

Genetic resources of energy crops: Biological systems to combat climate change

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Abstract

Bioenergy crop plants that function as solar energy collectors and thermo-chemical energy storage systems are the basis for biological systems that are expected to contribute to renewable energy production, help stabilize the rising levels of green house gases (GHG), and mitigate the risk of global climate change (GCC). Wide genetic resource bases, especially of wild and semi-domesticated perennial grasses and woody species of starch-, oil, and lignocellulose-producing plants, are available to select, breed, genetically-modify, and develop environmentally-friendly bioenergy crops. Plant species, with fast growth, tolerance to biotic and abiotic stresses, and low requirements for biological, chemical or physical pretreatments, are being evaluated as potential bioenergy crops. Currently, bioenergy systems based on traditional sources and first generation bioenergy crops, are not sustainable and their exploitation may contribute to environmental degradation. New genetic resources and technological breakthroughs are being employed to develop dedicated bioenergy crops (DECs) with better GHG profiles and with a suite of eco-physiological traits to maximize radiation interception, water- (WUE) and nutrient-use efficiencies (NUE), improved lignocellulosic accessibility to enzymatic degradation, and to confer environmental sustainability. Large-scale bioenergy crop plantations pose both opportunities and challenges, and will inevitably compete with food crops for land, water, nutrient resources and other inputs; whereas, biodiversity consequences of increased biofuel production will most likely result in habitat loss, increased and enhanced dispersion of invasive species, and pollution. Recent genetic modifications and breeding efforts of bioenergy crops aim at improving biomass yield, quality, and conversion efficiency. Improvements in composition and structure of bio-chemicals in bioenergy crops will enable the production of more energy per ton of biomass and will improve its caloric value, GHG profile, and GCC mitigation potential.

Introduction

There is a strong public interest in stabilizing the atmospheric abundance of CO₂ and other GHGs to mitigate the risk of GCC which places new and more challenging demands on agricultural productivity, land and water resources, biodiversity, environmental health, and ecosystem services (IPCC, 2007; Fraiture et al., 2008). Biomass has the potential to become one of the major global primary energy sources during the 21st century, and the future demand for biofuels is one component of the expanding human demand for photosynthetically-fixed carbon (Nass et al., 2007; Hoogwijk et al., 2009). Modernized bioenergy systems will be important contributors to future energy systems; whereas, biomass derived from bioenergy crops will play an important role in combating GCC and will increase the share of renewable energy sources worldwide (Karp and Shield, 2008). However, using biological systems to store carbon and reduce GHG emissions is a potential mitigation approach for which equity considerations are complex and contentious (IPCC, 2007; Lal, 2007). Other biology-based mitigation approaches include the development and use of biofuels as energy carriers that store energy derived from biomass (Kotchoni and Gachomo, 2008). Nevertheless, positive impacts on ecosystem services will be more important when dedicated energy crops are deployed on a large scale in the landscape (Landis et al., 2008; Muller, 2009). Biomass is a heterogeneous aggregation of different feedstocks, conversion technologies, and end-use with different traditional and connotations in different parts of the world. Traditional

biomass provides 38±10 EJ/yr as fuel wood, manure, and other forms (Smeets et al., 2007). Estimates of the bioenergy production potential vary from 33 to 1135 EJ/yr due to the uncertainty of land availability and yield of bioenergy crops (Hoogwijk et al., 2009). The use of bioenergy crops to reduce the negative effects and exploit possible positive effects of GCC is set to increase in the developing as well as the developed world. Theoretical biomass resources are potentially the world's largest sustainable bioenergy source comprising about 220 billion oven-dry tons or 4,500 EJ of annual primary production (if marine phytoplankton resource is included). Of these, in 2050, there may be 273-1381 EJ/yr provided by bioenergy crops (Smeets et al., 2007). The first generation bioenergy crops (FGECS) from which biomass is currently derived has not been domesticated for this purpose and the present methods for saccharification and fermentation are inefficient and expensive. It is expected that existing genetic diversity in the plant kingdom will provide important basic material for the development of bioenergy crops and for adapting crop species to GCC (Karp and Shield, 2008). There is a greater variety of highly productive bioenergy crops that can be grown in tropical developing countries compared to those that can be grown in temperate, developed countries; however, different bioenergy crops will be optimal for different climates. Nevertheless, there is uncertainty regarding sustainability of biofuel production in the face of GCC (Nass et al., 2007; Muller, 2009). In order for bioenergy crops to be grown within the context of a sustainable agro-

ecosystem, in which a variety of ecosystem services might be produced in addition to energy and food (Lobell et al., 2008; Tilman et al., 2009), the impact of biofuels on food prices remains the subject of considerable debate, as does their potential to contribute to energy security, GCC mitigation through GHG emissions, and agricultural development (Karp and Shield, 2008; Landis et al., 2008). The amount of biofuel that can be produced globally in an environmentally responsible way is limited, and land needs provide one of the major constraints (Nass et al., 2007; Kotchoni and Gachomo, 2008). The grand challenge for biomass production is to develop crops with a suit of desirable physical and chemical traits while increasing biomass production by a factor of 2 or more (Lal, 2007; Lal, 2008a). Conventional grain and oilseed crops and crop residues, perennial herbaceous and woody crops, perennial oilseed crops, halophytes, and algae, among others, are candidate bioenergy crops and are expected to combat GCC (Ferre et al., 2005; Eisenbies et al., 2009).

Bioenergy crops to combat climate change

Traditional bioenergy crops

Biomass has always been a major source of energy for mankind and presently contributes 10-14% of the world's energy supply. Traditional biofuels derived from natural vegetation or from crop residues are not new, have not always been good for health or for the environment and have competed with food production in developing countries where 70-75% of the energy used is in the form of biomass and almost 90% of it is used for food preparation (Kotchoni and Gachomo, 2008; Lobell et al., 2008). Throughout the developing world, firewood is still being gathered as a biofuel, and trees are likely to be damaged by exploitative, unregulated harvesting practices, resulting in wide ranging detrimental environmental and livelihood impacts (Eisenbies et al., 2009). Traditional biofuels still are the main energy source in a number of countries (e.g., Bhutan 86%, Nepal 97%); however, they are not sustainable; their exploitation may contribute to land degradation and desertification (Karp and Shield, 2008). Exploiting the indigenous plants as feedstocks for biofuels would need to include domestication programs to select for specific properties such as oil yields, quality, and content, as well as the ability to produce under managed systems so as to minimize the damage and exploitation of natural systems and to mitigate the impact of GCC (Chhetri et al., 2008). Agro-forestry as a traditional land-use adaptation may potentially support livelihood improvement through simultaneous production of food, fodder and firewood as well as mitigation of the impact of GCC. Innovations in domestication of useful species may strengthen the role of agro-forestry in developing countries (Singh, 2008).

First generation bioenergy crops (FGECs)

The vast majority of current liquid biofuels production is based on FGECs that can also be used for food; therefore, their raw materials compete with food for fertile land and inputs. Currently, a small number of food-crop species such as corn, sugarcane, oil palm and rapeseed are used globally to produce biofuels (Lobell et al., 2008). However, with the long-term goal of producing 1 Pg of lignocellulosic biomass in the US and 4-5 Pg in the world, crop residues are increasingly considered as sources of biomass. Potential availability of FGECs is limited by soil fertility and per hectare yields, and the effective savings of CO₂ emissions and fossil energy consumption are limited by the high energy input

required for crop cultivation and conversion (Blanco-Canqui and Lal, 2009). Biofuels derived from FGECs rely on fermentation of sugars to produce ethanol or on trans-esterification of plant oils to produce biodiesel. It is generally well understood that FGECs are limited in their ability to achieve targets for oil-product substitution, GCC mitigation, and economic growth (Chhetri et al., 2008; Carroll and Somerville, 2009; Lorenz et al., 2009). For most crops the annual change in above ground C is equal to zero if the whole biomass is taken away for energy production. The cost and sustainability of FGECs, other than sugarcane (Wang et al., 2008), have been criticized as expensive sources to meet environmental goals, and to provide energy alternative. These limitations can be partly overcome by the utilization of lignocellulosic materials from their residues (Eisenbies et al., 2009).

Second generation bioenergy crops (SGECs)

The SGECs are expected to be more efficient than FGECs and to provide fuel made from cellulose and non-oxygenated, pure hydrocarbon fuels such as biomass-to-liquid (BtL) fuel (Oliver et al., 2009). Biofuels produced biochemically or thermo-chemically from lingo-cellulosic SGECs, have more energy content (GJ/ha/yr) than most FGECs biofuels, could avoid many of the environmental concerns, and may offer greater cost reduction potential in the longer term (Petersen, 2008; Wang and Yan, 2008). However, technical barriers remain for growing and fuel production from SGECs. As with FGECs, the environmental consequences of the SGECs depend largely on the type of feedstock and how and where it is produced. The net GHG emissions from using either cellulosic ethanol or BtL are substantially less than for ethanol from grain-producing FGECs such as corn (Carpita and McCann, 2008; Carroll and Somerville, 2009). Early SGECs include perennial forage crops such as *Panicum virgatum* L., *Phalaris arundinacea* L., *Medicago sativa* L., *Pennisetum purpureum* Schumacher, and *Cynodon* spp. (Sanderson and Adler, 2008; Oliver et al., 2009). These are some of the most extensively studied species for cellulosic feedstock production. Historically, these have been used for grazing and forage and were the original energy feedstocks for draft animal power. Switchgrass (*Panicum virgatum* L.), a C₄ native warm-season perennial grass, demonstrated high productivity across many environments, is suitable for marginal and erosive lands, needs low water and nutrient requirements, and has positive environmental benefits (McLaughlin et al., 2006; Vogel and Mitchell, 2008). New cultivars with improved biomass yield and chemical composition have been released in the US (Sticklen, 2006; Boe and Lee, 2007; Boe and Beck, 2008); however, the plant received very little breeding although its genetic resources harbor tremendous genetic variability and great potential for germplasm improvement. Miscanthus (*Miscanthus x giganteus*), a cool hardy, vegetatively-propagated C₄ grass native to Asia, requires low amounts of and efficiently cycles N fertilizer. The capacity of *Miscanthus* to fix CO₂ ranges from 5.2 to 7.2 tC/ha/yr, which results in a negative C balance (Boe and Lee, 2007; Boe and Beck, 2008; Jakob et al., 2009; Leaky, 2009). Low-input, high-density mixtures of perennial grasses grown on degraded lands were advocated (Tilman et al., 2006) as better bioenergy sources than single species and may provide similar bioenergy gains and greater GHG benefits than current corn ethanol produced from crops grown in monoculture on fertile soil with high inputs. The use of indigenous perennial grass species is particularly promising, both because these are likely to be well adapted to local environment, and because they are less likely to

