Research paper

Agronomic characterization of Taiwan grass [Cenchrus purpureus (Schumach.) Morrone] and evaluation of its potential to produce bioethanol in the warm sub-humid climate of Mexico

Caracterización agronómica del pasto Taiwán [Cenchrus purpureus (Schumach.) Morrone] y evaluación de su potencial para la producción de bioetanol en clima cálido subhúmedo de México

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Abstract

The objective of this study was to evaluate the biomass production, chemical composition, proximate analysis, calorific value and theoretical yield of bioethanol of Taiwan grass under 6 cutting frequencies. The highest production of biomass (33 t DM/ha), cellulose content (41.3%), calorific value (17.5 MJ/kg DM) and potential bioethanol yield (7,936 L/ha) were recorded at a cutting frequency of 180 days. The highest moisture content of the dehydrated samples and ash and crude protein concentrations were observed at a harvest frequency of 30 days with 9.2, 12.1 and 10.5%, respectively. The highest concentrations of extractives were obtained at harvest frequencies of 60 and 120 days (13.9 and 13.7%, respectively), while lignin concentrations were greatest at harvest frequencies of 150 and 180 days (21.1 and 20.9%, respectively). The highest concentration of fixed carbon was observed at a harvest frequency of 90 days (18.5%), while the lowest concentration of volatile matter occurred at a harvest frequency of 30 days. The data indicate that Taiwan grass has significant potential for use to produce bioethanol but assessment of the carbon footprint, life cycle analysis, energy yield (energy produced:energy consumed) of the entire production process is needed to ensure there are positive effects on climate change and greenhouse gas emissions before this process is adopted.

Keywords: Biofuel, calorific value, chemical composition, cutting frequencies, Pennisetum.

Resumen

El objetivo de este estudio fue evaluar la producción de biomasa, composición química, análisis proximal, valor calorífico y rendimiento teórico de bioetanol del pasto Taiwán (Cenchrus purpureus Schum.) Morrone a seis frecuencias de corte. La producción más alta de biomasa, contenido de celulosa, valor calorífico y bioetanol se registró en el corte de 180 días con 33 Mg DM/ha, 41.3%, 17.5 MJ/kg DM, and 7936.2 L/ha, respectivamente. El contenido mayor de humedad, cenizas y proteína cruda se observó a la frecuencia de corte de 30 días con 9.2, 12.1 and 10.5%, respectivamente. La concentración mayor de extractivos fue obtenida en la frecuencia de corte de 60 y 120 días (13.9 y 13.7%), y la lignina las frecuencias de corte de 150 y 180 días mostraron los mayores valores (21.1 y 20.9%), La concentración más alta de carbono fijado se observó a los 90 días (18.5%), mientras que la concentración más baja fue en la frecuencia de corte de 30 días. De acuerdo con los resultados...
obtenidos el pasto Taiwán tiene potencial para ser usado para producir bioetanol, pero se necesita evaluar la huella de carbono, el análisis de ciclo de vida, el rendimiento de energía (energía producida;energía consumida) para asegurar que hay efectos positivos sobre el cambio climático y las emisiones de gases de efecto invernadero para que se adopte este proceso.

**Palabras clave:** Biocombustible, composición química, frecuencias de corte, *Pennisetum*, valor calorífico.

**Introduction**

The depletion of oil reserves and the increase of greenhouse gas emissions have caused a rising interest in the search for alternatives to liquid fuels from lignocellulosic biomass. Biofuels from biomass can be a valuable substitute and a complement to fossil fuels. In addition, they are environmentally friendly, due to the benefit of reducing greenhouse gases (Rio Andrade et al. 2012). The polysaccharides in the grasses can be used as raw material to produce biofuels, once they have been pretreated and decomposed into simple sugars for efficient fermentation. However, the biochemistry of the lignin attached to cellulose hinders the efficiency of hydrolysis and fermentation processes (Ladisch et al. 2010). Cellulose linked to lignin requires greater amounts of enzymes to hydrolyze it, because of its complex structure (Pu et al. 2011). However, in comparison with woody biomass, grass biomass contains lower lignin concentrations, which makes it less recalcitrant to the action of enzymes and leads to simpler pretreatment conditions (Mohapatra et al. 2017). Grasses are considered as dedicated energy crops due to their high yield per hectare, ready availability, utilization of the whole plant, high concentration of carbohydrates and lower lignin concentration than woody species (Ventura et al. 2015). On average, grass biomass contains 25–46% cellulose, 19–46% hemicellulose and 13–30% lignin (Ramos et al. 2013; Godin et al. 2013; Ventura et al. 2015). About 30–35 grass species and varieties are documented to be potentially sustainable feedstocks for cellulosic ethanol production (Mohapatra et al. 2017).

