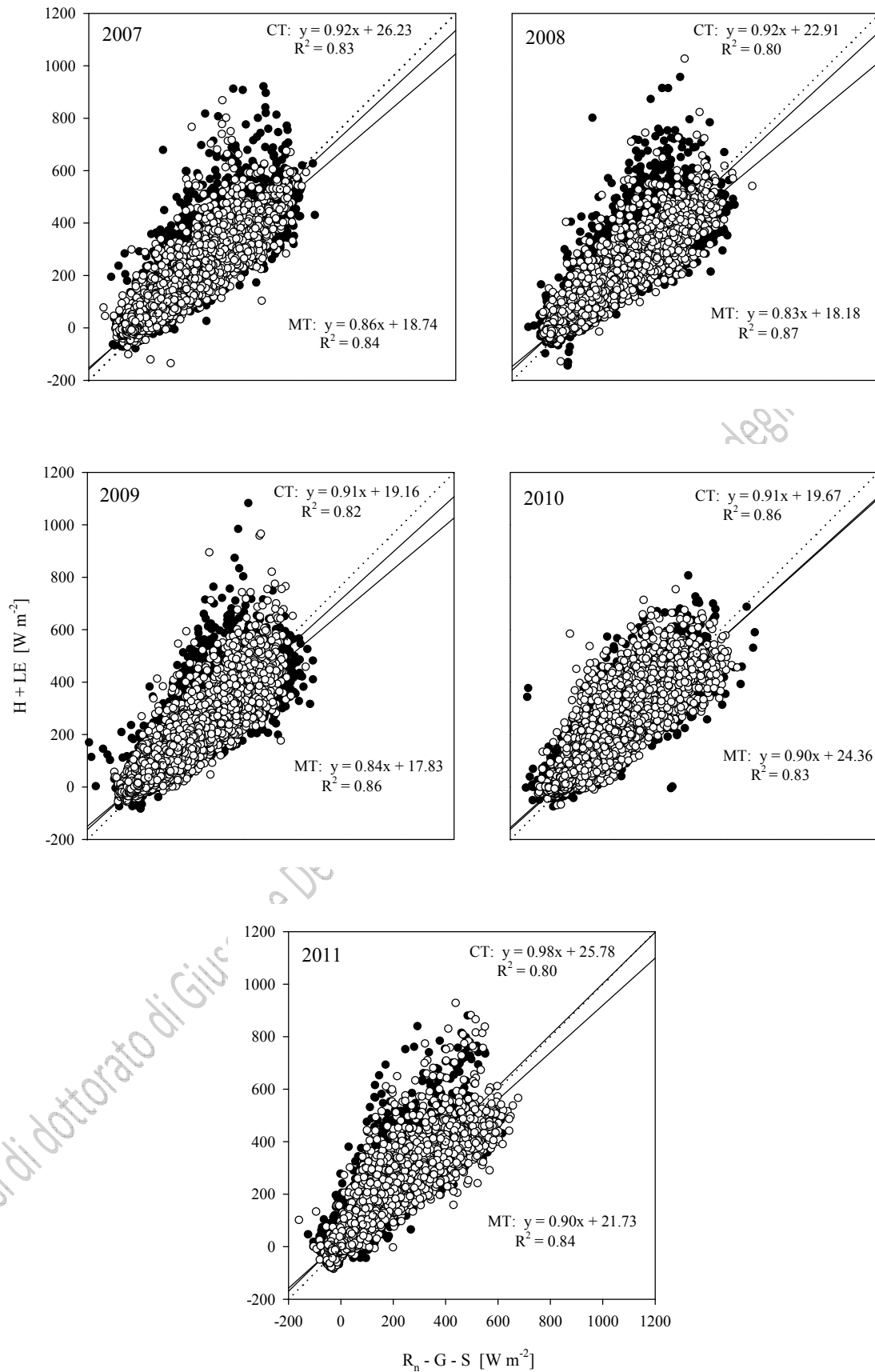


Figure 4.3: Energy balance closure of years 2007 to 2011 based on 30 min data for conventional tillage (CT; filled circles) and minimum tillage (MT; open circles). The dotted line represents 1:1 line.

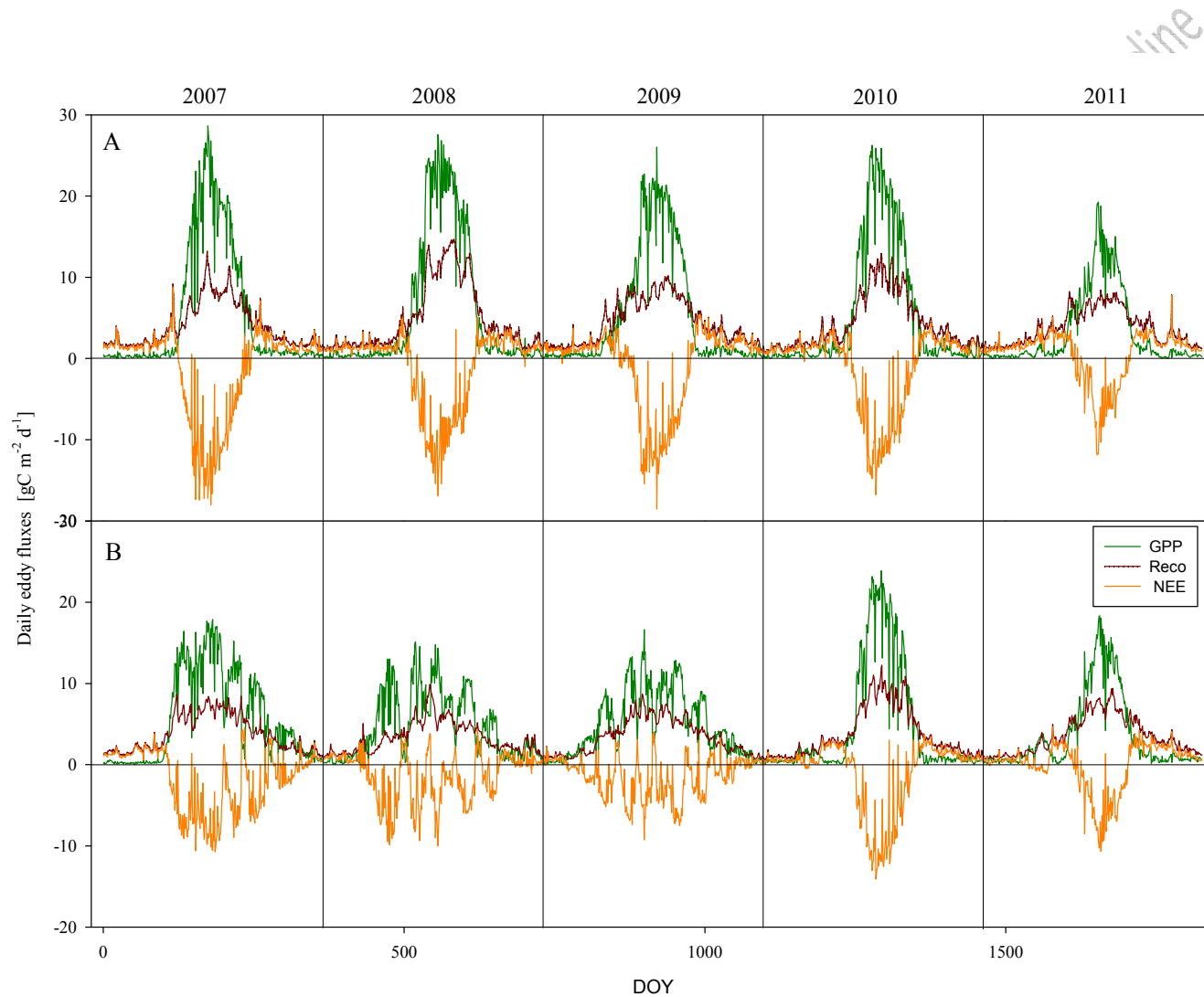


Figure 4.4: Daily gross primary production (GPP), total ecosystem respiration (R_e) and net ecosystem exchange (NEE) for conventional tillage (CT; panel A) and minimum tillage (MT; panel B).