adversely affect biodiversity than are non-native species, which are frequently invasive. On average, increasing species richness in perennial herbaceous polycultures increased productivity and weed suppression, but well-adapted species produced high biomass yield regardless of richness (Fransworth and Meyerson, 2003; Picasso et al., 2008). The contribution of non-edible plant oils (e.g., from *Jatropha curcas* L., Euphorbiaceae; 30-50% oil) and soapnut (*Sapindus mukorossi* and *S. trifoliatu*s; 52% oil), as new sources for biodiesel production have the advantage of not competing with edible oils produced from crop plants (Ram et al., 2008; Ranade et al., 2008). Other oil crops include *Azadirachta indica*, *Calophyllum inophyllum*, *Pongamia pinnata*, among 75 oil plants contain >30% or more oil in their seed, fruit or nut (Chhetri et al., 2008; Komar et al., 2009). Major plant families with oil-producing plants include Amariyllidaceae, Apocynaceae, Asclepiadaceae, Compositae, Convolvulaceae, Cruciferae, Euphorbiaceae, Flacourtiaceae, Lauraceae, Leguminosae, Malvaceae, Moraceae, Myrcaceae, and Palmae. The need is urgent to assess the impact of growing SGECs and producing biomass for bioenergy on the environment and how they may contribute to GHG mitigation (Kotchoni and Gachomo, 2008).

Third generation bioenergy crops (TGECS)

The TGECS include boreal plants, crassulacean acid metabolism (CAM) plants, *Eucalyptus* spp. and micro-algae (Patil et al., 2008; Schenk et al., 2008); the boreal and CAM plants are potential sources of feedstocks for direct cellulose fermentation (Carere et al., 2008; Borland et al., 2009), and *Eucalyptus* for bioenergy production through thermo-conversion (Carere et al., 2008; Wang and Yan, 2008); whereas, algae is a potential source of biodiesel. Successful development of TGECS depends heavily on a detailed understanding of the metabolism of cellulolytic bacteria, organisms that are capable of degrading cellulose and utilizing it as a source of C. Cellulose is generally degraded into H₂O and CO₂ in aerobic systems, while in anaerobic systems; CH₄ and H₂ are also produced. Success, however, will be dependent upon design decisions based on a detailed understanding of the extremely complex genetic, enzymatic, and thermodynamic mechanisms that direct C flow. In combination with other strategies including (meta)genomics, biodiversity studies, and system biology, metabolic engineering is a promising approach to the improvement of biofuel yields and the establishment of renewable, non-polluting energy source from TGECS that can mitigate GCC (Bush, 2007; Ehrlich and Pringle, 2008; Rubin, 2008). There is a large reservoir of boreal plant species that can be harnessed in CH₄ production. These plants are easy to cultivate, harvest and store, are tolerant to weeds, pests, diseases, drought and frost, and have good winter hardness and able to grow on poor soils with low nutrient inputs (Finckh, 2008). Boreal plants include perennial grasses (e.g., *Phleum pratense* [8-11 t dry matter/ha; or 2900-4000 m³ CH₄/ha], and *Phalaris arundinacea* [9-10; 3800-4200]) are among the most efficient producers of herbaceous biomass under boreal conditions (Lehtomaki et al., 2008). Boreal plants, such as pineapple (*Ananas comosus*), *Opuntia ficus-indica*, *Agave sisalana* and *Agave tequilana* are already being used to produce bioenergy with sizable GCC mitigation potential (Lehtomaki et al., 2008). *Opuntia* spp. are part of natural and agronomic ecosystems in many parts of the world, with commercial cultivation (for forage and fodder) with large (47-50 Mg/ha/yr) productivity. Agave as an economically viable sources of ethanol with zero-waste platform in Mexico produces 50 Mg/ha/yr with 27-38% sugar; and distilled

ethanol yields of 14,000 l/ha, and additional 33,500 l/ha from cellulose digestion (Lehtomaki et al., 2008). Plants with the crassulacean acid metabolism (CAM), as a photosynthetic adaptation that facilitates the uptake of CO₂ at night and thereby optimizes water use efficiency (WUE) of C assimilation in arid habitats, are potential drought-tolerant bioenergy crops (Fraiture et al., 2008). The WUE of CAM plants (CO₂ fixed per unit H₂O lost) can be 3- and 6-fold higher than that of C₄ and C₃ plants, respectively. Some CAM plants (e.g., Cardoon; *Cynara cardunculus* L.) serve as multi-functional bioenergy crops that can produce solid and liquid biofuels. On average, the heating value of the dry biomass with and without the seed is 18.5 and 16.5 GJ/t, respectively, and an energy ratio (input/output) of up to 1:27 was attained. The seed (15-20% of biomass) is 25% oil that can be converted into biodiesel; whereas, the biomass can be converted into ethanol (Borland et al., 2009; Grammellis et al., 2008). Of the >700 *Eucalyptus* spp. that are native to Australia, a large genetic resource exists to select and develop biomass species with fast growth, tolerance to harsh environments, indeterminate growth, coppicing, lignotubers, drought, fire, insect resistance, and tolerance to soil acidity and low fertility. *Eucalyptus* plantations in tropical countries are increasing due to fast growth, with rotations as short as 5 years and yields as high as 70 m³/ha/yr. Four species and their hybrids (*E. grandis*, *E. urophylla*, *E. camaldulensis*, and *E. globulus*) account for about 80% of plantations worldwide. *E. globulus* is widely adapted, used in breeding for fast growth, and comprises most plantations in Australia, Brazil, where Eucalypt oil is being produced in addition to biomass for biofuel and bioenergy production. Lignin content at about 34% is higher than most hardwood species, which suggests that short rotation plantations of *Eucalyptus* species can be ideal for bioenergy production through thermo-conversion (Rockwood et al., 2008; Wang and Yan, 2008). A number of TGECS oleaginous crops being tested (Carere et al., 2008) for biodiesel production include the seed of African palm (*Elaeis guineensis*) and babassu (*Attalea speciosa*) with 22 and 66% oil, respectively; fruit of avocado (*Persea americana* (7-35% oil), and coconut (*Cocos nucifera*, 55-60% oil); and grain of castor bean (*Ricinus communis* 45-48% oil), and peanut (*Arachis hypogaea*, 40-43% oil). Other species under study as sources for biodiesel production include *Acrocomia aculeate*, *Astrocaryum murumuru*, *Attalea humilllis*, *Jatropha curcas*, *Licania rigida*, *Oenocarpus bataua*, *Oenocarpus bacaba*, and *Theobroma grandiflorum* (Ram et al., 2008; Shao and Chu, 2008; Komar et al., 2009). High lipid algae is an efficient and promising genetic resource for future production of biodiesel and other biofuels with favorable environmental benefits and potential positive GCC mitigation impact (Patil et al., 2008). Depending on the species, algae contain 20-40% lipids by weight, can produce a wide range of feedstocks for the production of biodiesel, bioethanol, biomethane, and biohydrogen; however, algae cultivation can only occur under specific light, temperature and density conditions. Microalgae are veritable miniature biochemical factories, and appear more photosynthetically efficient than terrestrial plants, and are efficient CO₂ fixers. They can help reduce GHG emissions by capturing CO₂ released from power plants or by generating biomass through photosynthesis (Schenk et al., 2008). They can produce oil equivalent to 100 times of soybean per unit area. As compared with rapeseed (1,190 L of biodiesel/ha/year), *Jatropha curcas* (1,890 L of biodiesel/ha/year) and oil palm (5,950 L of biodiesel/ha/year), algae at 10 g/m²/day and 30% triglycerides can produce 12,000 L of biodiesel/ha/year, and at 50 g/m²/day and 50% triglycerides can produce 98,500 L of biodiesel/ha/year (Komar et al., 2009). Microalgal systems

have a higher photon conversion efficiency, can be harvested batch-wise nearly all-year-round, can utilize brackish and saline water resources (Williams et al., 2008), can couple CO₂-neutral fuel production with CO₂ sequestration, and produce non-toxic and highly biodegradable biofuels. The optimization of strain-specific cultivation conditions is of confronting complexity with many interrelated factors that can each be limiting. Microalgae can be improved for biofuel production through a series of processes, including screening a wide range of natural isolates, genetic engineering, selection and adaptation. Microalgal biofuels are likely to have much lower impacts on the environment and the world's food supply than conventional biofuel-producing crops (Patil et al., 2008; Schnek et al., 2008; Tilman et al., 2009).

