

Water and radiation use efficiency of sugarcane for bioethanol production in South Africa, benchmarked against other selected crops

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There are indications that high-fibre sugarcane genotypes may produce more biomass and use resources more efficiently than conventional sugarcane cultivars. The objective of this research was to gather quantitative information on resource use for selected conventional and high-fibre sugarcane genotypes and benchmark it against other bioethanol crops. Although conventional sugarcane initially grew slower than sorghum and Napier grass, it produced very high biomass (about 70 t ha⁻¹) and theoretical ethanol (first- and second-generation) yields (about 27 kL ha⁻¹) at 12 months, and used water relatively efficiently (about 5 kg m⁻³ and 2 kL m⁻³), outperforming all other crops except sorghum. The contribution of cellulosic ethanol to total ethanol yield varied hugely, from 89% for the high-fibre sugarcane hybrid to about 48% for conventional sugarcane, to as low as 14% for sugar beet. The high-fibre sugarcane hybrid grew faster initially and produced more biomass at eight months (56 t ha⁻¹ vs. 45 t ha⁻¹) than the conventional types, but then flowered,

reducing its growth rates markedly thereafter. It was also less sensitive to mild drought conditions. Results suggest that cellulosic ethanol production could be a feasible option that could be incorporated into conventional or biomass sugarcane production systems.

Keywords: bioethanol crops, biomass, high-fibre sugarcane, stalk fibre composition, theoretical ethanol yield

Introduction

There is increasing interest in renewable energy, including biofuel from crops. Bioethanol can be produced from the fermentation of soluble sugars in the storage organs of feedstock crops, while 2nd generation lignocellulose technology enables the production of ethanol from cell-wall sugars extracted from plant fibre (Ragauskas et al. 2006). This will greatly enhance the potential ethanol output from feedstock crops and address concerns regarding ethanol production from food crops in high potential production areas.

Potential bioethanol crops include maize, switchgrass, Miscanthus, sugarcane, sugar beet, sorghum and poplar. Compared to other crops sugarcane has abundant potential for producing high biomass yield (Alexander, 1985) and consequently high bioethanol yields from sugars in the juice (Renouf et al. 2008) and from leaf and stalk fibre (Waclawovsky et al. 2010, de Souza et al. 2013). Energy cane that produce high biomass rather than high sucrose yield and use natural resources more efficiently, are currently in development (Tew and Cobill 2008). These genotypes

could possibly be used for biomass production in marginal production areas where resource levels are low, such as low rainfall areas or areas with poor soils.

Very little quantitative information on radiation and water use or crop productivity is available for high-fibre sugarcane types in South Africa. Waclawovsky et al. (2006) quote commercial maximum yields of 29 t ha^{-1} of dry biomass and puts forward a theoretical maximum of 177 t ha^{-1} . Alexander (1985) hypothesizes that sugarcane yields can be increased two fold by using high-fibre cane and managing water and nitrogen to maximize biomass growth and not sucrose yield. Biomass yields can also be increased by reducing the growing period from 12 to 6 months and harvesting twice a year (Alexander 1985) to better exploit the faster initial growth.

Sugarcane models have been calibrated for commercial sucrose cultivars (about 12% fibre and 13% sugar content) and will need refinement and re-calibration for high-fibre sugarcane hybrids (about 30% fibre and 5% sugar content), before they can be used to assess resource use efficiency and productivity (Keating et al. 1999, Singels and Bezuidenhout 2002). Nair et al. (2012) highlighted the need for parameterization and validation of crop models for bioenergy crops, which can then be used for high resolution simulation of biomass production for planning purposes.

The overall objective of this research was to gather quantitative information on the productivity (biomass and bioethanol), water and radiation use efficiencies and drought tolerance of conventional and high-fibre sugarcane genotypes and to benchmark these against other selected bioenergy feedstock crops. The information gathered could be valuable in improving the capability of crop models to support

decision-making regarding sugarcane production for bioenergy in existing and marginal production areas for current and future climates.