Lignocellulosic biomass from C4 grasses is readily available in the tropical zones of Mexico, where varieties of *Cenchrus purpureus* (Schumach.) Morrone (syn. *Pennisetum purpureum* Schumach.; also known as Elephant and/or Napier grass) have been introduced in the past decades for use in animal feeding. Previous studies indicate that they have a great potential for growth and biomass production, ranging from 37 to 46 t DM/ha (Ramos et al. 2013; Calzada et al. 2014).

However, there are few studies showing the optimum harvesting age for highest production and chemical composition of the biomass to produce bioethanol, although it is reported that age of grass is the factor that most influences the chemical composition of cell walls (Rowell et al. 2012).

While *Cenchrus* grasses have been studied intensively, most evaluations have focused on the production of forage, nutritional value and animal performance (Grajales et al. 2018); fewer studies have evaluated cultivars of this species for bioethanol production (Ventura et al. 2015; Mohapatra et al. 2017). In Mexico, evaluation of the potential of grass biomass to produce cellulosic ethanol is limited. We consider that *Cenchrus* grasses have significant potential to provide biomass for bioethanol production, so designed this study to evaluate the biomass yield, chemical composition, heating value, proximate analysis and theoretical ethanol yield of Taiwan grass (*C. purpureus* cv. Taiwan) harvested at different cutting intervals to determine its potential as a bioenergy crop.

**Materials and Methods**

**Experimental site and sampling**

The experiment was carried out at the “Papaloapan” Experimental Site of INIFAP (18°06’N, 95°31’W; 65 masl) in Cd. Isla, Veracruz, Mexico, with an Awo climate and mean annual temperature of 25.7 °C (Garcia 2004). The soil type is a sandy-loam orthic Acrisol, with a pH from 4 to 4.7 and is poor in organic matter, nitrogen, calcium and potassium and medium to high in phosphorus and magnesium (Enríquez and Romero 1999). The average rainfall recorded during the study is presented in Table 1, with data from the Meteorological Station of the Papaloapan Experimental Site. The experiment started on 22 July 2013, when vegetative material (stems) of Taiwan grass (*C. purpureus* cv. Taiwan) was planted in plots 5 m wide by 16 m long, with 3 replications, and finished on 17 July 2014. Stems were sown in rows with a continuous cord with 4.33 germination points (plants) per linear meter and inter-row spacing of 0.5 m, giving a density of 87,033 plants per hectare. A fertilizer dose of 120:36:0 kg/ha of N:P:K was applied in 2 equal applications (at 43 and 112 days after planting). Six cutting frequencies (30, 60, 90, 120, 150 and 180 days) were compared with 3 replications arranged in a complete randomized block design with split-plots, where the major plot was the grass and the minor plot was cutting frequency. The study continued for 360 days except for the 150-day harvest interval where harvests ceased after only 300 days.
Table 1. Average rainfall during the study in Cd. Isla, Veracruz.

| Month | Jul | Aug | Sep | Oct | Nov | Dec | Jan | Feb | Mar | Apr | May | Jun | Jul | Total |
|-------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-------|
| Precipitation (mm) | 380 | 460 | 110 | 320 | 90 | 40 | 60 | 70 | 20 | 10 | 20 | 136 | 232 | 1,948 |