Halophytes

Salinity is one of the agricultural problems that result from, or are aggravated by, GCC. In view of the competitive nature of glycophyte (non-salt tolerant plants) biofuel sources for land and water resources, salt-tolerant plants, especially halophytes, provide alternative solid, liquid and gaseous biofuel sources that can thrive on brackish and saltwater or in saline soils (Jaradat, 2003; Fraiture et al., 2008). Halophytes are common feedstocks for fuel and food and feed in developing countries; they provide many ecosystem services, including C sequestration and GCC mitigation, rehabilitation of degraded land, stabilizing ecosystems by providing niches and protection for other flora and fauna. Halophytes can complete their normal annual life cycle under conditions of over 15 dS/m root-zone salinity; they are not a single taxonomic group, but represented by several thousand species of forbs, grasses, shrubs, and trees, most of which can be utilized or domesticated as bioenergy crops. Halophytes occupy important niches in many ecosystems due to the ease with which they adapt to diverse and harsh environments (O'Leary, 1993; Williams et al., 2008). Some of the species that are being used for fuel production in saline environments are found in the genera *Acacia*, *Eucalyptus*, *Casuarina*, *Melaleuca*, *Prosopis*, *Rhizophora*, and *Tamarix*. In addition, a number of frost-sensitive *Eucalyptus* spp. and frost-tolerant *Populus* spp. are among the best genetic resources for biomass production under saline conditions. For example, *Eucalyptus rudis* and *Acacia saligna* are candidates for domestication; they have several desirable attributes as bioenergy crops including high potential for rapid growth, easy establishment, and a wide genetic base (Aronson, 1989; Rockwood et al., 2008).

Halophytes can produce large biomass per unit area and unit time. Experimental *Salicornia* farms yield 17-20 Mg/ha/yr of biomass and about 2 Mg/ha/yr of combustible oil using seawater (~35 dS/m). Giant reed (*Arundo donax*), a perennial rhizomatous grass, tolerates ~18 dS/m salinity and produced 11,000 L of ethanol from 45 Mg/ha/yr of biomass using BtL technology. Very few halophytes have been identified so far as potential sources of liquid fuels besides *Arundo donax*; these include wild sugar beet (*Beta maritima*) and the nipa palm (*Nypa fruticans*). Similarly, a few halophytes, such as Kallar grass (*Leptochloa fusca*) are promising genetic resources for biogas production. The success and long-term sustainability of halophytes as bioenergy crops and for GCC mitigation will depend on continued efforts of selection and breeding (Lee, 1999). In addition, halophyte germplasm can furnish "climate-ready" genes for genetic engineering research and the development of new salt tolerant bioenergy crops (O'Leary, 1993; Jaradat, 2003).

Dedicated bioenergy crops

The development and deployment of dedicated energy crops have been proposed as a strategy to produce energy without impacting food security or the environment (Lobell et al., 2008; Jessup, 2009). The dedicated energy crops are mainly perennial herbaceous and woody plant species. Genetic resources for the development of dedicated energy crops with low requirements for biological, chemical or physical pretreatment are more environmentally friendly and will contribute more to GCC mitigation (Petersen, 2008; Taherzadeh and Karimi, 2008). Dedicated energy crops can have the added benefit of providing certain ecosystem services, including C sequestration, biodiversity enhancement, salinity mitigation, and enhancement of soil and water quality (Ehrlich and Pringle, 2008; Lal, 2008b). The value of these services will depend on the particular bioenergy system in question and the reference land use that it displaces (Dillon et al., 2007; Jessup, 2009). However, under certain GCC scenarios (Gillingham et al., 2008), dedicated energy crops will inevitably compete with food crops for land, water, nutrient resources and other inputs. There is a greater variety of highly productive dedicated energy crops that can be grown in tropical countries compared to those that can be grown in temperate developed countries. The private sector is prospectively defining criteria to choose plants with potential to serve as dedicated energy crops (Jessup, 2009). The criteria include cell wall composition, growth rate, suitability of growth in different eco-geographical regions, and resource-use efficiency. Some crops favored for investigation as dedicated energy crops include (1) cellulosic crops including short rotation trees and shrubs such as eucalyptus (*Eucalyptus* spp.), poplar (*Populus* spp.), willow (*Salix* spp.), and birch (*Betula* spp.); (2) perennial grasses such as giant reed (*Arundo donax*), reed canary grass (*Phalaris arundinacea*), switchgrass (*Panicum virgatum*), elephant grass (*Miscanthus x giganteus*), Johnson grass (*Sorghum halepense*) and sweet sorghum (*Sorghum bicolor*); and (3) non-edible oil crops such as castor bean (*Ricinus communis*), physic nut (*Jatropha curcas*), oil radish (*Raphanus sativus*), and pongamia (*Pongamia* spp.). Also, a number of woody oil plants (e.g., *Carcinia multiflora*, *Camellia oleifera*, *Cerasus humilis*, *Cornus wilsoniana*, *Elaeis quineensis*, *Euphorbia tirucalli*, *Jatropha curcas*, *Pistacia chinensis*, *Sapium sebiferum*, *Virnicia fordii* and *Xanthoceras sorbifolia*) considered as potential dedicated energy crops, have common characteristics, including short life cycle, long harvest period, several harvests per year, and short stature for easy harvest (Boe and Lee, 2007; Basha and Sujatha, 2007; Ranade et al., 2008; Shao and Chu, 2008; Komar et al., 2009). Short Rotation Coppice (SRC) are among the most promising dedicated energy crops for bioenergy production and GCC mitigation (Rae et al., 2009); they include *Salix*, *Populus*, *Robinia* and *Eucalyptus* species planted in rows to facilitate harvest and harvested in 3-5 year rotation. SRC products are combusted for heat or electricity generation, and can be processed to produce ethanol. SRC willow or poplar can be productive for 25-30 years and produce 7-12 oven dry t/ha/yr (Rowe et al. 2009). At identical biomass volumes, trees in SRC plantations with high wood density (mainly deciduous species) accumulate and sequester more C than trees with light wood density (mainly coniferous species). SRC plantations may result in more biomass and have larger potential for GCC mitigation than herbaceous perennial dedicated energy crops; however, they are more disturbing for biodiversity. Valuable but non-native broadleaved species (e.g., *Acer pseudoplatanus* L., *Castanea sativa* Mill., *Fagus orientalis* Lipsky.) may become more important in C

sequestration and GCC mitigation as short rotation woody crops (Ehrlich and Pringle, 2008). Hybrid DEC are feasible in the mid- to long-term and will undoubtedly enhance biomass and GCC mitigation potential. Criteria for the development of novel hybrid dedicated energy crops include (1) large-seeded crops with vigorous establishment to simplify biofuel production systems; (2) delayed flowering through photoperiodism to enhance greater biomass accumulation and potentially prevent seed-borne weed risks; and (3) sterility, based on cytoplasmic-, genetic-, or wide-hybridization, to enable larger bioenergy production, and reduced invasiveness potential (Fransworth and Meyerson, 2003).

Characteristics of bioenergy crops

There will be always a need to deploy new high-yielding dedicated energy crops that can be grown as crops in cropping systems with significantly improved phenotypic, architectural, physiological and biochemical characteristics in order to sustainably produce bioenergy and help combat GCC. Under such conditions, cultivars cannot be substituted regularly; therefore, perenniality would confer advantages in reducing energy cost of crop-establishment, and higher yields will generally lead to lower fossil energy input, water and nutrient use per unit biomass yield, and to better environmental protection (Boehmel et al., 2008). Due to the perennial nature of most SGEs, field resistance against diseases and pests should be multigenic (Finckh, 2008).

Agronomic and architectural traits

A bioenergy crop with low inputs for establishment, low fossil fuel inputs, adaptation to marginal lands, and high biomass and energy yield is expected to help reduce global warming and combat GCC. Agronomic characteristics of an ideal bioenergy crop include, but are not limited to, low proportional allocation of dry matter to reproductive structures, long canopy duration, perennial growth, sterility to prevent escape, and low moisture content at harvest. Most of these traits are found, for example, in the C_4 perennial grass *Miscanthus* and SRC, but not in many FGECs (Lewandowski et al., 2000; Jakob et al., 2009; Leaky, 2009). The architecture of a dedicated energy crop plant should help minimize plant-to-plant competition and effectively maximize competition with weeds, maximize radiation interception and WUE, accelerate drying in the field, and facilitate mechanical harvesting. This can be achieved by adjusting branching habit and having a thick, straight, upright stem and resistance to lodging. Trees (e.g., SRC) can be optimized to have short stature to increase light access and enable dense growth, large stem diameter, and reduced branching to optimize energy density for transport and processing.

Physiological and eco-physiological traits

A bioenergy crop plant can be viewed as a solar energy collector and thermo-chemical energy storage system, the yield of which, defined as the amount of C/ha/yr, is a function of the number of cells per unit area multiplied by the amount of C per cell. Therefore, biomass and bioenergy yields can be enhanced by increasing the number of cells/ha/yr, the amount of accumulated C per cell or both (Rae et al., 2004). Numerous physiological and eco-physiological traits needed to maximize radiation interception, radiation, water and nutrient-use efficiencies, and to confer environmental sustainability, should be targeted to enhance plant biomass and bioenergy production. For example, *Miscanthus x giganteus* over 3 years achieved an average

annual conversion efficiency into harvestable biomass of 1% (30 t/ha) and a maximum of 2% (61 t/ha), with minimal inputs; whereas, switchgrass achieved ~35% of that yield due mainly to differences in light intercept efficiency between these bioenergy crops (Boe and Lee, 2007; McLaughlin et al., 2006). (Eco)physiological traits that can help change thermal time sensitivity to extend the growing season and increase aboveground biomass without depleting belowground biomass include: high growth rate, response to light competition, canopies with low extinction coefficients, leaf traits for efficient light capture (including optimum LAI, and high SLA), C_4 or CAM photosynthetic pathway coupled with large WUE, long canopy duration, large capacity for C sequestration and nutrient cycling, and low nutrient (e.g., N and S) requirements and content of above-ground biomass (Lal, 2008b; Jakob et al., 2008). In addition, the large diversity available in germplasm of perennial SRC and lignocellulosic grasses for eco-physiological traits such as leaf area (LA), leaf area index (LAI), and specific leaf area (SLA), branching habit, and biomass partitioning patterns has been shown to influence clonal biomass production potential (Tharakan et al., 2001; Carroll and Somerville, 2009), will help develop improved bioenergy crops. Bioenergy crops with vegetative storage organs (e.g., stems in the C_4 sugarcane and roots in the C_3 sugar beet) are able to accept assimilates for storage over longer periods than grain crops. Vegetative storage reduces feed-back restriction to yield accumulation during environmental stress; and sucrose, as the storage product of PS, is the least transformed and therefore subject to smallest losses by subsequent metabolism (Wang et al., 2008). Biomass for bioenergy is harvested for its C not N content; therefore a high C:N ratio is preferable for maximum bioenergy production via combustion; however, a C:N ratio within the range of 25 to 32 is optimal for anaerobic digestion and is likely to contribute to high CH_4 yields of bioenergy crops (Long et al., 2006).