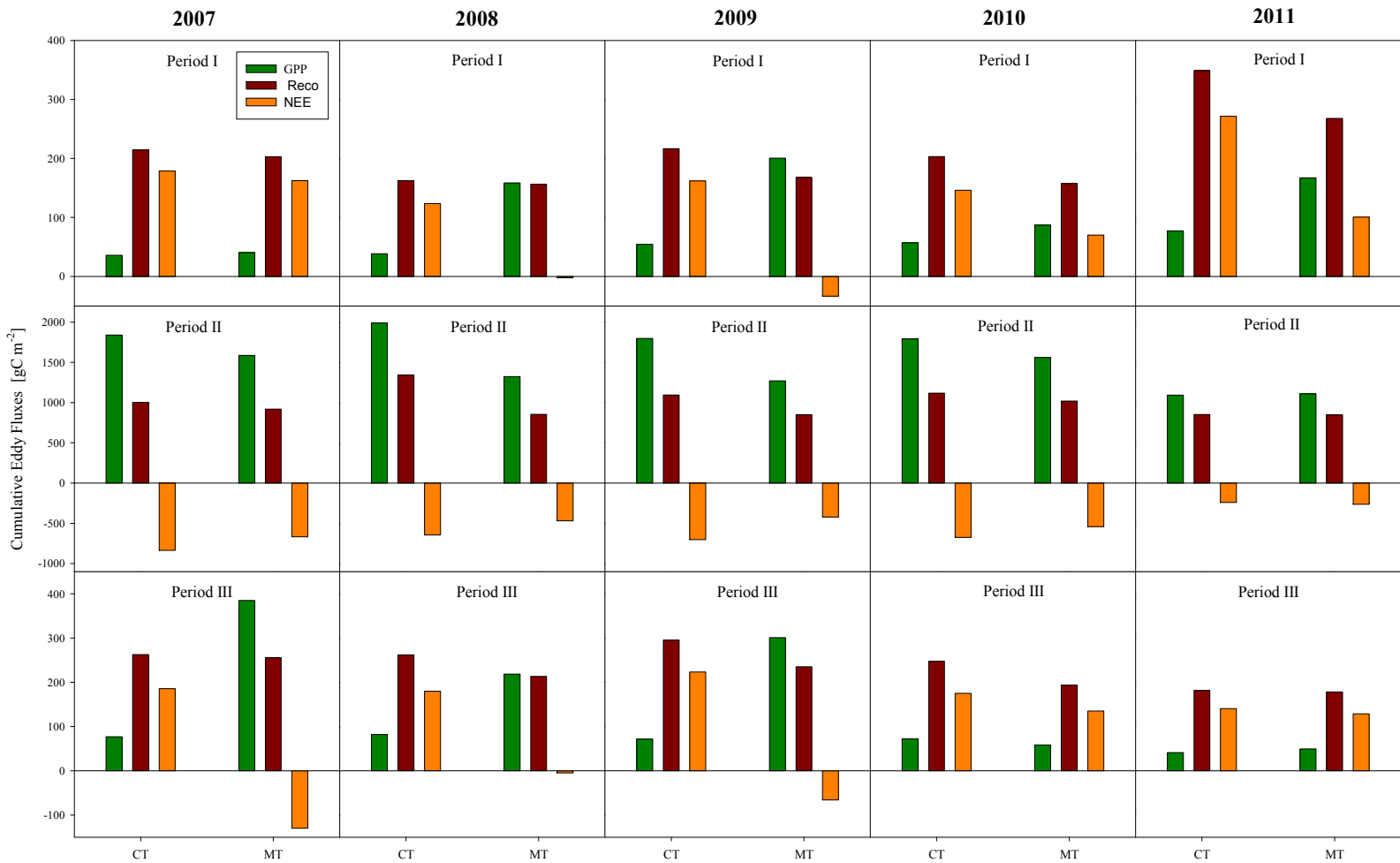


Figure 4.5: Cumulative gross primary production (GPP), total ecosystem respiration (R_e) and net ecosystem exchange (NEE) of conventional tillage (CT) and minimum tillage (MT) for each of the considered periods. During years 2007 to 2009 CT was planted with maize while in MT there was continuous alfalfa. In 2010 both treatments were planted with maize and in 2011 both treatments were planted with soybeans.

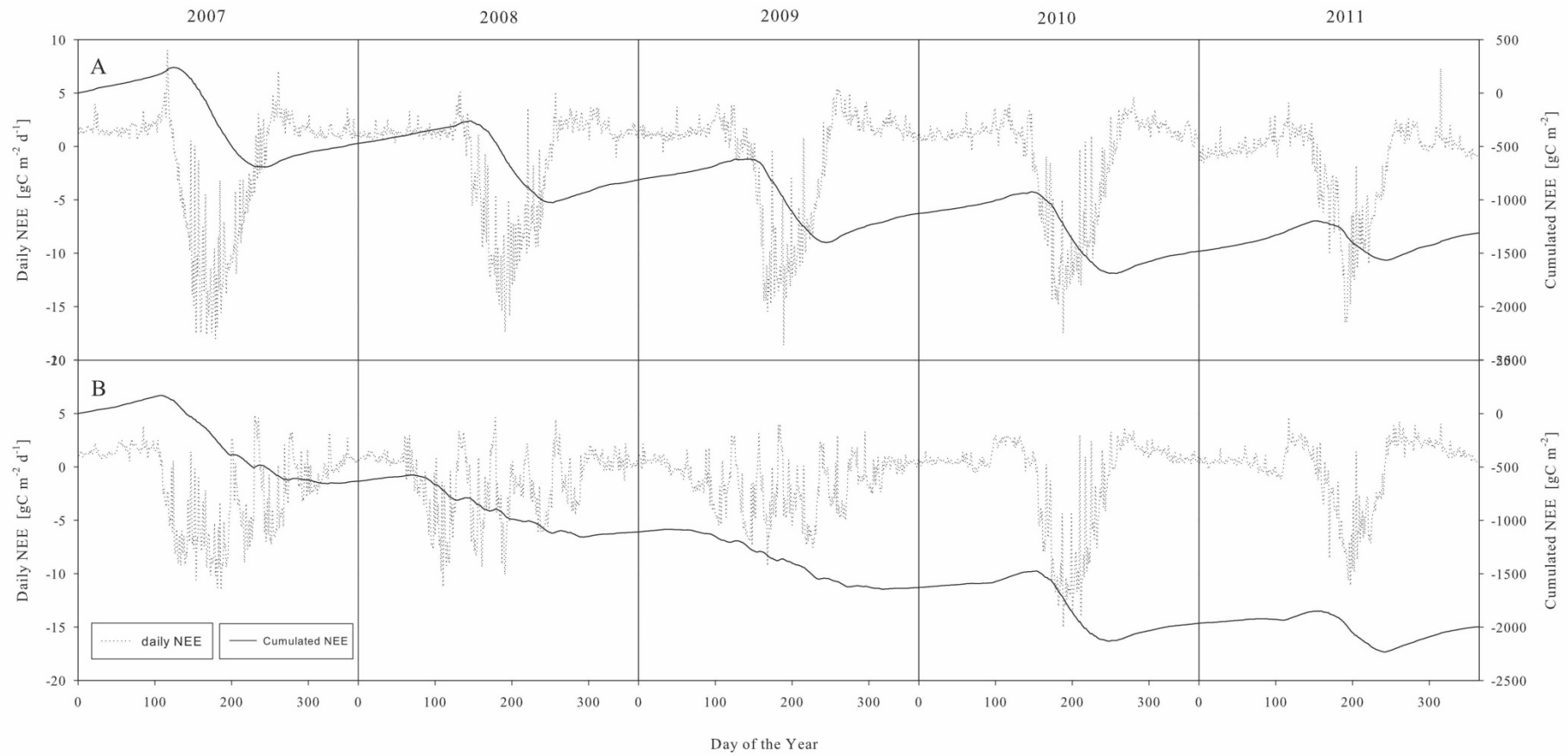


Figure 4.6: Daily and cumulative net ecosystem exchange (NEE) conventional tillage (CT; panel A) and minimum tillage (MT; panel B) during the whole 5-year rotation. The ecosystem is acting as a C sink when the cumulative NEE presents a negative slope and vice versa.

4.3.4 Crop productivity and overall Carbon budget

Measured total crop biomass (Table 4.3) was not influenced by MT neither during 2007-2009 despite treatments were planted with different crops ($P = 0.573$), nor in 2010 with maize ($P = 0.268$) nor in 2011 with soybeans ($P = 0.343$). The 2007-2009 harvest was significantly higher in MT ($P < 0.001$). Conversely, the grain yield in MT was significantly lower than in CT in 2010 ($P = 0.028$), while in 2011 there was no difference in harvest between treatments ($P = 0.450$). Accordingly, the apparent soil C input of MT was lower than in CT for 2007-2009 (1722 ± 32 gC m⁻² in CT and 639 ± 41 gC m⁻² in MT; $P < 0.001$), while MT had no effect on the apparent soil C input of 2010 ($P = 0.486$) and 2011 ($P = 0.269$), which amounted to 587 ± 33 gC m⁻² yr⁻¹ for CT and to 518 ± 84 gC m⁻² yr⁻¹ for MT in 2010, and to 237 ± 9 and 262 ± 17 gC m⁻² yr⁻¹ in 2011, for CT and MT respectively.