Materials and methods

Experimental details

A one hectare field trial was conducted at the South African Sugarcane Research Institute's Research Station near Komatipoort (25°33'10" S; 31°57'21" E, altitude 187 m above sea level) as a complete randomised block design with seven genotype treatments and four replications. The trial site consisted of two panels which were 21 m wide and 180 m long separated by a 5 m path along the length of the panel. Each panel comprised of 24 plots 7.5 m wide and 21 m long with no breaks between adjacent plots. The genotypes evaluated were two commercial sugarcane (complex hybrid of *Saccharum* spp.) cultivars (N19 and N31), a high-fibre sugarcane hybrid (04G0073, a cross between a conventional sugarcane clone and a *S. spontaneum* clone), an *Erianthus arundinaceus* clone (IK76-63) and three other crops, namely Napier grass (*Pennisetum purpureum* Land race), a forage sorghum hybrid (*Sorghum bicolor*, cultivar Big Kahuna) and sugar beet (*Beta vulgaris*, cultivar EB0809). N19 is commonly grown in irrigated regions of South Africa and is classified as having high sucrose content (McIntyre and Nuss 1996, SASRI 2006). Cultivar N31 is widely grown in rainfed regions of South Africa and is classified as high yielding with relatively low sucrose content (SASRI 2006). 04G0073 is a F1 hybrid cross between 88M0287 (complex hybrid of *Saccharum* spp. bred for sugar production) and US56158S (a *Saccharum spontaneum* clone) (pers. comm. M. Zhou, SASRI Plant Breeding Department, Mount Edgecombe). Selection of

04G0073 and IK76-63 was based on limited information available from SASRI plant breeding screening trials. Forage sorghum was selected above sweet sorghum as the focus was on high biomass yields.

Trial details are summarised in Table 1. Sorghum had to be replanted due to poor germination, which accounts for the six week difference in planting date compared to

Table 1: Cultivar, row spacing and plant and harvest dates of crops evaluated in the field trial

Crop	Species	Cultivar	Row spacing	Plant date	Harvest date
Sugarcane	<i>Saccharum spp.</i>	N31	1.5 m	12 Oct 2011	26 Oct 2012
Sugarcane	<i>Saccharum spp.</i>	N19	1.5 m	12 Oct 2011	26 Oct 2012
Sugarcane	<i>Saccharum spp.</i>	04G0073	1.5 m	12 Oct 2011	26 Oct 2012
Erianthus	<i>Erianthus arundinaceus</i>	IK76-63	1.5 m	12 Oct 2011	26 Oct 2012
Napier grass	<i>Pennisetum purpureum</i>	Landrace	1.5 m	12 Oct 2011	26 Oct 2012
Forage sorghum	<i>Sorghum bicolor</i>	Big Kahuna	0.75 m*	29 Nov 2011	26 Mar 2012
Sugar beet	<i>Beta vulgaris</i>	EB0809	0.75 m*	23 Apr 2012	26 Oct 2012

Forage sorghum planted at a recommended rate of 133 333 plants ha⁻¹ and sugar beet at 66 667 plants ha⁻¹.

the other crops. Growth and development was not affected negatively by the later planting as temperatures were well above the lower threshold values for sorghum during the entire growing period.

Based on soil analysis 120 kg N ha⁻¹ and 100 kg P ha⁻¹ was applied as Urea (46) and MAP (33) respectively to all plots one month after planting. After sorghum had been harvested the same plots were planted to sugar beet. Fertiliser was applied at a rate of 120 kg N ha⁻¹, 75 kg P ha⁻¹ and 75 kg K ha⁻¹ as Urea (46), MAP (33) and potassium chloride (KCL), respectively one month after the sugar beet was planted. Measurements of organic and mineral nitrogen and potassium in the soil on 22 August 2012 suggested that both these elements were not limiting growth in any of the treatments.

The soil was a shallow (0.60 m), well-drained, red sandy clay loam (clay content of 35%), classified as a Shortlands (Soil classification working group, 1991) with a profile field capacity (FC) value of 200 mm/0.6 m and permanent wilting point (PWP) value of 90 mm per 0.6 m. The available soil water content capacity (ASWC_{capacity}) for the profile was taken as 110 mm. The region is characterized by summer rainfall (long-term mean annual rainfall of 641 mm), hot summers and mild winters (see Figure 1).

Drip irrigation lines (emitters spaced at 0.6 m and delivery rate of 1.8 L h⁻¹) were spaced 1.5 m apart. Two water treatments were applied namely: (1) a well-watered control treatment (W100, irrigated to replace extraction on reaching a deficit of 10 mm) and (2) a water stress treatment (W50, receiving only 50% of the well-watered