Biochemical composition and caloric content

The caloric value of a material is an expression of the energy content, or heat value, released when burned in air. Plants differ in their biochemical composition (i.e., carbohydrates, proteins, lipids, organic acids, etc.) and in the amount of glucose to produce a unit of these organic compounds; therefore, plant composition determines the availability of energy from specific biomass type, when adjusted for moisture content, and results in differences in energy output. Besides their effect on energy yield, biomass yield and composition affect GHG profiles and GCC mitigation potential of bioenergy crops. For example, hybrid poplar produces the largest energy yield (6.15 MJ/m²/yr), followed by switchgrass (5.8) then reed canary grass (4.9); however, reed canary grass has the largest net GHG emission ratio of 3.65, as compared with switchgrass (2.42) and hybrid poplar (2.37) (Ferre et al., 2005; Boe and Beck, 2008). Energetic and environmental values of a particular type of biomass depend on the chemical and physical properties of the large molecules from which it is made; the gross energy content of biofuel produced per hectare determines its value in reducing global warming and in combating GCC. However, energy retained by plants is not proportional to accumulated biomass because proportions of the major chemical forms differ in energy density as well as between species and stages of growth. These are important issues in the energetics of plant growth and suitability of crops not only for bioenergy, but also for food and feed production (Lobell et al., 2008). Bioenergy crops that need less costly biological, physical, chemical or a combination of these pre-treatments is a better

source for energy and as bioenergy crops to mitigate GHG emissions. However, pre-treatment to remove ~65% of lignin from soft wood pulp and corn stover resulted in a nearly threefold increase in the yield of reducing sugars, 88% increase in the yield of glucose and a twofold increase in the initial hydrolysis rate (Monti et al., 2008; GEMIS, 2009). There are large differences between crops as a result of differences in feedstock characteristics and production environments. For example, oil palm yield ranged from 17.8 to 206 t/ha under different environments; whereas, its energy yield ranged from 134.6 to 155.8 GJ/ha. Similarly, within-species differences were reported under different environments for sugarcane yield (60.7-73.5 t/ha) and energy (95.4-115.5 GJ/ha), rapeseed yield (1.5-1.7 t/ha) and energy (21.1-23.9 GJ/ha), and soybean yield (2.4-2.7 t/ha) and energy (16.1-18.2 GJ/ha). The net energy value (NEV) is mainly affected by the productivity of the bioenergy crop; for example it ranged from -2.89 to 4.88 MJ/l of ethanol produced from corn grown under different dry land climatic and soil conditions, and from 3.68 to 6.85 MJ/l under irrigation (Persson et al., 2009; Schmer et al., 2008; Wang et al., 2008). The energy content of biomass (on a dry, ash-free basis) is relatively similar for most plant species (17-21 MJ/kg). However, energy contents per unit dry mass (MJ/kg) of the major chemical forms are different and range from 14 to 16 for sugars, starch, cellulose and hemicelluloses, ~ 17 for vegetative biomass, 25 for proteins and lignin, and from 38 to 40 MJ/kg for lipids. High levels of low molecular weight carbohydrates content (in un-polymerized state) is a valuable trait in bioenergy crops used for fermentation to help reduce energy inputs in the refining process and optimize GHG profiles. Quantitative and qualitative differences in carbohydrate content, for example, in sugar beet (94%), wheat and corn grain (70-76%), softwood (67%) and hardwood (66%) explain, to some extent, differences between these bioenergy crops in energy output as ethanol (Carroll and Somerville, 2009; Lorenz et al., 2009). Primary net energy yields (PNEY) of a number of SGECS (with no N application), including bioenergy corn (294 GJ/ha/year), willow (257), *Miscanthus* (224), and switchgrass (140), are variable and large as compared to those of FGECS such as grain and straw of winter oilseed rape, winter wheat, and winter triticale (~118); however, when adequate N was added for each crop, an increase in PNEY values of 2-100% was observed. Similarly, net energy ratios (output/input; NER) are equally variable among crops that produce ethanol or biodiesel and also differ between and within bioenergy crops. Most estimates for sugar beet (1.2-2.2), wheat (1.2-4.2), corn (1.2-1.8), soybean (1.4-3.4), and rapeseed (1.2-3.6) are small in comparison with sugarcane (2.2-8.4) and oil palm (8.6-9.6) (GEMIS, 2009). Cellulosic SGECS had higher biofuel yield and lower GHG emissions per hectare, and had a greater reduction in GHG emissions per unit biofuel produced than FGECS, resulting in greater reductions in GHG emissions associated with fossil fuels and better GHG profiles. This can be illustrated by the CO₂ profile (% of CO₂ release for the corresponding fossil fuel) of ethanol from FGECS, such as corn (90%), wheat (60-105%), sugar beet (30-70% in the EU and 15% in Brazil); and biodiesel produced from rapeseed (40-80%), and soybean (25-60%) as compared with ethanol produced from cellulosic feedstocks of SGECS (12-25%), or BtL biodiesel (15%). To illustrate the abatement potential of GHG emissions, it would be possible to abate about 3 billion tons of CO₂ emissions/yr if 38 EJ/yr of electricity and 51.5 EJ/yr of ethanol were produced from sugarcane (Monti et al., 2008; Carroll and Somerville, 2009). In addition to their role in combating GCC, terrestrial C sequestration of bioenergy crops offers multiple environmental benefits and ecosystem

services (Lal, 2008b; Landis et al., 2008). Forest ecosystems, for example, store C as lignin and other relatively resistant polymeric compounds (Singh, 2008); at present it is around 1.7 Pg C/yr. Interactions between cycles of N, P, H₂O, if moderated through judicious management, may enhance terrestrial C sequestration with positive impact on global warming and GCC mitigation. Improvements in composition and structure of biochemicals in bioenergy crops will enable the production of more energy per ton of biomass and will improve its caloric value, GHG profile, and GCC mitigation potential (Sticklen, 2006).

Genetic improvement of bioenergy crops

As we can retrospectively view the suite of traits that made certain wild plants desirable for domestication thousands of years ago to become today's food and feed crops, we are now prospectively defining criteria to choose wild plants as potential dedicated energy crops. Classical breeding and genetic modification techniques are already available to develop crops with desired morphological, phenological, and biochemical traits as dedicated energy crops (Lee, 1999; Baenziger et al., 2006). These include large energy yield, large C:N ratio, modified lignin biosynthesis, pre-processing *in planta* via expression of cellulases and cellulosomes, and cell wall lignocellulose characteristics that make the feedstock more amenable to processing by one or a combination of biological, physical, chemical pre-treatments. Several technologies, besides classical breeding, are already available to improve these traits, including genomic approaches to screen natural variation and the use of genetic modification to produce transgenic plants (Gressel, 2008; Ortez, 2008). Plant genetic resources have already been mined to improve lingocellulosic biomass accessibility to enzymatic degradation (Bouton, 2007; Carere et al., 2008; Rubin, 2008). A thorough understanding of how gene products function in the synthesis and architectural construction of the cell wall will help modify plants to engineer lignin and cellulose so that they breakdown more easily, speed-up plant growth and increase yield (Ortiz, 2008); whereas, classical breeding is expected to substantially contribute to the design of optimal bioenergy crop plants. Classical breeding was responsible, for example, for most of the nine-fold increases in grain yield since the advent of hybrid corn. Although yield improvement through breeding and potential increases through the application of biotechnology have been reported for willow, whereby dry weight yield reached 15 t/ha/yr; however, using classical breeding only or with novel gene discovery, yield is expected to reach 25 and 40 t/ha/yr by 2025, respectively (Baenziger et al., 2006). Genetic improvement is needed to develop bioenergy crops more adapted to adverse environmental conditions with higher growth rate and high caloric value. Many of the traits that need manipulating to improve yield are unlikely to be amenable to simple genetic modification and will require a combination of approaches including classical and novel gene discovery via QTL-assisted breeding (Murray et al., 2008a). The wealth of genomic resources and tools for food crops (e.g., corn, sorghum and rice) can be put to immediate use to make similar advances in biomass yield, quality and GHG profiles (Rubin, 2008). The high degree of genetic synteny among grass or *Poplar* spp. genomes should facilitate the translation of gene-function discovery in these species to more genetically recalcitrant grass species, such as switchgrass and *Miscanthus*, and SRC species, respectively (Ferre et al., 2005; Bouton, 2007).

Genetics of bioenergy crops

The open question for genetic research on bioenergy crops is will it allow plant yields to increase faster than the projected 1% per year? Genetic correlations between yield-related traits need to be investigated to identify “early diagnostic” indicators of biomass and bioenergy yields and energy density. Increased understanding of the genetic and physiological mechanisms that control yield-related traits (e.g., tiller density, number of phytomers per tiller, and mass per phytomer in perennial grasses, and leaf size, cell number, and stem and sylleptic branch number per plant in SRC species) would be useful to achieve large gains in yields of lignocellulosic bioenergy crops. Also, intensified search for genetic resources with wide genetic diversity for yield-related traits, climate-ready genes (e.g., from halophytes), and transgenic solutions to biotic and abiotic stresses, are expected to accelerate yield gains in bioenergy crops (Gressel, 2008; Ortez, 2008). The considerable genetic diversity within potential bioenergy crops (e.g., *Acacia saligna*, *Jatropha* spp and *Panicum virgatum* L.) suggest that a number of important traits (e.g., biomass yield, WUE), exhibit genetically-induced variation (Ranade et al., 2008; Komar et al., 2009). However, the high level of genetic variation in these and other species is likely to complicate agronomic studies designed to unravel the impact of genotype x environment interaction on biomass yield and energy-related traits (Basha and Sujatha, 2007). Such interaction is a characteristic of widely distributed species, which are also likely to have locally specific adaptation. Prolonged selection and breeding of bioenergy crops could result in producing highly differentiated cultivars with lower genetic diversity than their wild progenitor populations as was the case in food crops. Gene flow between newly domesticated bioenergy crops and wild populations could lead to the introduction of adaptive or maladaptive genes, disruption of co-adapted gene complexes, and genetic assimilation.