The Carbon Use Efficiency in the cropping period (i.e. whole year for alfalfa) was not influenced by MT (2007-2009 $P = 0.508$; 2010 $P = 0.966$ and 2011 $P = 0.411$), in fact the CUE was 0.56 ± 0.01 in CT and 0.56 ± 0.00 in MT for 2007-2009, 0.61 ± 0.02 in CT and 0.62 ± 0.07 in MT during 2010, while in 2011 this index decreased to 0.38 ± 0.02 and 0.42 ± 0.03 in CT and MT, respectively, due to the lower biomass production of soybeans. The Carbon Sequestration Efficiency (CE), indeed, was found to be negatively influenced by MT in 2007-2009 ($P = 0.009$), positively influenced by MT in 2011 ($P = 0.017$), while in 2010 there were no differences in CE among treatments ($P = 0.401$). In particular, CE was equal to 0.09 ± 0.06 in CT and -0.91 ± 0.21 in MT during 2007-2009, 0.07 ± 0.06 in CT and -0.08 ± 0.15 in MT during 2010, while in 2011 CE was -1.59 ± 0.06 and -0.54 ± 0.26 for CT and MT, respectively. Considering all years, CE was significantly higher in CT (-0.07 ± 0.05) in comparison to MT (-0.51 ± 0.03).

The comparison between NEP_{eddy} and NEP_{GA} showed a good agreement during the whole rotation (Figure 4.7). Thus, the NECB was calculated using data from growth analysis in order to test for differences among treatments.

Finally, considering the overall 5-years rotation, MT increased significantly NEP ($P = 0.048$). However, since MT increased also the amount of harvested C ($P < 0.001$), it ended up with a NECB significantly lower in comparison to CT ($P = 0.015$; Figure 4.8).

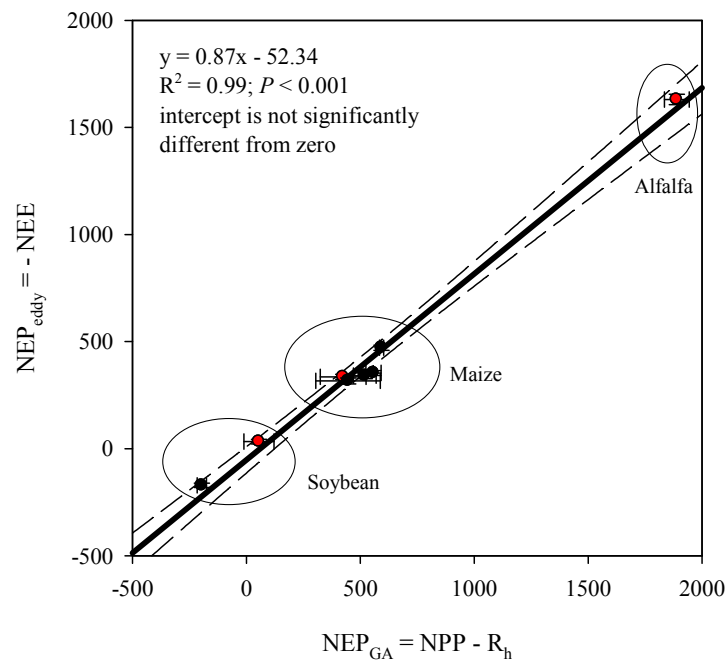


Figure 4.7: Comparison between net ecosystem production measured by Eddy Covariance ($NEP_{eddy} = -NEE$) and net ecosystem production measured through growth analysis ($NEP_{GA} = NPP - R_h$). Circles highlight the different crops planted during the rotation. Data for alfalfa refers to the cumulative NEP for the 2007-2009 period. The black and red dots represent data from conventional tillage (CT) and from minimum tillage (MT), respectively. The dashed lines represent 95% confidence intervals.

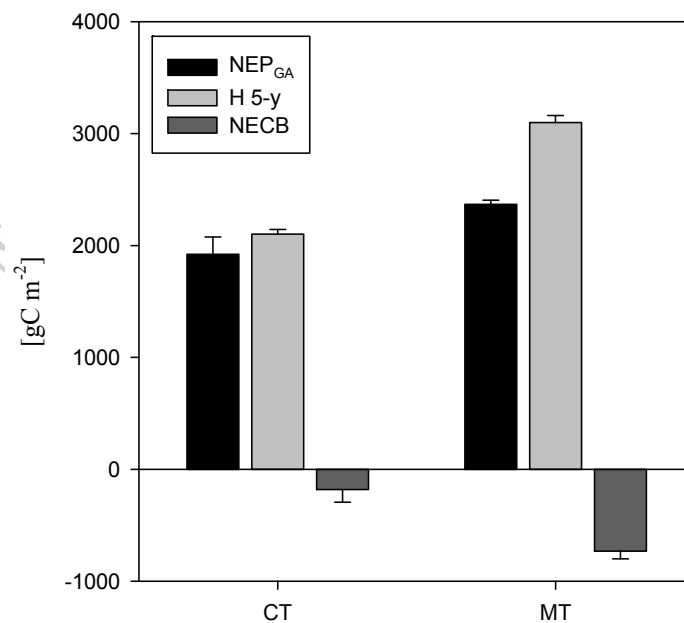


Figure 4.8: Net ecosystem production measured through growth analysis (NEP_{GA}), harvested C (H) and net ecosystem carbon balance (NECB) of conventional tillage (CT) and minimum tillage (MT) for the overall 5-year rotation. Vertical bars represent standard error ($n = 3$).

4.4 Discussion

From our results, MT management was not able to reduce the overall C emissions in comparison to conventional practices, even if it improved the ecosystem C budget outside the growing season. In particular, MT has been effective in reducing R_h during the tilled periods of 2007-2009, 2010 and 2011, when the residues of the previous crop were included into soil after plowing in CT, and thus were more accessible to microorganisms (Dungait *et al* 2012). Moreover, daytime temperatures in MT were lower than CT during the tilled period since crop residues normally reduce surface temperature by reflecting solar radiation and insulating the soil surface (Shinners *et al* 1993, van Wijk *et al* 1959). This effect could potentially lead to a reduction in C losses through lower respiration rates, as it is a widespread opinion that an increase in temperature would lead to an increase in CO_2 release from soils (Cox *et al* 2000, Luo 2007).

Likewise, eddy covariance data showed the main discrepancies between treatments during tilled periods, when a higher GPP and consequently a lower NEE were found in MT (Figure 4.5). The differences among treatments in eddy fluxes result from the combined effect of plowing in CT and of the presence of alfalfa or of the cover crop in MT. In detail, according to eddy data, MT was able to improve the cumulative NEE of the 5-year rotation (Figure 4.6), contrary to the results of Baker and Griffis (2005), in which eddy covariance measurements did not support the hypothesis that reduced tillage and a spring cover crop will improve the NEP of a corn/soybean rotation, even though they supposed that a fall-planted cover crop (such in our experiment) might be more effective. Thus, year-round cropping could improve NEP in agro-ecosystems, even not considering tillage impacts on C budget. In fact, as reported by Ceschia *et al* (2010), when a summer crop is combined with a cover crop, bare soil periods are shorter and soil C losses could be offset, at least partially, by net C assimilation from the cover crop.

Growth analysis revealed that MT had no effects on total crop biomass (NPP). Conversely, MT increased the harvest during the period 2007-2009 (+47%) as alfalfa has been harvested several times per year thus leading to a higher C export in comparison to maize in CT. Taking into account 2010 and 2011, instead, even if our crop yields are aligned with the mean C export (i.e. harvest) of $462 \pm 251 \text{ gC m}^{-2} \text{ yr}^{-1}$ reported by Ceschia *et al* (2010), MT resulted in a significant reduction (-14%) of the grain yield for maize, while the grain production of soybean was not affected by management. These results perfectly agree with other studies about crop yields of maize and soybean following no-till adoption (Chen *et al* 2011, Ogle *et al* 2012). However, there are contrasting opinions about crop yields under conservation agriculture. In particular, while some experiments found no significant differences in yields (Stavi *et al* 2011, Kumar *et al* 2012, Patrignani *et al* 2012) and some others found an increased productivity, even if in prairies (Lafond *et al* 2011), a recent review sentenced that yields for spring-sown crops with no-till management have often been lower than those with conventional practices (Soane *et al* 2012).

Considering the NECB, at the end of the rotation, the studied cropland acted as a C source under both tested managements (Figure 4.8). The final C balances found in the present study for both treatments are in agreement with the mean NECB of $138 \pm 239 \text{ gC m}^{-2} \text{ yr}^{-1}$ reported by Ceschia *et al* (2010) averaging the results from 15 European cropland sites. Our results agree also with the net loss of $90 \pm 50 \text{ gC m}^{-2} \text{ yr}^{-1}$ estimated by Janssens *et al* (2003) for croplands, even though this value has been recently questioned by studies that suggested that European cropland soils are close to the equilibrium, acting as small sources ($3 \text{ gC m}^{-2} \text{ yr}^{-1}$ in Schulze *et al* 2009; $17 \pm 33 \text{ gC m}^{-2} \text{ yr}^{-1}$ inventory estimation by Ciais *et al* 2010) or as small sinks ($16 \pm 15 \text{ gC m}^{-2} \text{ yr}^{-1}$ in Gervois *et al* 2008; $15 \pm 15 \text{ gC m}^{-2} \text{ yr}^{-1}$ ORCHIDEE-STICS estimation by Ciais *et al* 2010).