**Biomass production**

At each harvest a central area of 2 × 3 m was harvested (6 m²) from each plot at 20 cm above ground level. The harvested biomass was weighed on a precision scale (Ohaus, Mod. GT-4000) before a representative sample (15% of the total biomass) was taken, weighed and dried in an oven (Felisa, Mod. FE-243A) at 55 °C until constant weight to determine dry matter yield. Dried samples were ground in a Thomas-Wiley® mill (Arthur H. Thomas Co., Philadelphia, PA, USA) and sieved to pass through a No. 40 mesh (0.42–1.00 mm) and retained on a No. 60 mesh (0.25–0.42 mm). This sieved material was used to perform the chemical and calorific determinations. Following sampling the remaining grass on each plot was cut and removed.

**Proximate analysis**

Moisture content, volatile matter (VM) and ash (on a dry matter basis) were determined according to ASTM E871, ASTM E872 (ASTM 2012) and ASTM D 1102-84 (ASTM 2009) standards, respectively. Fixed carbon (FC) was computed by subtracting the concentrations of ash and volatile matter from the oven-dry sample mass [FC = 100-(VM+Ash)]. The moisture content of the samples was determined on an Ohaus MB45® scale with 3 samples per plot, giving 9 determinations for each cutting frequency.

**Higher heating value**

Higher heating value (HHV) was determined using an adiabatic bomb calorimeter (Isotheribol, Parr 1266) following ASTM E711 (ASTM 1996) standard at 30±0.5 °C, with pellets weighing 1 gram. Five determinations were performed per plot with a total of 15 samples per cutting frequency. Energy production was calculated by multiplying the biomass yield per hectare by HHV.

**Chemical composition**

Extractive release was carried out by following the TAPPI T-264 standard, including lipids (galactolipids, triglycerides and phospholipids), waxes, fat-soluble vitamins, pigments and steroids (Barbosa et al. 2017). Holocellulose concentration was determined by the acid chloride method and ASTM D1104 (ASTM 1977) standard was used for cellulose determination. Hemicellulose was calculated as the subtraction of cellulose from holocellulose. Lignin was determined according to TAPPI T-222 standard and nitrogen concentration by the semi-micro Kjeldahl procedure (AOAC 1990). Two samples per plot were determined giving a total of 6 determinations per cutting frequency.

**Theoretical ethanol yield (TEY)**

The TEY of grass biomass for each cutting frequency was estimated as follows (Badger 2002):

\[
\text{TEY} = (B + B1),
\]

where:

- for cellulose: \(B = C \times \text{RE} \times \text{E} \times \text{GFE}\);
- for hemicellulose: \(B1 = H \times \text{RE} \times \text{E} \times \text{XFE}\);
- \(C\) = kg of cellulose/tonne of dry biomass;
- \(H\) = kg of hemicellulose/tonne of dry biomass;
- \(\text{RE}\) = Recovery efficiency (0.76 for cellulose; 0.90 for hemicellulose);
- \(\text{E}\) = Ethanol stoichiometric yield (0.51);
- \(\text{GFE}\) = Glucose fermentation efficiency (0.75); and
- \(\text{XFE}\) = Xylose fermentation efficiency (0.50).

The unit of bioethanol yield, calculated with this formula, is kg/ha/yr. The density of ethanol (0.789 kg/L) was used to show the results in L/ha/yr.

**Statistical analysis**

The experimental design was a randomized complete block design with: whole plot being genotype and subplot cutting frequency (30, 60, 90, 120, 150 and 180 d), with 3 replications. An analysis of variance (ANOVA) was carried out to investigate the effects of study factors on response variables by using the SAS/GLM procedure and treatment means were compared with the Tukey test (P≤0.05). The data were analyzed to estimate the effect of cutting frequency using SAS for Windows version 9 (SAS 2011).
Results

Biomass production

As harvest interval increased, biomass yields (Table 2) increased from 10.2 t DM/ha/year with harvesting every 30 days to 38.4 t DM/ha/year with harvesting every 180 days (increase of 278%).

Proximate analysis

The results for concentrations of moisture, ash, fixed carbon, volatile matter and higher heating value and energy production are presented in Table 2.