Breeding of bioenergy crops

Breeding of bioenergy crops implies breeding for adaptation to long-term GCC and the replacement of crops having high inter-annual yield variability with new ones having more stable yields, and may involve innovative plant design via accelerated domestication. It is unrealistic to assume that plantations of bioenergy crops can be started with little or no domestication; large deployment of wild species in the landscape as bioenergy crops is bound to lead to unforeseeable biological and environmental problems (Petersen, 2008). Biomass and bioenergy yields of lignocellulosic crops could increase significantly over time since breeding research, including genetic modification of bioenergy crops, is at an early phase compared with food crops. Breeding challenges of bioenergy crops include long-yield cycles, complex genetics, multiplication, and conducting expensive long-term experiments involving perennial species and their interaction with the environment. A basic breeding program for bioenergy crops entails collection and evaluation of genetic resources, genetic analyses and development of criteria for selection, development of novel tools for selection and testing novel varietal concepts, and genetic improvement for biomass yield and energy-related properties (Bouton, 2007; Dillon et al., 2007). Breeding objectives of dedicated energy crops include the improvement of biomass yield, quality, and conversion efficiency, either through selection among progeny obtained by crossing parents with desirable traits, or as a way to enhance the agronomic performance of

promising mutants and transgenic plants (Gressel, 2008; Grattapaglia et al., 2009). Tree (e.g., SRC) breeders, for example, must reduce the number of years required to complete a generation of testing and its deployment, improve understanding of the genetic control of desirable timber traits, and produce fast growing SRC cultivars (Rae et al., 2009). Traditional breeding has increased yield performance of perennial grasses (e.g., switchgrass by 20-30% from existing parental types) and several SRC species (Bouton, 2007; Vogel and Mitchell, 2008). Breeding bioenergy crops for improved NUE, especially under low N conditions, will help lower N₂O emissions. Further improvements both in genetics and agronomics, when achieved, will further improve biomass yield, conversion efficiency, and net energy yield of dedicated energy crops (Schmer et al., 2008). Self-incompatibility in some perennial grasses (e.g., switchgrass) may allow for the development of high yielding single cross hybrids, and the use of F1 hybrids will have the potential of dramatically increasing biomass yield. Transformation methods (e.g., *Agrobacterium*-mediated transformation of switchgrass) can be used to incorporate value-added genes that cannot be transferred through crossing and selection. A transgene (e.g., for reduced lignin content) should not cause environmental harm; however, a bioenergy crop with reduced lignin content may be less environmentally fit because of its increased pest problems and the need for chemical control (Petersen, 2008). Heritability for biomass yield in perennial grasses is high enough to allow plant breeders to predict and demonstrate adequate gain from selection; however, yield gains per cycle varied from zero to a maximum of 6% and were not linear across cycles (Bouton, 2007; Boe and Beck, 2008). Significant breeding advances have been documented in several perennial grass species for dry biomass yield (DBY), and the potential for increasing their DBY is significant because of the large genetic variation available within the species. For example, genotypes of Bermuda grass (*Cynodon dactylon* L. Pers.) bred for high DBY produced twice as much as the unimproved, and recent yield trials indicated that switchgrass yields were 50% greater than those achieved in early 2000 (Anderson et al., 2009). Lignocellulosic yield of perennial grasses and SRC trees parallels their DBY, which is total yield of all harvested components with only the water removed; therefore, improvements in DBY should be part of all future breeding efforts. Finally, the development of an index for instant determination of “energy value” can be a valuable tool for plant breeders and growers to tailor hybrid selection and crop management to give the highest DBY possible. Improvement of these traits can be achieved through conventional breeding and selection based on existing genetic variation or through transgenic and GM technologies. The latter can be used to introduce new genes, modify existing genes or interfere with gene expression (Gressel, 2008).

Genomics and genetic modification of bioenergy crops

The next generation of bioenergy crops is being developed (Bush, 2007; Grattapaglia et al., 2009; Rubin, 2008) using marker-assisted breeding and the creation of hybrids and transgenics with a broad portfolio of proven traits, such as DBY, plant architecture, tolerance to biotic and abiotic stresses, NUE, and WUE. Genomic information gathered from across the biosphere, including potential bioenergy crops and microorganisms able to breakdown biomass, will be vital for improving the prospects of significant cellulosic biofuel production from SGECS and dedicated energy crops with reduced conversion costs and favorable GHG profiles (Oliver et al., 2009). Many of the traits targeted in the

genomes of energy-relevant plants for optimization in potential cellulosic bioenergy crops are those that would improve growth on poor soils and minimize competition with food crops over land-use, and affect growth rate, response to competition for light, branching habit, stem thickness and cell wall chemistry (Rubin, 2008). Genetic engineering could produce crop plants with reduced biomass conversion costs by developing crop cultivars with less lignin, crops that self-produce cellulase enzymes for cellulose degradation and ligninase enzymes for lignin degradation, or plants that have increased cellulose (i.e., polysaccharides) or an overall larger DBY using genes for delayed flowering (Lee, 1999; Sticklen, 2007; Lobell et al., 2008). Genetic modification (GM) could be a useful tool in developing fast-growing bioenergy crops to gain higher yields from lower inputs, and to reduce GHG emission through lower inputs and reduced or no tillage of perennial bioenergy crops. GM bioenergy crops offer great potential for GCC adaptation and mitigation through multiple resistances or tolerance to biotic and abiotic stresses, herbicides, salinity and environmental toxicity (O'Leary, 1993). Preprocessing *in planta* via expression of cellulases and cellulosomes could potentially reduce the cost of enzymatic saccharification of lignocellulosic biomass (Ortiz, 2008). Alterations of the ratios and structures of the various macromolecules forming the cell wall are a major target in bioenergy crop domestication and development. This allows for easy post-harvest de-construction of these macromolecules at the cost of a less rigid plant. The genetic engineering industry is actively seeking ways of using GM to simplify and streamline processes to breakdown cellulose, hemicellulose and lignin, so as to produce inexpensive and environmentally-friendly biofuels more easily and efficiently from plant biomass (Sticklen, 2007). Tree genomic research already identified genes for increased C partitioning to above-ground woody matter, increased cellulose availability for enzymatic digestion, manipulated genes for N metabolism, delaying senescence and dormancy, and increased PS and adaptation to drought and salinity. Mapping of genomes of ~40 feedstock, feedstock model crops, and eight energy-producer microorganisms is already in draft form, in progress or completed. Genomic information and resources are being developed that will be essential for accelerating their domestication. *Populus trichocarpa* was the first tree and potential bioenergy crop to have its genome sequenced (Tuscan et al., 2006). Quantitative trait loci hotspots serve as useful targets for directed breeding for improved biomass productivity that may also be relevant across additional poplar hybrids and QTL mapping identified regions of genetic control for biomass yield (Rae et al., 2009).

Genetic models and ideotypes of bioenergy crops

The ideotype concept was fundamental to understanding the physiological reasons behind the breeding success of the green revolution of the 1960s. A model plant was expected to yield a greater quantity of grain, oil, or other useful product when developed as a crop cultivar. The cereal ideotype's phenotypic characteristics were a short stem, small erect leaves, a low number of tillers, and a large and awned ear. Such a plant was designed to be a weak competitor to reduce intra-crop interference and thereby maximize yield per unit area. Yield is a property of a population of plants, and is poorly correlated with the performance of an individual plant in the population. Advancing appropriate genetic model systems for bioenergy crops is essential in the development of systems approaches to improve cell wall architecture and plant anatomies for the end-use of biofuel production and for GCC adaptation and mitigation. Therefore, the ideotype for a

bioenergy crop seems to be quite different from that of a food crop. A number of traits to maximize radiation interception, WUE and NUE have been suggested (Ehrlich and Pringle, 2008; Karp and Shield, 2008) to develop bioenergy crop ideotypes. Additionally, traits that may provide a variety of ecosystem services, such as C sequestration, biocontrol, pollination and biodiversity conservation, as components in the sustainable production of bioenergy should be considered in bioenergy crop models and ideotypes. Corn and sorghum are suggested as genetic models for the improvement of perennial C₄ bioenergy grasses. Both crops have close evolutionary relationship with future bioenergy perennial grasses, C₄ photosynthetic pathway, historical depth of genetic knowledge and a rapidly growing resource of genetic tools. Also, rice (*Oryza sativa*) and brachypodium (*Brachypodium distachyon*), a grass with a small genome, are suggested as comparative models for grass cell biology. These model crops can help provide answers to (1) how C₄ metabolism arose, (2) how C₄ grasses partition C into sugar stores versus cell wall mass, and (3) what are the genetic basis of several physiological and architectural traits, such as tillering, canopy formation, stalk reserve retention, perennial growth habit, and water and nutrient use efficiencies. Identifying whether these traits are determined by major genes or quantitative trait loci (QTL) is of foremost importance (Murray et al., 2008b; Murray et al., 2009). The SRC is considered to be amenable to ideotype breeding, and poplars are recognized as model systems for woody species, with a broad genetic base for breeding, an extensive understanding of genetics, biology, and physiology, the availability of sequenced genome and a well-established set of molecular tools that can be used for improvement of bioenergy SRC and other tree species. Among 32 willow genotypes, at least two alternative growth strategies were identified: (1) a large number of thin stems, relatively low LAI and SLA; or (2) larger-diameter stems, and high LAI and SLA; both strategies gave high yields, therefore, multiple SRC ideotypes may need to be selected (Ray et al., 2004).