Generally, we can conclude that MT resulted in a significantly lower NECB in comparison to CT, even though MT improved the ecosystem C budget in terms of NEP by reducing significantly the C emissions from the ecosystem (R_h). However, MT led to an increase in the C exports from the ecosystem through harvest that caused a reduction in the apparent soil C input and finally in NECB. In fact, even though soil respiration data, R_h : NPP ratio and eddy data showed the tendency by MT to minimize C losses mainly due to the lower R_h in MT during period I, the carbon sequestration efficiency index (CE) clearly highlighted that MT had a negative impact on the C budget at the end of the rotation. In particular, while CE in CT was near the range reported by Ciais *et al* 2010 for croplands (-0.03 to 0.03), in MT this index was significantly lower. Thus, since CE in croplands is closely linked to soil C accumulation or release (Guzman and Al-Kaisi 2010, Ciais *et al* 2010), our results suggest that MT management is not going to increase soil C stock, even if several studies concluded that an increase in SOC through long-term application of no-till is possible (Al-Kaisi *et al* 2005, Abreu *et al* 2011, De Sanctis *et al* 2012). However, taking into account only the last two years of the crop rotation (2010-2011), MT showed to improve significantly the NECB, thus supporting the statement by Bernacchi *et al* (2005), that maize/soybean ecosystems may even turn from C source to C sink in consequence of a conversion to no-till management, and supporting also the results of several studies that found a NBP (i.e. NECB) close to neutrality in maize/soybean rotations, which acted either as low C sinks (Baker and Griffis 2005, Dobermann 2006, Hollinger *et al* 2005, 2006) or low C sources (Verma *et al* 2005, Grant *et al* 2007).

4.5 Conclusions

The overall conclusions of this study are that MT did not bring to large changes in soil physical properties, even though significant differences in mean soil temperature and SWC were detected during some of the periods considered, and especially in the tilled period.

However, MT reduced significantly R_h in comparison to CT during the tilled and the fallow periods. This led to a R_h and R_s significantly lower in MT considering the whole 5-year rotation. Cumulative eddy fluxes highlighted the differences among treatments during the tilled periods, when MT presented a higher GPP thus leading to an improvement in NEE in comparison to CT.

Since there were no differences among treatments in terms of biomass (NPP), the reduction in R_h improved significantly the C budget of the agro-ecosystem in terms of NEP. However, considering the C export from the ecosystem, MT was found to increase the harvest during the first three years of the rotation and to reduce the grain yield of maize, while the grain yield of soybean was not affected by management. Consequently, even if MT was able to reduce the C emissions, the higher amount of harvested C in MT resulted in a significantly lower NECB at the end of the 5-year rotation.

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5

Short-term cropland responses to temperature extreme events during late winter³

Summary

In recent years, several studies have focused on terrestrial ecosystem response to extreme events. Most of this research has been conducted in natural ecosystems, but few have considered agroecosystems. In this study, we investigated the impact of a manipulated warmer or cooler late winter-early spring on the carbon budget and final harvest of a soybean crop (*Glycine max* (L.) Merr.). Soil temperature was altered by manipulating soil albedo by covering the soil surface with a layer of inert silica gravel. We tested three treatments: cooling (Co), warming (W), mix (M) and control (C). An automated system continuously measured soil heterotrophic respiration (R_h), soil temperature profiles, and soil water content across the entire year in each plot. Phenological phases were periodically assessed and final harvest was measured in each plot. Results showed that treatments had only a transient effect on daily R_h rates which did not result in a total annual carbon budget significantly different from control, even though cooling showed a significant reduction in final harvest. We also observed anticipation in seed germination in both W and M treatments and a delay in germination for Co. Moreover, plant density and growth increased in W and M and decreased in Co.

Keywords: climate change, ecosystem manipulation, extreme events, carbon dynamics, soil respiration, soil warming, heat waves.

³ De Simon G, Alberti G, Delle Vedove G, Peressotti A, Zaldei A and Miglietta F (*submitted*). Short-term cropland responses to temperature extreme events during late winter. *Biogeosciences*.

5.1 Introduction

Soil respiration (R_s) is the second largest carbon flux in most ecosystems (Davidson *et al* 2002). Thus, the possible increase in R_s due to climate warming and the consequent feedbacks on climate change have led to a growth in concern about the effects of climate change on soil organic matter dynamics (Cox *et al* 2000, Jones *et al* 2003, Knorr *et al* 2005, Davidson and Janssens 2006, Heimann and Reichstein 2008).

The soil carbon pool is reported to be quite considerable (from 1220 to 1576 PgC for the 0-100 cm layer; Tarnocai *et al* 2009), while estimated global R_s in 2008 amounted to 98 ± 12 PgC, with an increasing rate of 0.1 PgC yr^{-1} between 1989 and 2008, implying a global R_s Q_{10} of 1.5 (Bond-Lamberty and Thomson 2010). Hence, understanding the sensitivity of respiratory processes to temperature is central for quantifying the climate-carbon cycle feedback (Mahecha *et al* 2010).

So far, several studies have focused on the effects of gradual climatic trends (e.g. global warming, increasing atmospheric CO₂ concentration; Jentsch *et al* 2007), but in recent years there has been a growing interest in the impacts of climate extremes and climate variability on the carbon cycle (Easterling *et al* 2000, Jentsch *et al* 2007). In fact, such extreme events can have an even greater influence on ecosystems and societies than gradual shifts in mean temperatures and precipitation regimes (Jentsch and Beierkuhnlein 2008). Moreover, there is general agreement about the fact that there will be an increase in both mean values and overall variability of occurrence of extreme weather events (Meehl *et al* 2000, Jentsch *et al* 2007, Jentsch and Beierkuhnlein 2008). In particular, temperature extremes (heat waves) are predicted to become more frequent, intense and longer lasting (Karl and Trenberth 2003, Meehl and Tebaldi 2004), especially in certain areas like central-western Europe, where the length of summer heat waves has doubled and the frequency of hot days has almost tripled in recent decades (Della Marta *et al* 2007). Regarding cold extremes, instead, a recent paper foresees an increased likelihood of cold in the European region (Fereday *et al* 2012).

Since the total global cropland area amounted to 1.53×10^9 ha at the end of the last millennium (Biradar *et al* 2009), and agriculture has been estimated to account for 10–12% of total anthropogenic greenhouse gas emissions in 2005 (Loubet *et al* 2011), it is clear that studying the effects of climate change and extremes on agro-ecosystems is a key issue in carbon dynamics and climatic research. Here, we investigate the response of soil respiration and ecosystem productivity to soil temperature manipulation (warming and cooling) in an agro-ecosystem during late winter-early spring, when soils are usually ploughed and soil organic matter is more accessible for micro-organisms (Dungait *et al* 2012). In particular, the specific objective of this work is to assess the response of a soybean crop (*Glycine max* (L.) Merr.) to a manipulated warmer or colder late winter-early spring, particularly focusing on soil heterotrophic respiration (R_h) and final harvest.

Our experimental hypotheses were that warm extreme events do not affect crop carbon input (C_{input}), while a warmer late winter-early spring leads to an increase in R_h (carbon output; C_{output}) and, consequently, to a detectable loss of soil carbon (C_{budget}). On the contrary, we hypothesized that a colder late winter-early spring leads to lower C_{output} and thus a higher C_{budget} .

5.2 Methods

5.2.1 Study site and experimental design

The experiment was carried out in Beano (46°00' N 13°01'E, 65 m a.s.l.), north-eastern Italy. Mean annual temperature at the site is 13.7 °C and mean annual precipitation is around 1200 mm (2000-2007). An analysis of the occurrence of local climate extremes was performed using data for two decades (1991-2000 and 2001-2010) at a meteorological station close to the study field (~10 km). In particular, the average and standard deviation (σ) of the daily maximum (T_{max}) and minimum temperature (T_{min}) in the winter-early spring period (from January to April) were calculated. Then, similarly to De Boeck *et al* (2010), we considered heat waves as periods encompassing at least 7 consecutive days above $T_{\text{max}} + \sigma$ and cold waves as periods encompassing at least 7 consecutive days below $T_{\text{min}} - \sigma$. The mean length of extreme events was expressed as the number of days above or below temperature threshold divided by the number of events.

The location is characterized by intensive, fertilized and irrigated farming. Soil is classified as a Chromi-Endoskeletal Cambisol (FAO 2006) with the following characteristics in the 0–30 cm horizon: total soil organic carbon (SOC) = $48.4 \pm 8.5 \text{ tC ha}^{-1}$, total N = $4.2 \pm 1.1 \text{ t N ha}^{-1}$, soil bulk density = $1.25 \pm 0.15 \text{ g cm}^{-3}$, soil field capacity = 23% v/v, wilting point = 12% v/v, and pH = 7.1 ± 0.02 (Alberti *et al* 2010). In this field, irrigated maize (*Zea mays* L.) has been cultivated during the last 30 years. In winter, the soil is ploughed to a depth of 0.35m, while in spring, soils are ploughed to 0.05m in preparation for sowing.