Moisture content of dehydrated grass. As harvest interval increased, moisture content decreased (P<0.05) since plants advanced in physiological development, i.e. from 9.2% at 30-day harvests to 7.0% at harvest intervals greater than 120 days (Table 2).

Ash. Ash concentration decreased (P<0.05) as plants progressed in physiological development from 12.2% at 30-day harvests to 4.5% at 120-day and longer harvest intervals (Table 2).

Fixed carbon. There was no consistent effect of harvest interval on concentration of fixed carbon in the grass with highest value of 18.6% at 90-day harvest intervals and a mean of 16.4% for the remainder (P<0.05) (Table 2).

Volatile matter. The concentration of volatile matter in the grass was similar for harvest intervals of 120, 150 and 180 days (mean 79.3%), which was higher than for the other cutting frequencies, with the lowest value for 30-day harvests (71.3%) (Table 2).

Higher heating value and energy production

Energy concentration in the harvested grass increased from 15.6 MJ/kg at 30-day harvests to 17.2 MJ/kg at 90-day harvests and then plateaued (P<0.05). Energy production increased progressively with harvest interval from 158.5 GJ/ha at 30-day harvests to 675.7 GJ/ha at 180-day harvests (P<0.05) (Table 2).

Chemical composition

Extractives. There was little consistency in the concentrations of extractives in the harvested grass with those from 90- and 180-day harvests being lowest at 7.2 and 7.8% (P>0.05), while remaining treatments varied from 10.2 to 14.0% (P>0.05) (Table 3).

Holocellulose. Holocellulose concentrations did not vary between treatments (P>0.05) with an overall mean of 72.3% (Table 3).

Cellulose. Cellulose is the main feedstock to produce ethanol, since it is a glucose polymer; its concentration increased from 38.3% at 30-day harvests to 42.8% at 90-day harvests (P<0.05) and then plateaued (Table 3).

Hemicellulose. Concentration of hemicellulose in the grass declined as harvest interval increased to 90 days but then plateaued (P<0.05) (Table 3).

Lignin. Lignin concentration increased linearly as plants advanced in physiological development from 17.7% at 30-day harvests to 21.0% at 150- and 180-day harvests (P<0.05) (Table 3).

Crude protein. Protein concentration decreased as plants advanced in physiological development from 10.5% at 30-day harvests to 2.7% at 150-day harvests (P<0.05) (Table 3).

Bioethanol yield

Theoretical ethanol yields that can be produced from the grass biomass for the various treatments are presented in Table 4. Bioethanol yield per hectare increased progressively (P<0.05) as harvest

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Table 2. Average biomass yield (DM), proximate analysis and calorific power of Taiwan grass at 6 cutting frequencies.

| Cutting frequency (days) | Yield (t/ha/yr) | Proximate analysis (%) | Higher heating value (MJ/kg) | Energy production (GJ/ha) |
|--------------------------|-----------------|------------------------|-----------------------------|--------------------------|
|                          | Moisture¹       | Ash                    | Fixed carbon                | Volatile matter          |                             |
|                          |                 |                        |                             |                           |                             |
| 30                       | 10.2 ± 2.60d    | 9.2 ± 0.26a            | 12.2 ± 0.87a                | 16.5 ± 1.42b             | 71.3 ± 2.0c                | 15.6 ± 0.43c                | 158.5 ± 45.09d              |
| 60                       | 10.5 ± 2.03cd   | 8.3 ± 0.28b            | 8.1 ± 0.65b                 | 17.1 ± 0.74ab            | 74.8 ± 0.53b               | 16.2 ± 0.25bc               | 170.2 ± 34.64d              |
| 90                       | 14.0 ± 1.22c    | 7.4 ± 0.26c            | 6.6 ± 0.6 c                 | 18.6 ± 0.66a            | 74.8 ± 1.03b               | 17.2 ± 1.22a               | 239.8 ± 10.92c              |
| 120                      | 18.8 ± 3.40b    | 6.6 ± 0.19d            | 4.7 ± 0.30d                 | 16.4 ± 1.94b            | 78.8 ± 1.89a               | 17.0 ± 0.32ab              | 319.4 ± 61.39b              |
| 150                      | 18.2 ± 2.42b²   | 6.9 ± 0.26d            | 4.0 ± 0.12d                 | 15.7 ± 1.98b            | 80.3 ± 2.07a               | 17.2 ± 0.78a               | 313.2 ± 45.03b              |
| 180                      | 38.5 ± 6.80a    | 7.5 ± 0.57c            | 4.7 ± 0.21d                 | 16.4 ± 0.91b            | 78.9 ± 0.95a               | 17.6 ± 0.66a               | 675.7 ± 56.12a              |