Environmental impacts of bioenergy crops

The global C fixed by all crops is already exceeded by the C released by fossil fuel combustion, thereby imposing hard biophysical constraints on food and feed production (Lal, 2008b). Biofuel production from bioenergy crops has a variety of positive and negative effects on local and regional environments, and may help relax some of these constraints (Petersen, 2008). The energy output and GHG balances of bioenergy cropping systems differ depending on the type of biomass sources, conversion and end-use technologies, system boundaries and reference energy system with which the bioenergy chain is compared. Therefore, assessing the environmental performances of bioenergy crops and their biofuels is a complex task (Boehmel et al., 2008). It implies covering a wide range of different bioenergy crops as sources of diverse feedstocks, conversion technologies, land-use options, and issues related to land-use change, as well as aspects related to the substituted products, including fossil transport fuels, as well as food and feed. Bioenergy crops are dependent on the functioning and integrity of ecosystems and particularly on ecosystem services related to soil, air, water, and biodiversity; therefore, bioenergy crops will have environmental effects beyond their impacts on GHG emissions. Multi-functional bioenergy crop plantations produce additional environmental benefits (Grammelis et al., 2008; Landis et al., 2008), either (1) dedicated to environmental services, such as vegetation filters for waste water and sewage sludge treatment, and shelter belts against soil

erosion; or (2) generating more general benefits, such as soil C sequestration, increased soil fertility, and removal of toxic elements (Lal, 2008b). Legitimate concerns exist about the relative climate benefits of various biofuels and competition for the limited land resources between food, fiber, fuel and other ecosystem services (Lobell et al., 2008; Jessup, 2009). Agricultural cultivation of bioenergy crops accounts for most of environmental impact; therefore, a trade-off is needed between minimizing GHG emissions and reducing ecological impact knowing that most biofuels that may reduce GHG by >30% have a higher ecological impact than fossil fuel. The overall environmental impact of bioenergy production is to a large degree determined by the scale of direct and indirect land use change, whether for total GHG balance or the conservation of natural resources and biodiversity (Searchinger et al., 2008). Herbaceous perennial grasses may provide improved soil structure and function, which would reduce run-off and erosion risk. Grassland ecosystems are usually more biodiversity-friendly than cropping systems. Perennial poly-cultures offer a low-input, less polluting, and more efficient alternative to annual monocultures for bioenergy production. The use of diverse native perennial grasses may be a viable alternative to monocultures of grass species as they require fewer inputs, promote biodiversity, and reduce the risk of becoming invasive (Fransworth and Meyerson, 2003). The conversion of biomass to liquid biofuels is 2.3- to 3-fold less efficient than that for converting oil to liquid fuels, while crude oil and biomass are converted to heat and electricity in stationary plants with almost equal efficiencies. BtL emerged with a better environmental profile relative to cellulosic ethanol when indirect land-use change is considered (Schenk et al., 2008). The risk that biofuel deployment could accelerate and worsen the current unsustainable trends of deforestation and depletion of natural resources in the framework of accelerated growing population, and food and feed demand; deforestation only accounts for 20% of worldwide GHG emissions (Karp and Shield, 2008). Also, many of the SRC plantations established today are causing a range of environmental and social problems, including loss of biodiversity, soil erosion, and displacement of local people.

Land use (LU) and land-use changes (LUC)

The land used to grow bioenergy crops for biofuels increased from 13.8 Mha in 2004 (~1% of global cropland) to 26.6 Mha in 2007; for example, corn area increased 19% between 2006 and 2007 in the US and resulted in reduced crop diversity in many parts of the Midwest (Gillingham et al., 2008; Searchinger et al., 2008). Biofuel production opportunities in developing countries are being fueled by the apparent relative availability of land to grow bioenergy crops; however, this raises concerns about potential added social and environmental pressures, including environmental consequences resulting from LUC such as GHG emissions and loss of biodiversity. Growing millions of hectares of land under bioenergy crops will put intense pressure on land both for food production and for natural resources conservation and sustainable utilization, and the clearing of natural ecosystems to grow bioenergy crops may create a C debt of greater GHG emissions than the fossil fuels they replace. Therefore, the effect of large-scale cultivation of bioenergy crops will ripple through the global economy and the global ecology via changes in commodity prices and consequent LUC (Muller, 2009; Sanderson and Adler, 2008). It must be ensured, however, that any further land expansion for biofuel production will provide a positive contribution to GCC mitigation knowing that LUC is source of the most

significant GHG emissions. Therefore (Tilman et al., 2009), integrated biotechnological solutions are needed to reduce the adverse environmental impact and limit increased cultivated land under bioenergy crops. Nevertheless, deployment of bioenergy crops at a large scale will result in major LUC. It was estimated that land area required to meet 100% of biodiesel world demand by 2030 by growing *Jatropha* would be 173 Mha, oil palm (48 Mha), or soybean (361 Mha). Whereas, land area required to meet 100% ethanol demand growing corn would be 147 Mha, sugar cane (70 Mha), or sweet sorghum (116 Mha). Mean annual CO₂ emissions (Mt CO₂/yr by 2030) from land conversion to bioenergy crops under different scenarios where each crop is assumed to meet 100% of the biodiesel demand if planted to *Jatropha* (537), oil palm (857), or soybean (1119); and to meet 100% of ethanol demand if planted to corn (706), sugar cane (216), or sweet sorghum (360) are likely to be greater than the savings expected from the first 30 years of growing these bioenergy crops (Gillingham et al., 2008). Converting rainforest, grassland, peatlands, and savannas to produce biofuels in Brazil, SE Asia, and the US may result in a biofuel C debt by releasing 17-420 times more CO₂ than the annual GHG reductions that these biofuels would provide by displacing fossil fuels (Nass et al., 2007). On the other hand, estimates of soil C stock change (t C/ha), as an indirect contributor to GHG emissions and global warming, due to direct LUC from crop reserve program (CRP) in the US, temperate grassland, or tropical grassland to biofuel production (e.g., sugar beet, sugarcane, oil palm, rapeseed, and soybean) can be extremely large. It ranged from -9 to -13 t C/ha; and an extreme value of -31 t C/ha was reported for a tropical rain forest when converted to soybean production. However, estimates of above-ground C stock changes were almost zero, except when converting temperate forest or tropical rain forest to cultivate any of these crops (-35 to -120 t C/ha) and the only gain was when converting tropical grassland to oil palm production (63 t C/ha) (Lal, 2008a).

In order to be a viable energy source, bioenergy crops will have to environmentally and economically compete successfully with other LUs for a share of the finite land resources, and the extent to which bioenergy crops displace other crops will influence global LU and the global agricultural system. Most LUC studies (Searchinger et al., 2008) have found that replacing gasoline with ethanol modestly reduces GHG if made from corn and substantially if made from cellulose or sugarcane. Because growing bioenergy crops removes CO₂ from the atmosphere, biofuels from bioenergy crops can, in theory, reduce GHG emissions relative to fossil fuels. Continued development of FGECs (e.g., corn, soybean and sugarcane) could lead to desertification as more land in natural ecosystems is hastily brought under cultivation. Use of US crop lands for biofuels may lead to increased GHG emissions from LUC. Corn-based ethanol, instead of producing a 20% savings, nearly doubles GHG emissions over 30 years and increase GHG for 167 years. Biofuels from switchgrass, if grown on US corn lands, may increase GHG emissions by 50% (Casler et al., 2007a,b; Vogel and Mitchell, 2008). Although abandoned agricultural land (globally estimated at 385-472 Mha) is not a major source of biomass, it can be an important source at a regional level. Worldwide, the weighted mean above-ground biomass production potential is about 4.3t/ha/yr; however, in tropical grassland regions it can be as high as 7-20 t/ha/yr. Improvement in bioenergy crop yield, composition, and processing technology will minimize land area needed to produce target energy (McKendry, 2002).

Water footprint of bioenergy crops

At present, the contribution of bioenergy crops in overall agricultural water demand is modest, this may increase as rising energy prices, geopolitics, and concerns over the impacts of GHG emissions drive increased biofuel production. This potentially leads to more intensive competition between food and biofuel for land and water resources, particularly in already water-scarce parts of the world (Gerbens-Leenes et al., 2009). Large-scale bioenergy crop plantations pose both opportunities and challenges to the water sector, and much depends on the choice of species, genotypes, location of production, prevailing management practices, and water management options; however, water scarcity may prove to be the limiting factor for the establishment and growth of bioenergy crops, and for biofuel production in many contexts. For example, deep-rooted bioenergy crops are usually more drought tolerant and capable of sequestering more C; however, they are more likely to modify the water and nutrient dynamics in soils, and they may negatively impact biodiversity (Ehrlich and Pringle, 2008). Growing bioenergy crops for biofuels currently accounts for $\sim 100 \text{ km}^3$ (1%) of all water transpired by crops worldwide, and about 44 km^3 (2%) of all irrigation water withdrawals. Many crops (e.g., corn, sugar cane, oil palm) have high water requirements at commercial yield levels and are best suited to high-rainfall tropical areas, unless they can be irrigated (Fraiture et al., 2008). Water requirements of different types of bioenergy crops per unit of energy produced varies largely due to several plant, environmental and management factors (GIMES, 2009; Gerbens-Leenes et al., 2009). For example, total water requirements estimated as evapo-transpiration (ET) in m^3/GJ for rapeseed is 100-175, oil palm (46-250), soybean (143-500), sugarcane (18-35), sugar beet (70-180), corn (100-300), wheat (40-350), sweet sorghum (56-230), and lignocellulosic bioenergy crops (11-170). Water footprint (WF; m^3/GJ) of bioenergy crops relates the energy yield of a crop to its actual water use under actual field conditions during the growing season, and depends on crop type, agricultural production system and climate. WFs show large variations for similar crop types, depending on the agricultural production systems and climate conditions. Most estimates of WFs show variations of a factor of 4 to 15. On average, estimates were small ($24 \text{ m}^3/\text{GJ}$) in the Netherlands, medium (58-61) in the US and Brazil, and large (143) in Zimbabwe (Gerbens-Leenes et al., 2009). These estimates are 70-400 times larger than the WF of other primary energy carriers. When expressed per L, the WF ranges from 1,400 to 20,000 L of water per L of biofuel; most of which (>90%) is used in the production of the feedstock. Therefore, crops and conversion technologies (e.g., combustion vs. ethanol or biodiesel) should be selected for the highest WF. Moreover, it is more efficient to use total biomass than a fraction of the crop (e.g., seed) for biofuel production, especially when water is a limiting factor (McKendry, 2002). The WF of ethanol appears to be smaller than that for biodiesel. For example (GEMIS, 2009), a WF of $50 \text{ m}^3/\text{GJ}$ was obtained when corn, sugar beet, or sugarcane were used to generate electricity; whereas rapeseed and *Jatropha* spp. were at a disadvantage with a WF of $400 \text{ m}^3/\text{GJ}$. When used for ethanol production, the WF for sugar beet and potato (60 and $100 \text{ m}^3/\text{GJ}$, respectively) were better than for sweet sorghum ($400 \text{ m}^3/\text{GJ}$). The WF estimate for soybean and rapeseed when used to produce biodiesel ($400 \text{ m}^3/\text{GJ}$) were better than that for *Jatropha* spp. ($600 \text{ m}^3/\text{GJ}$).