The experiment started on March 1st 2011 and lasted 1 year until February 28th 2012 in order to complete the annual carbon budget. A soybean crop (*Glycine max* (L.) Merr.) was sown on May 4th 2011 (DOY 124), during the effective treatment period (see below for details).

The experiment setup consisted in 12 plots (3 replicates \times 4 treatments): 9 plots of $5 \times 2.5 \text{ m}$ (treated) and 3 control plots of $10 \times 10 \text{ m}$ deriving from a previous experiment (Alberti *et al* 2010). The plots were arranged in three blocks. Soil respiration measurements were performed every 2 hours using three closed dynamic soil respiration systems based on the measurement of the increase in CO_2 concentration within an automated chamber during a fixed amount of time using a non linear regression method (Delle Vedove *et al* 2007, Alberti *et al* 2010). Heterotrophic respiration (R_h) was measured using two automated chambers per plot. Soil below the chambers

was isolated with a root exclusion stainless-steel cylinder opened at both ends (32 cm diameter, 40 cm height). The steel cylinders were placed in the field after sowing and removed after final harvest. Soil temperature profiles (four type-T thermocouples for each depth: 0, 2.5, 5 and 10 cm depth, the superficial ones protected from direct solar radiation) and soil water content (Decagon EC-5; 5-10 cm depth) were also continuously monitored in each plot. All variables were measured at 0.1 Hz and then averaged half hourly. Air temperature, humidity (HMP45AC, Vaisala), and precipitation were measured at a nearby weather station (Alberti *et al* 2010).

5.2.2 Warming-Cooling method

We adopted a passive method to alter soil temperature, which consisted of changing soil surface albedo by covering it with a layer (0.5 cm thickness) of inert silica gravel (95.7% SiO₂; pH 7-8 in water solution; density 2.65 g cm⁻³; granulometry 1.2-1.8 mm). Using gravel of two different colours (black and white), we set up 4 treatments: Cooling (Co; white gravel), Warming (W; black gravel), Mix (M; 4:1 black and white gravel) and Control (C; bare soil). The main advantages of this system are the low cost and the fact that no electrical power is required, while the main disadvantage is the inability to determine, a priori, a soil temperature range. Moreover, since this method depends on incident radiation over soil, treatments were not effective at night time, on cloudy-rainy days, or after complete crop canopy closure. However, the aim of the experiment was to test the effects of soil temperature manipulation mainly outside the growing season (i.e. late winter-early spring).

5.2.3 Phenology and ecosystem productivity

Seed germination was monitored in each plot and seed birth trend for each treatment interpolated through a logistic function. The day when the first derivative of the logistic curve (i.e. seed birth rate) was at a maximum was assumed to be the day of germination for the treatment. Moreover, during the growing season, the height and phenological phases of the crop, according to Fehr *et al* (1971), were periodically assessed, in order to detect if there were differences in crop development due to treatments.

Crop productivity and crop yield were assessed at final harvest by destructive sampling on September 22nd 2011 (DOY 265). All plants in a 1.7 × 1.0 m subplot per plot were collected. After oven-drying at 70°C for 48 h, above- and below-ground dry biomass and dry grain yield were determined. Furthermore, on a subsample of 10 plants, root:shoot ratio and harvest index were calculated. Crop residues after harvest were estimated on a 1 m² subplot for each plot.

Considering that we are dealing with a crop rotation, the carbon input to the ecosystem (C_{input}) was assumed to be equal to the sum of the crop residues of the previous crop year (CR_{y-1} ; 2010) and those of the studied crop year (CR_y ; 2011), minus the amount of carbon respired between the harvest of the previous year and the beginning of the experiment (Rh_{y-1} ; already monitored in control plots by the same soil respiration systems):

$$C_{input} = CR_{y-1} + CR_y - Rh_{y-1}$$

Assuming that the carbon losses from the ecosystem (C_{output}) during the study year were equal to the cumulative heterotrophic respiration (Rh_y), the total carbon budget (C_{budget}) was calculated as follows:

$$C_{budget} = C_{input} - C_{output} = CR_{y-1} + CR_y - Rh_{y-1} - Rh_y$$

5.2.4 Data analysis

Measured soil respiration data were averaged across four periods during the day (00:00-6:00; 6:00-12:00; 12:00-18:00; 18:00-24:00). Days without at least three periods of data were discarded from further analysis so as not to under- or over-estimate soil respiration since a complete daily trend was not available. Moreover, data for days when all three replicates per treatment were not available were also discarded. In total, 14% of days were not considered in the analysis. Missing data were then gap-filled using surface temperature according to van't Hoff equation (Lloyd and Taylor 1994):

$$R_h = Ae^{kT}$$

where R_h is soil heterotrophic respiration and T is soil surface temperature (Pavelka *et al* 2007). Coefficients A and k were derived by non-linear regression. Sensitivity of soil respiration to soil temperature (Q_{10}) was then calculated as:

$$Q_{10} = e^{10k}$$

Data were analyzed at the end of the year and independently for the following five periods: period I, pre-treatment (DOY 60-67); period II, effective treatment period (from treatment application to complete crop canopy closure, DOY 68-158); period III, after complete crop canopy closure (from complete crop canopy closure to final harvest, DOY 159-264); period IV, after final harvest period (from final harvest to ploughing, DOY 265-326); period V, after ploughing (DOY 327-60). The slope of the cumulative soil respiration curve of each period was considered as the mean daily heterotrophic respiration rate of that period.

The effects of the various treatments on soil albedo, soil temperature, and water content were tested by repeated measures ANOVA with post-hoc tests (Tukey test), while the effects of treatments on daily heterotrophic respiration rates and ecosystem productivity were tested by one-way ANOVA (post-hoc Tukey test). Analyses were performed with SPSS (© IBM Corp.). All errors presented in text and graphs are standard error of the mean unless reported otherwise.

5.3 Results

5.3.1 Soil temperature and soil water content

Both maximum and minimum mean air temperatures (T_{\max} , T_{\min}) and their standard deviations were larger over the period 2001-2010 in comparison to 1991-2000 (T_{\max} increased from 12.2 ± 5.3 to 13.0 ± 5.6 °C, T_{\min} from 1.8 ± 4.5 to 2.8 ± 4.6 °C [mean \pm standard deviation]). The occurrence of heat waves and their mean length also increased, growing from 6 heat waves lasting 9 days in 1991-2000 to 9 heat waves with a duration of 14 days in 2001-2010. Cold waves also increased from 4 to 8 events per decade, however their mean length decreased from 13 to 10 days. Thus, average temperatures, their variability and the occurrence of heat and cold waves during late winter-early spring increased in the last decade, in agreement with the expected climatic trends.

All treatments significantly modified soil surface albedo in comparison to C ($P < 0.001$; Mean albedo: Co 62.6%; W 9.6%; M 15.7%, C 22.5%). Thus, while there were not any significant differences in soil temperature at any depth among treatments before gravel application (DOY 60-67; $P > 0.05$), changes in soil albedo significantly modified soil temperatures during effective treatment periods (period II; $P < 0.001$; figure 5.1(a)). In particular, maximum soil temperature deviations from control were obtained at the soil surface and were equal to -6.8 °C for Co and +5.7 °C for W treatment, while the mean differences in soil temperature at 5 cm depth during period II amounted to -3.00 ± 0.12 , $+2.06 \pm 0.08$ and $+1.24 \pm 0.09$ °C for Co, W and M, respectively. During this period, all treatments created a quite homogeneous soil temperature alteration along the soil profile, at least up to 10 cm depth (data not shown). Soil temperature diurnal fluctuations were wider in W and narrower in Co (compared to C), but in all treatments they were gradually smaller as depth increased.

After complete crop canopy closure (period III; DOY 159-264), there were no significant differences in soil temperature among treatments, except for Co, which had significantly lower soil temperatures compared with C ($P = 0.008$) during the first days of this period due to a delay in crop development (see below). In the first days after final harvest (period IV; DOY 265-326), soil temperature of treatments W and M were significantly ($P < 0.001$) higher than C as harvest residues had not been redistributed over the soil yet, since plants were collected for laboratory

measurements. Finally, after ploughing (period V; DOY 327-60), no significant differences in soil temperature were detected ($P > 0.05$).

Regarding soil moisture, during both period I and II, no significant differences were detected among treatments ($P > 0.05$). Unfortunately, just after canopy closure lightning damaged most of the soil water content sensors. Thus we were not able to assess if there was a difference in soil water content or not in the subsequent periods. However, because the crop is irrigated (a total of 310 mm during period II and III), soil water content was always close to field capacity (0.25 ± 0.04 [mean \pm standard deviation] considering period III, IV and V; data from TDR at the weather station).