Means within a given column followed by different letters are significantly different by Tukey’s test (P≤0.05). ¹Moisture of dried biomass. ²Two harvests at 150-d intervals, i.e. only 300-days production.
Table 3. Average chemical composition (%) of Taiwan grass at 6 cutting frequencies.

| Cutting frequency (days) | Component (%) | Component (%) | Component (%) | Component (%) | Component (%) | Component (%) |
|-------------------------|---------------|---------------|---------------|---------------|---------------|---------------|
|                         | Extractives   | Holocellulose¹ | Cellulose¹     | Hemicellulose¹ | Lignin¹       | Crude protein |
| 30                      | 10.2 ± 2.09abc| 71.4 ± 3.51a  | 38.3 ± 0.91lb | 33.1 ± 4.22ab | 17.7 ± 0.94c  | 10.5 ± 0.71a  |
| 60                      | 14.0 ± 3.45a  | 72.2 ± 1.62a  | 37.7 ± 0.78b  | 34.6 ± 1.68a  | 16.8 ± 0.36c  | 6.4 ± 1.09b   |
| 90                      | 7.2 ± 1.41c   | 71.9 ± 1.49a  | 42.8 ± 0.90a  | 29.1 ± 1.54c  | 19.6 ± 0.18b  | 4.6 ± 0.18c   |
| 120                     | 13.8± 1.38a   | 74.4 ± 0.62a  | 42.7 ± 0.57a  | 31.7 ± 0.60abc| 19.8 ± 0.41b  | 3.5 ± 0.52cd  |
| 150                     | 11.6 ± 0.95ab | 72.2 ± 0.56a  | 43.2 ± 0.37a  | 29.0 ± 0.39c  | 21.1 ± 0.23a  | 2.7 ± 0.45d   |
| 180                     | 7.8 ± 1.82bc  | 71.4 ± 0.60a  | 41.4 ± 1.96a  | 30.0 ± 1.54bc | 21.0 ± 0.91a  | 3.5 ± 0.51c   |

Means within a given column followed by different letters are different by Tukey’s test (P≤0.05). ¹Values based on moisture and extractive free weight.

Table 4. Theoretical annual bioethanol yield from Taiwan grass at 6 cutting frequencies.

| Cutting frequency (days) | Component (%) | Component (%) | Component (%) | Component (%) |
|-------------------------|---------------|---------------|---------------|---------------|
|                         | Glucose       | Xylose        | Total         | L/ha/yr        |
| 30                      | 1,138 ± 317.6c| 758 ± 161.5c  | 1,896 ± 469.9d| 2,400 ± 594.8d|
| 60                      | 1,146 ± 212.4c| 829 ± 146.8c  | 1,975 ± 353.4d| 2,499 ± 447.4d|
| 90                      | 1,739 ± 168.8bc| 926 ± 41.0c  | 2,665 ± 203.6cd| 3,373 ± 257.7cd|
| 120                     | 2,341 ± 450.2b| 1,365 ± 237.9b| 3,706 ± 687.5b| 4,692 ± 870.3b|
| 150                     | 2,291 ± 314.7b| 1,214 ± 159.9b| 3,504 ± 473.9bc| 4,436 ± 599.9bc|
| 180                     | 4,010 ± 1,025.1a| 2,259 ± 366.2a| 6,270 ± 1,385a| 7,936 ± 1,754a|

Means within columns followed by different letters are significantly different by Tukey’s test (P≤0.05). ¹Two harvests at 150-day intervals, i.e. 300-days production.