Impact of bioenergy crops on biodiversity

Worldwide, habitat loss will be the major driver of biodiversity loss in the next 50-100 years and land conversions, particularly deforestation and conversion of grasslands and savannas, to bioenergy crops are the greatest threat to biodiversity. However, positive impacts on biodiversity may be realized as a result of ameliorating the rate of change of atmospheric composition and global climate and if bioenergy crops and cropping systems can help reduce GHG emissions (Boehmel et al., 2008). Large-scale biomass plantations (e.g., oil palm) often entail the destruction of large areas of rain forest, reducing biodiversity, and impact services and products of the forest ecosystem. Although it can be produced in an environmentally-friendly manner to help mitigate GCC and preserve biodiversity, oil palm, as it is currently practiced, contributes to GHG emissions, impacts local environments and replaces important C sinks (McKendry, 2002). Biodiversity consequences of increased biofuel production will mostly result in habitat loss, increased and enhanced dispersion of invasive species, and pollution resulting from fertilizer and herbicide use. Moreover, extinction of genetically distinct populations, decreases in effective population sizes, and habitat uniformity due to large-scale deployment of bioenergy crop monocultures are likely to have negative effects on future biodiversity (Ehrlich and Pringle, 2008). Vegetatively-propagated species (e.g., *Panicum virgatum* and *Miscanthus* spp.), which are more likely to tolerate poor soils and grow in dense stands are likely to become invasive. Invasive species will have large negative impact on native biodiversity. Non-native species and genotypes may facilitate native species extinction, alter the composition of ecological communities, and alter ecosystem processes such as water filtration and nutrient cycling. Therefore, it is suggested (Fransworth and Meyerson, 2003) to identify plant traits that contribute to or avoid invasiveness in potential bioenergy crops, and incorporate desirable traits such as sterility, reduced seed production, and inability to reproduce vegetatively, into the germplasm of bioenergy crops to minimize potential invasiveness. However, sterility in bioenergy crops (e.g., poplar) may deprive pollinators of a food source and negatively reduce a valuable ecosystem service (Ferre et al., 2005). Biological control using insects as natural enemies is an ecosystem service that is strongly influenced by local landscape structure. The development of lignocellulosic dedicated energy crops and biofuel production processes that use a variety of feedstocks could increase diversity in agricultural landscapes and enhance arthropod-mediated ecosystem services (Landis et al., 2008). On the other hand, pollution from fertilizers and pesticides associated with large-scale deployment of bioenergy crops as monocultures is anticipated to impact terrestrial and aquatic biodiversity; eutrophication caused by nutrient pollution often leads to changes in biogenic habitats (Fransworth and Meyerson, 2003).

Climate change, C-sequestration and GHG mitigation

A proliferation of studies, advocating or opposing biomass for bioenergy to combat GCC, cover a wide range of bioenergy crops, production schemes, and conversion processes, and present a range of positive and negative results and recommendations (Lal, 2008b). For example, biofuels derived from biomass in China (6.5×10^{10} metric t/yr) and crop residues (714.7×10^6 t) could account for 10 and 16% of national energy supply in 2010 and 2020, respectively. These biofuels help reduce GHG emissions of SO_2 (54% of national

emissions in 2003), NO_x and CO₂ (30% of emissions in 2003) (Fan et al., 2007). On the other hand, studies in Europe and the US reported GHG improvements (over fossil fuels) of FGECs ranging from -35 to 65% for sugar beet, from -55 to 85% for rapeseed, and from -40 to 78% for soybean; whereas, positive values (40-70%) were reported for oil palm in Malaysia and sugarcane (85-100%) in Brazil as compared with 67 to 115% for lignocellulosic ethanol and 60 to 115% for biodiesel from a number of SGECS (Nass et al., 2007; Oliver et al., 2009). One important factor which is often overlooked when considering the use of biomass to assist alleviating global warming and mitigating GCC impact, is the time lag between the instantaneous release of CO₂ from burning fossil fuels and its eventual uptake as biomass, which can take many years (McKendry, 2002). However, information on the environmental impacts of a few important bioenergy crops and their biofuels (e.g., corn, sugarcane, and rapeseed) is incomplete, or contradictory, and highlight major gaps in our knowledge that need to be addressed before a truly quantitative assessment of these and other bioenergy crops can be made. Specifically, knowledge gaps on potential environmental impacts of biofuel production and use on acidification, due to the manufacturing and use of synthetic fertilizers, eutrophication and toxicity, photochemical ozone, and abiotic depletion need to be addressed (Kotchoni and Gachomo, 2008). Advanced biofuels (Gutterson and Zhang, 2009), including lignocellulosic parts of FGECs and most SGECS, mitigate GCC by allowing further GHG emission reductions (e.g., ethanol produced from wheat straw only releases 20 gCO₂/km along its life cycle while petrol releases on average 163 gCO₂/km). They reduce pressure on food crops and on land use as they require less land to grow the same amount of feedstock (whole crop vs. stover), and produce useful by-products (e.g., for the chemical industry, or as fertilizer). Biofuels derived from dedicated energy crops (e.g., SRC) have greater potential for GCC adaptation and mitigation. They can enhance and maintain soil structure and function which will help cope with floods and droughts, reduce nutrient loss and pesticide pollution, and they require fewer inorganic fertilizer inputs (Gillingham et al., 2008; Gutterson and Zhang, 2009). Contradictory estimates of C balance of bioenergy crop production systems are a product of different methods and models used to assess C release and fixation. Perennial crops are more suitable for C sequestration than annuals due to several agronomic, physiological, and structural characteristics (Lal, 2008b; Sanderson and Adler, 2008). The efficiency of C sequestration is reduced when C and N are not adequately balanced. The rate of C sequestration ranges from negative or zero under arid and hot climates to approximately 1000 kg C/ha/yr under humid and temperate climates. With choice of the appropriate species and prudent management, biofuels produced from dedicated energy crops (e.g., SRC poplar, willow, switchgrass, *Miscanthus*, karnal grass, *Andropogon*, *Pennisetum*) can sequester C in soils, offset fossil fuel emissions and reduce the rate and abundance of atmospheric CO₂ and other GHGs (Gutterson and Zhang, 2009). When planting fast growing dedicated energy crops (e.g., SRC) with the purpose of C sequestration, the overall impact on the full budget of GHG depends on the preceding land cover, type of management, and LUC of the plantation. For example, a decrease of 61 (top 10 cm) to 25% (top 45 cm) of C was observed under poplar trees in land converted from natural forest (Lewandowski and Schmidt, 2006). Bioenergy crops are often promoted to help reduce global warming; they provide the greatest GHG benefits if grown on land where C losses from conversion are small. Estimated GHG emissions from land conversion for bioenergy cropping under different