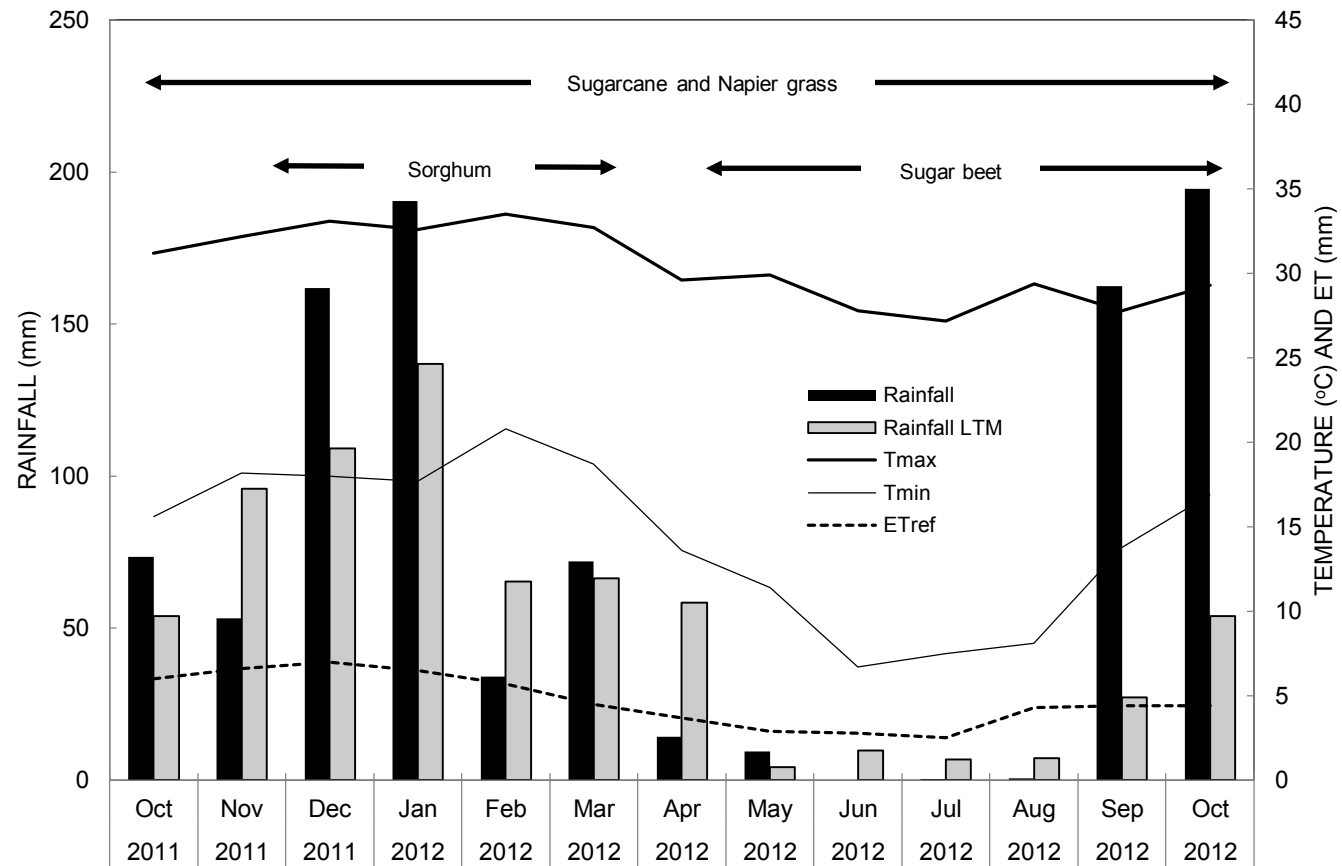


Figure 1: Rainfall, long-term mean rainfall (Rainfall LTM), monthly average minimum (Tmin) and maximum (Tmax) air temperature and monthly average reference cane evapotranspiration (ETref, as defined by McGlinchey and Inman-Bamber 1996) encountered during the growing season

treatment). Irrigation in the W100 control treatment was managed strictly according to the measured soil water content (SWC) records of individual plots, and SWC never dropped below the stress point (50% of $ASWC_{capacity}$). Whenever the W100 treatment was irrigated, only half the irrigation amount was applied to the corresponding genotype in the W50 treatment. During the latter part of the growing season the W50 treatments occasionally received additional irrigation as the soil water content approached PWP. Irrigation water was withdrawn on 3 October 2012 (24 d before harvest) and the crop allowed to dry off naturally. During this period a total of 137 mm of rainfall was however recorded.

Measurements

Volumetric soil water content

Volumetric SWC was measured in all 48 plots using a neutron water meter (Model 503DR CPN Hydro probe, Campbell Pacific Nuclear, Concord, CA, USA) calibrated for the specific soil. Measurements were taken before and after irrigation events at 0.25 m, 0.40 m and 0.55 m soil depth.