Discussion

This study has demonstrated that Taiwan grass has considerable potential for biomass production, which can then be utilized to produce bioethanol. It is obvious that the longer the intervals between harvests the greater the biomass production per annum up to 180-day intervals, as longer intervals were not studied in this work. While bioethanol production also increased as interval between harvests increased, the increase in production was not as great as for biomass yields.

Biomass yield

The maximum biomass yield obtained of 38.5 t DM/ha at 180-day harvest intervals was somewhat less than the 46.3–58.4 t DM/ha/yr obtained by Ramos et al. (2013) with 3 cultivars of Cenchrus but slightly greater than the 11-25 t DM/ha/yr reported for other Cenchrus purpureus cultivars (Habte et al. 2020). These differences are not surprising as biomass yield is affected by genotype, soil properties including fertilization, age of the plant, agronomic management and amount and distribution of rainfall (Liu et al. 2014; Ventura et al. 2015). The increases in biomass yield as harvest interval increased are similar to those reported by Calzada et al. (2014).

Proximate analysis

Fixed carbon. Crude fiber (CF) in the grass from 90-day harvest of 4,010 and 2,259 kg/ha/yr from cellulose (glucose)and hemicellulose(xylose) sources, respectively. Maximum total theoretical bioethanol yield was 6,270 kg/ha/yr or 7,936 L/ha/yr.

Volatile matter. The concentrations of volatile matter obtained for the different cutting frequencies were lower than the values reported for 2 cultivars of Cenchrus...
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In plants (Sannigrahi et al. 2010). They function as interfere with carbohydrate and lignin characterization easily extractable with water or solvents that can chlorophyll, organic acids, alditols, and polyphenolics), of grass biomass (waxes, fats, oils, resins, free sugars, hemicellulose during the production process. hemicellulose can also be used to produce bioethanol. On the other hand, lignin is a complex polymer formed of glucose, xylose, galactose, mannose and glucuronic acid. Some wild type microorganisms have the metabolic capacity to use xylose and galactose to produce ethanol, while the glucose from hemicellulose can also be used to produce bioethanol. The distribution of cellulose in grasses is commonly 10% in leaves and 20–40% in stalks (Cafall and Mohnen 2009), so it was not surprising that cellulose concentration in biomass was higher at harvest intervals of 90 days than at 30- and 60-day intervals as stem percentage increases as grasses mature. In previous studies, Santiago et al. (2016) and Lima et al. (2014) reported 42.6% for Taiwan grass (at 270 days of age) and 43.4% for U. brizantha (at 180 days of age), which are similar to values obtained for the longer harvest interval suggests that this parameter will not be affected significantly by duration between harvests.

**Calorific value**

Higher heating value, also known as the gross calorific value or gross energy calorific value, is directly related to the potential of material for production of bioethanol and is an important characteristic for evaluating materials (Ramírez et al. 2012). It is the amount of heat released during the combustion of one gram of fuel to produce CO₂ and H₂O at its initial temperature and is usually used to define the energy content of fuels and thereby their efficiency (Godin et al. 2013). The higher heating values (14.9–16.5 MJ/kg DM) reported for 9 cultivars of C. purpureus (Ramos et al. 2013; Mohammed et al. 2015) were similar to the energy values obtained in this work (15.6–17.6 MJ/kg DM). In contrast, an energy value of 18 MJ/kg DM has been reported for Switchgrass (Panicum virgatum) (Ram and Salam 2012).

**Chemical composition**

The composition of biomass produced is important in considering a plant’s potential for bioethanol production as it influences the heating value and combustion processes (Brosse et al. 2012). For example, hemicellulose is a polymer with units of glucose, xylose, galactose, mannose and glucuronic acid. Some wild type microorganisms have the metabolic capacity to use xylose and galactose to produce ethanol, while the glucose from hemicellulose can also be used to produce bioethanol. On the other hand, lignin is a complex polymer formed by units of phenyl propane (p-coumaryl, coniferyl and syiapyl alcohol) and presents a problem in ethanol production as it can prevent the release of cellulose and hemicellulose during the production process.