bioenergy crop scenarios in Africa, for example, range from 29 (*Jatropha* spp. and sugarcane) to 71 Mt CO₂ (soybean and corn) annually. However, some biofuel systems (i.e., whole life cycle of biofuel production) can increase GHG emissions relative to the fossil fuels they replace, thus aggravating global warming, with the agricultural production of biofuels being responsible for a substantial share of GHG emissions and water quality degradation (Gutterson and Zhang, 2009). The wide range of GHG savings of biofuels can be largely attributed to the type of energy inputs used to transport bioenergy crops to “biorefineries” and to make the biofuels, and to differences in co-product allocation methods (Adler et al., 2007; GIMES, 2009). The reduction in GHG emissions is more than 10-fold when using switchgrass for heating–replacing coal – than when producing ethanol from corn – replacing gasoline, and GHG improvement values >100% reported for sugarcane are due to credits for co-products in the sugarcane industry. GHG emissions (gC-eq./MJ) and energy ratio in MJ differ due to biomass source and conversion method (McKendry, 2002). For heat generation, SRC and *Miscanthus* provide clear C savings as compared with liquid biofuels from FGECs due to extra GHG emissions during hydrolysis, fermentation and distillation. On average, GHG emissions and energy ratio for *Miscanthus* for heat generation (0.512 and 35.86, respectively) are more favorable than those for corn ethanol (5.4 and 1.5, respectively) (Gutterson and Zhang, 2009). Nitrogen fertilizer inputs contribute to large portions of the GHG emissions. Due to its potency (298 times greater than CO₂), even very small changes in the N balance and rate, N₂O emissions can significantly affect the overall GHG balance results of bioenergy crops. Generally, spring-sown FGECs have potential to lower N₂O because they require lower N inputs than the winter-sown. The high global warming potential of N₂O of FGECs, as compared to dedicated energy crops, is due to their large N fertilizer requirements, and those grown in high rainfall or irrigated environments have the highest N₂O production (Searchinger et al., 2008; Cherubini et al., 2009). Changes in cropping systems in response to GCC will alter C and N flows resulting from changes in crops, residue amounts and qualities, and mineralization of organic matter; therefore, GHG emissions could be reduced more efficiently by managing C and N simultaneously (Tubiello et al., 2007; Eisenbies et al., 2009). Annual bioenergy crops may contribute more N₂O than perennials since their rates and frequency of fertilizer applications are larger. Bioenergy crops grown under high rainfall need more N fertilizers and have the highest N₂O emissions as denitrification (Sanderson and Adler, 2008). Although efficient, the starch grain-based route consumes more energy, and therefore, emits more CO₂, than the sucrose-based route. The GHG savings in t CO₂-eq./ha/yr for ethanol from sugarcane (10-16), ethanol from corn and sugar beet (0.5-11), biodiesel from rapeseed, soybean and sunflower (0.4-4), and ethanol from lignocelluloses (2-7). The GHG savings for electricity and cogeneration of sweet sorghum ranged from 2-29, and for *Miscanthus* and switchgrass from 2 to 33; however, when these two bioenergy crops were used for heat generation, the GHG savings ranged from 14 to 46, and 13 to 58 t CO₂-eq./ha/yr, respectively (GEMIS, 2009). The main GHG implications related to crop residue removal are: (1) crop productivity showed a decrease in yield of 0.05-0.15 t dry/ha because of a lower net mineralization of N in soils, (2) N₂O emissions decreased slightly with increasing straw removal, at a rate of 0.1-0.25 kg N/t dry straw, and (3) straw removal contributed to global warming due to the change of soil C stocks, in comparison with the case in which straw left in the ground (Lehtomaki et al., 2008; Eisenbies et al., 2009)

Nutrient composition, cycling and loss

The implications for GHG balances of bioenergy cropping systems arise from an increase of synthetic fertilizer application to replace nutrients removed with biomass or lost from the soil through leaching, and to sustain crop yields (Heggenstaller et al., 2008). Biomass is a complex heterogeneous mixture of organic and inorganic matter; it contains various solids, fluids, and minerals of different origins and in different ratios and chemical bonds (El-Nashaar et al., 2009). Biofuel quality and chemical composition have not received adequate attention given that it is an important aspect in the introduction of bioenergy crops. Biofuel quality changes due to crop-specific mineral uptake, may change with biomass partitioning, and can drastically impact net energy output thus limiting the effectiveness of conversion processes and decreasing energy value, which, in turn, is negatively related to ash content; with every 1% increase in ash concentration decreasing the heating value by 0.2 MJ/kg (Monti et al., 2008; Schmer et al., 2008; Persson et al., 2009). As bioenergy applications often demand different bioenergy crops and biomass quality traits, new bioenergy crops and cultivars with specific mineral contents (e.g., low ash, N, Cl, and K) are needed to fit this demand. The chemical composition of biomass depends on several factors, including plant species, plant organ and its age, growing conditions, management practices, including fertilizer and pesticide application, and harvest time and pre-treatment (El-Nashaar et al., 2009). Nutrients in biomass can be classified as major (>1.0%), minor (0.1-1.0%), or trace (<0.1%) elements according to their elemental concentration on dry weight basis. The major nutrients, in decreasing order are C, O, H, N, Ca, and K. The minor nutrients include Si, Mg, Fe, P, Cl and Na; whereas, the most important trace elements are Mn and Ti, and the ash-forming elements may include some or all of the aforementioned. Bioenergy crops are classified on the basis of their chemical composition and mineral content to evaluate their suitability for different conversion processes (El-Nashaar et al., 2009). Similar contents of C, H, O and significant differences in the contents of N and ash-forming nutrients have been reported for different bioenergy crops (Zhao et al., 2009). Variations reported for woody bioenergy crop species in Al, Mn, Na, and Si were larger than variations in Ca, Cl, Fe, K, Mg and P. In general, bioenergy crops have high Cl and S contents, which are strongly associated with corrosion and HCl emissions, with annual and fast-growing crops having the greatest contents of ash, moisture, and highly mobile Cl, K, Mg, N, P, and S in comparison with woody bioenergy crops (Monti et al., 2008). On the other hand, the large diversity reported for sulfur content in about 700 *Eucalyptus* species offers the potential for sulfur-free biofuel (Rockwood et al., 2008). The extended growing season, high ET rates and extensive root systems of perennial dedicated energy crops (e.g., SRC, *Miscanthus*, and switchgrass) has led to much interest in the effects they may have on N cycling, leaching and related changes in water quality (Casler et al., 2007a). Perennial dedicated energy crops have inherent advantages in their ability to recycle nutrients, achieve major reductions in N and P losses to the environment, and to fully exploit the growing season. It was estimated that switchgrass can replace corn and soybean on productive lands with 75-90% reduction in N and P losses; whereas, increasing corn cultivation would lead to an increase of 10-30% in the annual average flux of dissolved inorganic N to the Mississippi River (Heggenstaller et al., 2008), and the cultivation of oilseed rape can be questioned on environmental grounds as *Brassica* spp. emit more methyl bromide than any other crop. Legumes are less productive

than cereals or perennial bioenergy crops for biomass production; however, when included in bioenergy cropping systems, the energy savings obtained through less N input must be balanced with the loss of potential yield. The potential biological nitrogen fixation (BNF) for most legume species is in the range of 200-300 and a maximum of 450 kg N/ha/yr for the potential bioenergy crop *Lupinus albus*. However, the cost of N uptake for inoculated *Lupinus albus* is 2.9-6.1 gC/gN and when supplied by N fertilizer the cost would be 0.8-2.4 gC/gN. Nevertheless, legumes can play a positive role as fast growing cover crops to provide additional biomass yield, enrich SOM, and provide protein feedstock for the chemical industry (Sanderson and Adler, 2008).

Life cycle analysis (LCA)

Bioenergy cropping systems vary with respect to length of the plant life cycle, yields, feedstock conversion efficiencies, nutrient demand, soil C input, and N losses. These factors affect the magnitude of the components contributing to net GHG fluxes and N losses (Adler et al., 2007; Heggenstaller et al., 2008). Assessment of the GHG implications of LU and LUC to bioenergy crops is a very complex and contentious issue. Life cycle analysis is used to reveal the validity of bioenergy as a means to reduce GHG emissions (Berndes et al., 2003). It requires (1) an accounting of all GHG emissions associated with growing, processing and transporting bioenergy crops and biofuels, (2) the land categories that will be cleared in response to increased biofuel demand, (3) the C stocks present in those land categories along with the rates of release of C associated with the land conversion, (4) potential C uptake rates in those land categories if the current LU pattern continues, (5) the quantity of fossil fuels to be replaced by biofuels to meet the projected demand, (6) the bioenergy crops selected (e.g., C₃, C₄; or oilseed, lignocellulosic crops, etc.), (7) biofuel yields and the likely rates of change in future yields, and (8) the quantities of by-products of bioenergy crops and their potential uses (Cherubini et al., 2009; Davis et al., 2009). The Global Emission Model for Integrated Systems (GEMIS, 2009), used in conducting LCA reported in this review, maintains a database for energy, material and transport systems and includes the total life cycle in its calculations of impacts. The GEMIS dBase covers for each process: efficiency, power, direct air pollutants, GHG emissions, solid wastes, liquid pollutants and land use. Most LCA studies reported significant net reductions in GHG emissions and fossil energy consumption when ethanol and biodiesel are used to replace fossil fuels (Cherubini et al., 2009; Davis et al., 2009). A few studies examined impacts of LUC on local air pollution, acidification, eutrophication, and ozone depletion, and concluded that the positive impacts on GHG emissions may carry an additional environmental cost (Davis et al., 2009). Several LCA studies concluded that bioenergy is the superior LU option delivering the greatest mitigation benefits where bioenergy crop growth rates are high, biomass is used efficiently, initial C stocks are low, and a long-term view is taken (Cherubini et al., 2009). Land converted from row crops to perennial dedicated energy crops showed an increase in C sequestration of up to 1.1 t C/ha during five years. Other studies reported increases in soil C at rates of 0.2-1.0 t C/ha/yr for several decades (Searchenger et al., 2008). Pay-back time for grassland ecosystems converted to sugarcane or oil palm is <10 yr because these ecosystems have the lowest C reserves and the highest yielding bioenergy crops (Berndes et al., 2003; Davis et al., 2009). Therefore, C sequestration related to LUC might broaden the GHG mitigation benefits of bioenergy crops

beyond GHG emission savings; however, the positive impacts on GHG emissions may carry a cost in other environmental areas, so that a much more careful analysis is needed to understand the trade-offs in any particular situation (Cherubini et al., 2009). The wide range of the combined direct and indirect LUC, estimated in g CO₂-eq.-C/MJ, of rapeseed to Fatty Acid Methyl Esters (FAME) in the EU (117-260), oil palm to FAME in Indonesia (45-84), soybean oil to FAME in Brazil (51-100), sugarcane to ethanol in Brazil 36-48, corn to ethanol in the US (72-130), and SRC to BtL in Brazil (17-34), demonstrates the large variability within and among crops, production systems, and countries; with GHG emissions from indirect LUC considered as more important than emissions from direct LUC (GIMES, 2009). Land-use change from native habitat to bioenergy crops lead consistently to significant GHG emissions and a negative C balance, or C-debt. The N₂O emissions, due to N fertilizer production and field application, constitute a serious uncertainty source in the LCA results of many biofuel pathways (Nass et al., 2007).

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