5.3.2 Heterotrophic respiration

Daily soil heterotrophic respiration ranged from 0.15 to 7.95 gC m⁻² d⁻¹ with an annual average equal to 1.77 ± 0.07 , 1.27 ± 0.05 , 1.96 ± 0.23 and 1.48 ± 0.08 gC m⁻² d⁻¹ in C, Co, W and M, respectively. There were no statistically significant differences in mean daily R_h rates before gravel application (i.e. slope of the cumulative respiration; $P > 0.05$), while during period II R_h rates in Co and W were less than and greater than C, respectively ($P = 0.117$ and $P = 0.007$; figure 5.1(b)). In this period, mean daily R_h rates were 2.81 ± 0.13 in C, 2.20 ± 0.21 in Co, 3.73 ± 0.43 in W and 2.78 ± 0.20 gC m⁻² d⁻¹ in M. In period III, Co and M treatments had significantly lower daily R_h rates compared with C ($P < 0.01$). R_h rates presented the same trend in all treatments: an increase from period I to period II followed by a decrease during periods III, IV and V. For all treatments, maximum daily R_h rates were found in period II.

R_h increased exponentially with soil temperature in all treatments, however the van't Hoff equation explained only a small part of variance (C: $R^2=0.46$; Co: $R^2=0.09$; W: $R^2=0.28$; M: $R^2=0.12$; $P < 0.0001$). Calculated Q_{10} values were equal to 1.63 ± 0.12 , 1.20 ± 0.07 , 1.50 ± 0.10 and 1.28 ± 0.07 for C, Co, W and M, respectively. Q_{10} of Co was significantly lower than the Q_{10} of C. Seasonal patterns of R_h were similar for all treatments: we observed an increase in spring as temperature increased, a decrease thereafter to values around 1-2 gC m⁻² d⁻¹ and a further decline in autumn after final harvest.

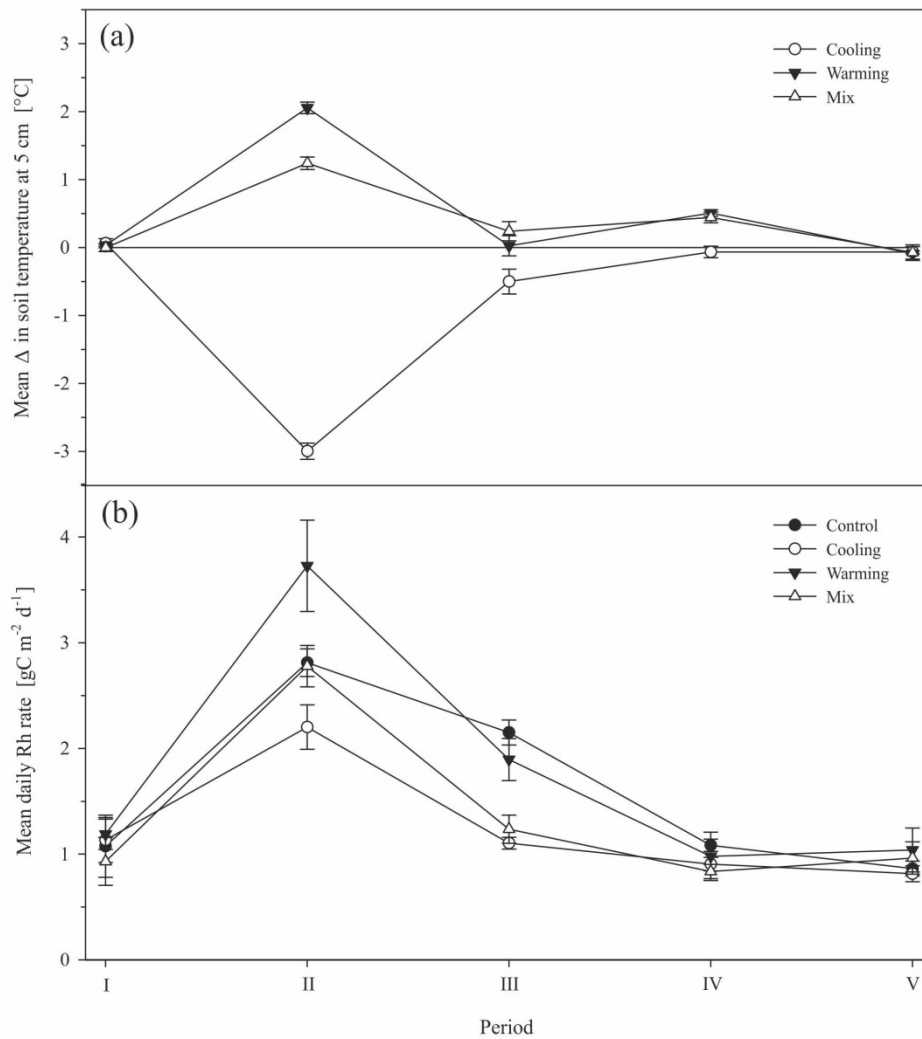


Figure 5.1: Mean difference in soil temperature at 5 cm depth (treatment – control; (a)) and mean daily heterotrophic respiration rates by treatment (b) for each of the study periods considered (I to V). Period I: pre-treatment (DOY 60-67); Period II: effective treatment period (from treatment application to crop canopy closure, DOY 68-158); Period III: after crop canopy closure (from crop canopy closure to harvest, DOY 159-264); Period IV: after harvest period (from harvest to ploughing, DOY 265-326); Period V: after ploughing (DOY 327-60). Vertical bars represent standard error ($n = 3$).

5.3.3 Phenology and ecosystem carbon budget

Treatments had a transient effect on crop development (i.e. on succession of phenological phases). Cooling treatment caused a lower plant density compared to C at all sampling dates (figure 5.2(a)), even if not significant ($P > 0.05$), while in W, plant density was always higher than C (significantly only on May 13th, 9 days from sowing; $P = 0.015$). However, at the end of growing season, plant density was not significantly different between any of the applied treatments and C ($P > 0.05$).

Concerning seed germination (first stage of Fehr-Caviness scale; Fehr *et al* 1971), soil temperature manipulation led to earlier germination in comparison to C in both W and M treatments (-4 days), whilst germination was delayed by 3 days in Co (figure 5.2(b)). The survey of crop development through phenological phases (table 5.1) showed that treatments C and W approximately followed the timetable for phenological phases reported by Fehr and Caviness (1971) in Setiyono *et al* (2010). Conversely, in the cooling treatment, phase VE was delayed by 8 days, V6 by 22 days, R5 by 27 days and finally R6 by 12 days. In comparison to C, Co presented a delay of one or more stages during the entire growing season. Nevertheless, at the end of the crop year, plants reached full maturity in all treatments.

Regarding crop height, plants were significantly smaller in Co than C from May 30th ($P < 0.05$), while plant height was not significantly different from C in W on any sampling date ($P > 0.05$). Finally, plant height was significantly lower in M compared to C only during the last part of the growing season (from August 3rd; $P < 0.05$).

As for the carbon budget (figure 5.3), there were no significant differences in C_{input} among treatments ($P = 0.46$), even if a significant reduction in harvest was recorded for Co in comparison to C at the end of the growing season (-37.5%; $P = 0.02$), while in W and M final harvest was not significantly different from C (final harvest: 4.0 ± 0.3 in C, 2.5 ± 0.1 in Co, 4.0 ± 0.4 in W and 3.4 ± 0.1 $t_{DM} ha^{-1}$ in M). Annual total R_h (C_{output}) and total carbon budget (C_{budget}) of all treatments did not differ significantly from C ($P > 0.05$).