**Extractives.** Extractives are non-structural compounds of grass biomass (waxes, fats, oils, resins, free sugars, chlorophyll, organic acids, alditols, and polyphenolics), easily extractable with water or solvents that can interfere with carbohydrate and lignin characterization in plants (Sannigrahi et al. 2010). They function as metabolic intermediaries and energy reserves and are responsible for the color, smell and resistance to wilting of grasses (Olanders and Steenari 1995). However, they cannot be converted to ethanol, so lignocellulosic biomass with a higher concentration of extractives will produce a lower yield of ethanol (Santiago et al. 2016). Gomes et al. (2015) suggest that biomass extractives present problems because they cause difficulties in the operation of industrial equipment through stickiness. In previous studies, Cardona et al. (2013) reported 16.9% for elephant grass (C. purpureus), while Mateus et al. (2012) reported 10.7% for Maralfalfa (C. purpureus), which is similar to the average obtained in the present study. The absence of any consistent relationship between level of extractives in the grass and harvest interval suggests that this parameter will not be affected significantly by duration between harvests.

**Holocellulose.** Holocellulose is formed by cellulose and hemicellulose (Jacobsen and Wyman 2000) and a higher concentration of holocellulose will produce higher amounts of bioethanol. However, pretreatments, hydrolysis and fermentation will directly determine the bioconversion of glucose and xylose to bioethanol (Victor et al. 2015). The values recorded in this study (71.4–74.4%) are similar to the values reported for Panicum maximum (now: Megathyrsus maximus) (69.9%) and Brachiaria brizantha (now: Urochloa brizantha) (71.7%) by Lima et al. (2014), as well as for elephant grass (C. purpureus) (72%) and C. purpureus cv. Enano (71.2%) by Wongwatanapaiboon et al. (2012). Since holocellulose concentrations in the biomass produced in our study were not related to harvest interval, frequency of harvests is unlikely to affect this parameter for the grass.

**Cellulose.** The distribution of cellulose in grasses is commonly 10% in leaves and 20–40% in stalks (Cafall and Mohnen 2009), so it was not surprising that cellulose concentration in biomass was higher at harvest intervals of 90 days than at 30- and 60-day intervals as stem percentage increases as grasses mature. In previous studies, Santiago et al. (2016) and Lima et al. (2014) reported 42.6% for Taiwan grass (at 270 days of age) and 43.4% for U. brizantha (at 180 days of age), which are similar to values obtained for the longer harvest intervals in our study. On the other hand, Rueda et al. (2016) registered concentrations of 37.7 and 36.7% for C. purpureus cv. Muaklek at 90 days of age, which is similar to concentrations for the 30- and 60-day harvests.

**Hemicellulose.** Hemicellulose is a complex carbohydrate polymer that constitutes 25–50% of the biomass in Gramineae (Ebringerová et al. 2005). The concentrations
we recorded are towards the bottom of this range and generally below the 37.6% recorded for elephant grass by Wongwatanapaiboon et al. (2012) but similar to the 31.0% reported for King grass (Cenchrus hybrid) by the same authors. Similarly, Lima et al. (2014) reported an average of 28% for C. purpureus. To produce bioethanol, hemicellulose concentration must be low because not all ethanol-producing microorganisms can metabolize xylose and galactose (hemicellulose-forming units). Increasing the harvest interval increased biomass production and reduced hemicellulose concentration, making the biomass obtained more suitable for bioethanol production.

Lignin. Lignin is a complex polymer constituted by units of phenyl propane (p-coumaryl, coniferyl and synapyl alcohol) and represents 10–30% of the total biomass in Gramineae (Limayem and Ricke 2012). While concentrations of lignin in biomass from our study increased as age at harvest increased as indicated by McCan and Carpita (2008), even at the longest harvest interval the concentration was lower than the 24% reported by Lima et al. (2014) for C. purpureus cultivars but greater than the 16.3% reported for Maralfalfa (C. purpureus) by Mateus et al. (2012). In terms of bioenergetics evaluations, high concentrations of lignin are undesirable, as the architecture and biochemistry of its bonds makes the hydrolysis of cellulose and hemicellulose difficult.

Crude protein. It is important to evaluate the concentration of CP in biomass because it can interfere with lignin quantification and change the chemical composition of biomass (Du et al. 2020). High concentrations of nitrogen limit the bioconversion of total sugars to ethanol (Santiago et al. 2016). The reduction in CP concentration, as stage of development at harvest increased, would favor bioethanol production from the material produced.

Bioethanol yield

The polysaccharides from cell walls can be used as raw matter to produce bioethanol and other biofuels once they have been pretreated and hydrolyzed into simple sugars for efficient fermentation (Rio Andrade et al. 2012). In the current study, potential yields of bioethanol were estimated according to the procedure of Badger (2002) and strongly favored the longest harvest interval, largely as a reflection of higher DM yields at this harvest frequency, despite increasing lignin concentration in the more mature material. Wongwatanapaiboon et al. (2012) reported bioethanol yields of 6,331 L/ha/yr for Mott grass (C. purpureus) and 6,717 L/ha/yr for guinea grass (M. maximus), both from rather aged biomass, which were 20 and 15% lower than the 7,936 L/ha/yr obtained in our study at 180-day harvest intervals.

It seems that Taiwan grass is a suitable source of biomass for production of second-generation bioethanol, with the potential to produce 8,000 L/ha/yr. It could compete with other first-generation primary sources like sugarcane juice and corn (grain) that can produce 6,900 L/ha and 2,900 L/ha, respectively (Somerville et al. 2010). Another reason for using C4 grasses for bioenergy is that they are more efficient in the use of water than C3 grasses (Weiide et al. 2013). These authors suggest that it is necessary to evaluate structural and non-structural components of the cell wall in order to produce bioethanol profitably and sustainably. Ethanol-producing microorganisms cannot convert 100% of fermentable sugars to ethanol, because they need to use part of these sugars to perform some other vital metabolic functions. Therefore, the theoretical yield based on 100 g of glucose that would produce 51.4 g ethanol and 48.8 g CO2 would not be possible (Badger 2002).

Current research is being undertaken by the United States Department of Energy with the objective of accelerating the conversion process from lignocellulosic biomass to liquid bioethanol. It is expected that by 2030 around 30% of the gasoline currently consumed worldwide will be replaced by bioethanol from plant material. A significant benefit would be that the use of bioethanol from cellulosic biomass could reduce the greenhouse gas emissions about 86% (Wang et al. 2007). Nevertheless, there is controversy about the environmental and economic benefits of biofuels. Science-based information will help to guide decisions about the crop, cultivation strategies, age of harvest and the bioethanol production process. Then, the environmental and economic impacts of biofuel production will become clearer. Furthermore, in terms of the balance of energy consumed:energy produced, cellulosic ethanol is less efficient than ethanol from starch and other traditional sugar crops. For example, for switchgrass and Miscanthus the ratios are 10.8–11.3:1 and 22:1, respectively. By comparison, the ratios for traditional sugar crops and corn are 8.1–10:1 and 1.4–2.3:1, respectively (Byrt et al. 2011). Obviously, starchy products are a much more efficient source of energy production than cellulosic materials. From an ethical point of view, it is not advisable to obtain fuel ethanol from raw materials considered as food for...
humans. For the production of ethanol from biomass to be economically and technically viable, the production process must be improved.

Conclusions

Climatic conditions, the type of soil in the Gulf of Mexico and the agronomic management developed in this study favor the growth of Taiwan grass. The highest yield of biomass was 38 t DM/ha/yr with a production of 675 GJ energy/ha at a harvest interval of 180 days. The plasticity, regrowth speed and resistance to pests and diseases make the grass an appropriate raw material to produce liquid biofuel in Mexico. Nevertheless, it is necessary to study the carbon footprint, life cycle analysis, energy yield (energy produced:energy consumed) of the entire production process to ensure there are positive effects on climate change and greenhouse gas emissions before this process is adopted.

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