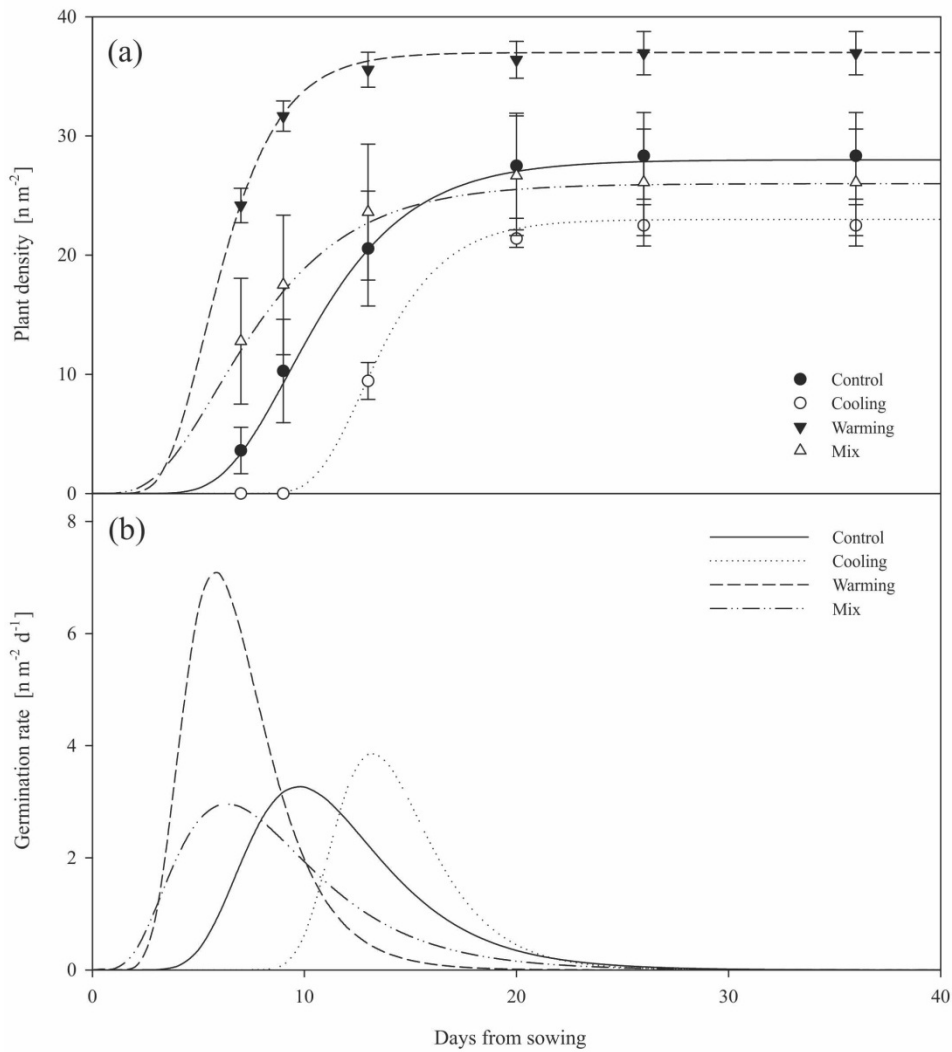


Figure 5.2: Crop development: plant density (a) and germination rate (b). Vertical bars represent the standard error ($n = 3$).

Figure 5.3: Total carbon input (C_{input}), carbon output (C_{output}) and carbon budget (C_{budget}) of the studied agro-ecosystem. Negative values represent carbon losses. Vertical bars represent the standard error ($n = 3$).

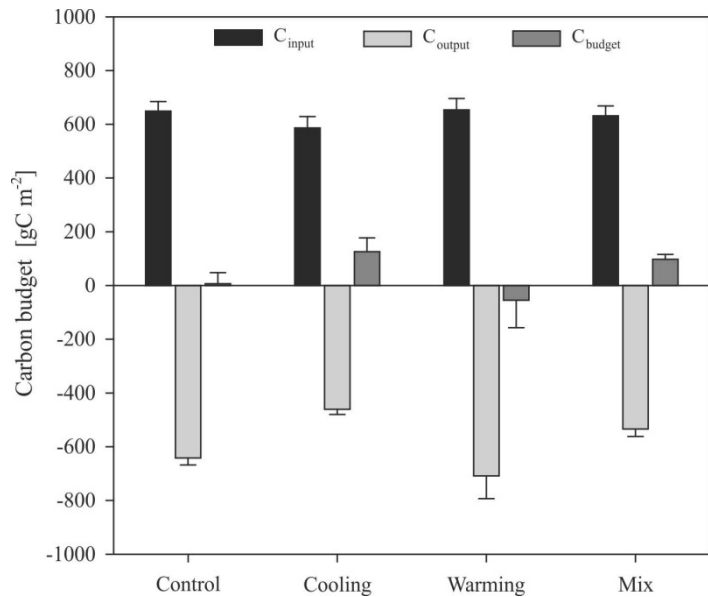


Table 5.1: Cultural practices and phenological phases according to Fehr-Caviness scale (Fehr *et al* 1971). Phenological phase abbreviations: VE = Emergence; V1 = First node; V2 = Second node; Vn = nth node; R1 = Beginning bloom; R2 = Full bloom; R3 = Beginning pod; R4 = Full pod; R5 = Beginning seed; R6 = Full seed.

Date	Days from sowing	Phenological Phase			
		Control	Cooling	Warming	Mix
11/05/2011	7	VE	-	VE	VE
13/05/2011	9	V1	-	V2	V1
17/05/2011	13	V2	VE	V2	V2
24/05/2011	20	V2	V1	V3	V2
30/05/2011	26	V3	V2	V3	V3
09/06/2011	36	V4	V3	V5	V5
14/06/2011	41	V5	V3	V6	V5
20/06/2011	47	V6	V4	V6	V5
30/06/2011	57	R1	V6	R1	R1
03/08/2011	91	R5	R4	R5	R5
09/08/2011	97	R5	R4	R5	R5
17/08/2011	105	R6	R5	R6	R6
24/08/2011	112	R6	R6	R6	R6
Cultural practices					
19/04/2011	-15	Weed control: 5 kg/ha Ammonium sulphate (N 21%) + 2.5 kg/ha Glifosate 36%			
04/05/2011	0	Soybean sowing: Nikko Dekalb 1-			
06/05/2011	2	Weed control: 1.2 l/ha S-metolachlor 86,5% (960 g/l) + 0.8 l/ha Linuron 36,5% (425 g/l)			
10/05/2011	6	Pest control: 2 kg/ha Metaldehyde 4.9% + 2 kg/ha Metaldehyde 3.5%			
11/06/2011	39	Weed control: 2.2 l/ha Cicloxidim 21% (200 g/l) + 7 g/ha Methyl Tifensulfuron 75% + 1 l/ha Imazamox 3,7% (40 g/l)			
22/09/2011	145	Harvest			
23/11/2011	203	Plowing (35 cm depth)			

5.4 Discussion and conclusions

In this study we were able to enhance/decrease soil temperatures in a realistic way, thus mimicking the effects that might be associated to extreme events (i.e. both cold and heat waves; Meehl *et al* 2000, Jentsch and Beierkuhnlein 2008).

There is a large consensus that future increases in temperature will lead to an increase in heterotrophic soil respiration, suggesting that an overall increase in the frequency of extreme temperature events (heat waves) might lead to a substantial emission loss of soil carbon from croplands to the atmosphere. This could eventually lead and to a positive biospheric feedback to global warming (Granier *et al* 2007, Heimann and Reichstein 2008, Ciais *et al* 2005). On the other hand, the occurrence of cold waves, especially during late winter-early spring, might have opposite effects.

The overall net effect of extreme events on the carbon balance of a cropping system is the result of the difference between carbon gains and losses. When soil water is in ample supply, as in irrigated systems, an increase in temperature may indeed translate into increased soil carbon losses. Our results show that heterotrophic respiration was stimulated by soil warming possibly leading to a rapid depletion of the most labile soil carbon stock. As a consequence, the enhanced respiration pulse was followed by reduced respiration rates when the substrate, and not the temperature, subsequently became limiting (figure 5.4). It is also interesting to consider that a higher soil temperature promoted initial growth of the crop by affecting, to some extent, its phenology: crop germination was brought forward by 4 days in response to soil warming, in agreement with Menzel *et al* (2006) who predicted an earlier onset of germination of 2.5 days °C⁻¹ in a future global warming scenario. However, this did not translate into an earlier flowering date and, more importantly, into a larger biomass and crop yield at the end of the growing season, an effect that was also observed in the warming experiments analyzed by Rustad *et al* (2001) and, more recently, by Wu *et al* (2011). Our hypothesis is that the faster soil organic matter decomposition, which is driven by increased soil temperature, leads to a faster mineralization in a period in which the crop, and its rooting system, is still unable to uptake most of the nutrients that can become available in the soil. This was demonstrated in the current study given the fact that the net carbon balance ($C_{input} - C_{output}$) of the crop grown on artificially warmed soil was the same as that in the control (figure 5.3). This suggests substantial homeostasis exists in the carbon balance when observed on a seasonal timescale, that finally restricts the effect of heat waves occurring during late winter-early spring mainly because of substrate limitations to respiration in the last part of the season (Kirschbaum 2004, Eliasson *et al* 2005, Knorr *et al* 2005, Davidson and Jentsch 2006, Hartley *et al* 2007). However, such an effect may also be attributed to changes in microbial diversity and physiology (Allison *et al* 2010), but this second hypothesis is unlikely due to the short duration of the warming effect that was considered in this study. It is worth noting,

that this result is of interest for other types of investigations and in particular for ongoing studies that are trying to address the effect of changes in soil surface albedo on the carbon balance and crop productivity. For instance, the agricultural use of biochar, the dark carbonaceous residue of biomass pyrolysis, may in fact have similar effects to our soil warming treatment as its application prior to sowing can lead to a substantial decrease in surface albedo in the period that precedes full canopy cover (Genesio *et al* 2012). Our study shows that such changes in the surface energy balance do in fact alter soil temperature, cause detectable priming of soil organic matter decomposition and enhance soil CO₂ efflux during late winter-early spring. However, on a seasonal timescale, there is hardly any affect on the overall carbon budget of the crop, unless other effects of the amendant (i.e. increased water holding capacity or improved plant nutrition) cause an increase in crop yields.

As expected, heterotrophic respiration was slightly diminished in the soil cooling treatment, and the overall carbon savings that were made during late winter-early spring could not be compensated by higher respiration rates during the crop growing period. In fact, when the soil warmed up and its temperature became comparable to that of the control, heterotrophic respiration remained lower. The most likely interpretation for such an effect is that the large albedo-driven decrease in soil temperature that was observed in the period before complete crop cover (up to -5.3°C at 5 cm depth on DOY 131) may cause changes in the soil microbial functions, leading to reduced organic matter decomposition rates (Muhr *et al* 2009). On the other hand, soil cooling also caused decreased harvest that did not affect overall C-input. This was not due to changes in phenology, which was scarcely affected by soil cooling, but rather to a decrease in the number of germinating seeds that led to a decrease in plant density and, possibly, a reduction in soil nutrient availability in response to lower SOM mineralization rates. The observation that cold spells may lead to reduced crop yields is not novel (Fuller *et al* 2007) but the effect of extreme cold events in the late winter-early spring on carbon accumulation in soils poses some interesting considerations. Recent studies on alternative tillage practices in crop management (Licht *et al* 2005, Al-Kaisi *et al* 2005) have already reported a decrease in soil temperature and an increase in carbon accumulation driven by no-tillage, even though they did not relate the increase in carbon storage to the decrease in soil temperature. Such an association is especially important in the light of recent climate modelling studies that support the idea of an increased likelihood of cold in Europe as a direct consequence of ice cover reduction in the arctic (Fereday *et al* 2012). The preliminary nature of our results and the large uncertainty associated with such climate predictions (based on arctic ice melting effects) prevents excessive generalization of the idea of a complex feedback mechanism by which global warming will eventually cause ice cover reduction, translating into a higher frequency of cold winters over Europe and thus eventually leading to a net carbon sequestration in agricultural soils. But this idea certainly calls for new and more extensive field

studies that can actually address the mechanisms associated with a decrease in soil temperature and the conservation of soil carbon stocks in croplands.

A final consideration examines the likely consequences of an increase in the amplitude and frequency of warming and cooling extreme events on the carbon balance of crop systems. Our study highlights that late winter-early spring heat waves are unlikely to cause depletion of soil carbon as resource supply rather than reaction kinetics appears to be the key limiting factor for heterotrophic respiration; on the other hand, substantial soil cooling occurring at the same stage in crop development may cause net carbon accumulation in soils. When combined, an increase in the frequency of both types of extreme events (Meehl and Tebaldi 2004 and Fereday *et al* 2012) is therefore unlikely to have large effects on the soil carbon balance of European irrigated croplands. A conclusion that certainly warrants further investigation, involves the use of validated simulation models capable of capturing short-term soil warming and cooling effects on the dynamics of soil organic matter, soil carbon fluxes and stocks, and their critical determinants.

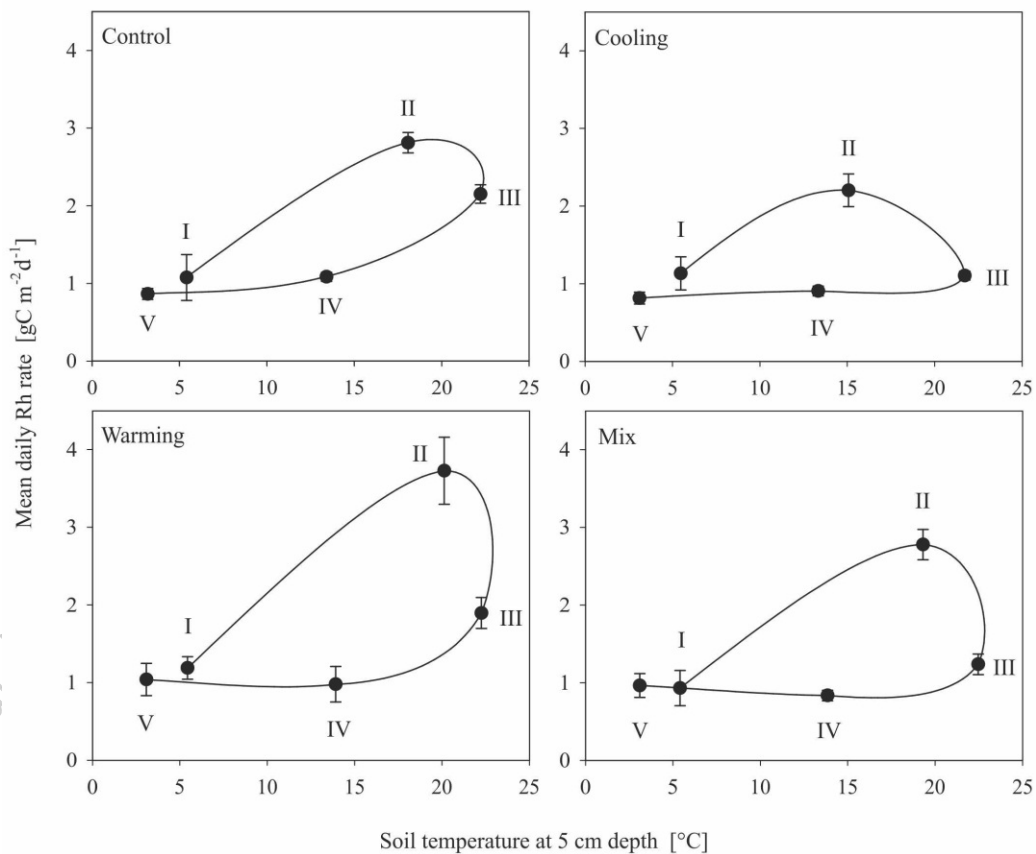


Figure 5.4: Heterotrophic respiration rates vs. soil temperature at 5 cm by treatment and period (I to V). Vertical and horizontal bars represent the standard error ($n = 3$).

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Overall conclusions

In this thesis, the effects of management and climate extremes on the C dynamics in forests and croplands have been investigated.

Considering forests and management, our study based on the chronosequence approach revealed that the studied management (i.e. shelterwood system) had no influence on the soil C stock in beech and spruce forests of Italian Alps, probably due to the low human disturbances on soil micro-climate. In fact, model simulations for both forests confirmed that the applied management allows for a rapid recovery of the ecosystem, as NEP was found to decrease to negative values only in consequence of the seed cut and for a few years. For these forests, it would be interesting to investigate the C fluxes through continuous soil respiration and/or eddy covariance measurements in the period immediately after the seed cut, in order to better understand the impacts related to harvest operations and to quantify the effects on the forest C budget.

Concerning croplands and management, we found that conservation agriculture does not necessarily lead to a benefit in terms of C budget. In fact, the results of our experiment showed that the management of a cropland through minimum tillage + cover crops resulted in a strong decrease of the overall C budget (NECB), even though this type of management led to a substantial improvement of NEP in comparison to conventional tillage. In particular, the tested management was found to change cropland C fluxes, especially during the tilled period, reducing both R_h and R_s , increasing GPP and thus reducing the NEE. However, this management has considerably increased the amount of harvested C during the first three years of the rotation when alfalfa was present thus negatively influencing NECB in comparison to conventional practices. In this field of research, it would be interesting to test also for other types of management and for different crop rotations. Moreover, it would be useful to integrate this comparison between managements with continuous ecosystem measurements of N_2O and CH_4 as cheaper analyzers for eddy covariance measurements are now available on the market and taking into account also the C emissions related to agricultural operations in order to get to a full GHG accounting, and thus truly assess the climate change mitigation potential of conservation agriculture.

Regarding croplands and climate extremes, both warm and cold extreme events during late winter had not a significant impact on the C budget of croplands. In particular, warm extremes caused only a transient increase in R_h that led to a faster consumption of the labile SOC, while cold

extremes showed only slightly diminished R_h rates. However, both kind of events had no effects on the annual R_h from the ecosystem. On the contrary, our treatments showed to influence phenology, particularly leading to an anticipated germination in consequence of soil warming and to a delayed germination due to soil cooling. Moreover, cooling led also to a delay in crop development and to a significant reduction in final harvest caused by a lower plant density. Overall C budget at the end of the year was not affected by both warm and cold extremes. However, we cannot exclude that in a longer time span, an higher frequency of such events could alter the decomposability of the non-labile SOC, thus likely leading to a positive feedback on climate change. Therefore, it would be interesting to study the long-term impacts of temperature extremes on croplands, particularly referring to the effects of these events on the “old” SOC, probably combining C isotopes and micro-meteorological measurements.

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