Non-destructive plant measurements

Fractional interception of photosynthetically active radiation (FI_{PAR}) was measured fortnightly using a ceptometer (Model AccuPar LP80, Decagon Devices, Pullman, WA, USA) in a marked 2 m section of cane row. Ten readings were taken below, and one above, the canopy between 11:00 and 13:00.

Destructive plant measurements

Destructive samples of aboveground biomass (2 m row length per plot) were taken at four, eight and 12 months of age in all four replicate plots. Plant material was split into stalk, green leaf (including cane tops) and dead leaf (trash) material. Fresh mass was determined and sub-samples dried at 85 °C (until constant mass).

Fibre, brix and sucrose content in stalks were determined from a sub-sample of shredded stalk material (16 stalks per plot). The shredded material was treated conventionally in a blender, filtered and the liquid portion passed through a polarimeter and a refractometer to determine pol and brix readings of the extract (Schoonees-Muir et al. 2009). Fibre%_{cane} was calculated from dry matter content (determined by weighing fresh and dry stalk material) and the Brix reading, brix%_{cane} was derived from the brix reading and fibre%_{cane}, while pol%_{cane} was calculated from the pol reading and fibre%_{cane} (SASRI 2013). Brix and sucrose mass was calculated from stalk fresh mass and brix and sucrose content, respectively. The difference between brix and sucrose mass was taken as the mass of hexoses (glucose and fructose) in stalks.

A second subsample of 16 stalks per plot of selected treatments (the W100 treatments for sugarcane, Napier grass, and sorghum) were analysed for stalk fibre composition. Not all treatments could be analysed because of the prohibitive cost of the process. A minimum of 8 kg of fresh plant stalk was washed, ground and dried for 48 h to obtain approximately 1 kg of dry fibre. The samples were treated and

analysed using the NREL methodology for their lignin, cellulose and hemicellulose contents (Sluiter et al. 2005a, Sluiter et al. 2005b, Sluiter et al. 2011). The concentrations of sugars that can be derived from cellulose and hemicellulose were determined with high performance liquid chromatography and include cell wall glucose, cell wall cellobiose, cell wall xylose, and cell wall arabinose.

Calculations

Crop water use

Seasonal crop water use (CWU) or evapotranspiration (ET) was estimated using the soil water balance equation:

$$\Sigma ET = \Delta S + \Sigma I + \Sigma R_{\text{eff}} - \Sigma DR \quad (1)$$

where ΔS is the change in storage (the difference in ASWC at the end and start of the experiment), ΣI and ΣR_{eff} are seasonal total irrigation and effective rainfall respectively and ΣDR is seasonal total drainage plus runoff. R_{eff} was calculated by assuming an interception loss per rainfall event equal to sugarcane reference evapotranspiration (ET_{ref} , as defined by McGlinchey and Inman-Bamber 1996) on the given day. ΣDR was calculated by summing individual daily drainage events (runoff was assumed to be zero as the fields were flat) for a season, with drainage for individual events calculated according to Equation 2:

$$DR = (ASWC + I + R_{\text{eff}} - ET) - ASWC_{\text{capacity}} \quad (2)$$

Water-stress days

Water-stressed days were defined as days during the active growing season (excluding the 24-day drying off period) when profile SWC was less than 145 mm (50% of plant available water capacity).

Thermal time

Thermal time (TT in units of °Cd) was calculated according to Equation 3:

$$TT = (T_{ave} - T_{base}) \cdot \Delta t \quad (3)$$

where T_{ave} is the average daily air temperature (calculated from the maximum and minimum air temperature), T_{base} the base temperature (16°C) and Δt the time interval. Thermal time was accumulated from the plant date up to 50% (TT50) and 80% PAR interception (TT80).

Radiation interception

Polynomial regression lines were fitted to the FI_{PAR} data to estimate FI_{PAR} for each day of the growing season. Estimated FI_{PAR} values were then converted to FI for global shortwave radiation (FI_{SR}) in a two-step approach using Beer's law. First daily leaf area index values were calculated, using a light extinction coefficient of $k_{PAR} = 0.77$, which was then converted back to FI_{SR} values using a light extinction coefficient of $k_{SR} = 0.55$. Daily intercepted radiation was calculated by multiplying daily FI_{SR} values with recorded daily global shortwave radiation values. The chosen

k_{SR} value agrees well with those reported in the literature for sugar beet (Rinaldi and Vonella 2006) and for sorghum (Ceotto et al. 2013) and thus applied to all crops.

Ethanol yield

The following equations (Zhao et al. 2009) were used to calculate the theoretical first-generation ethanol yield present in stalk juice (ETH_{SJ}), and second-generation ethanol yield from stalk (ETH_{SF}) and leaf fibre (ETH_{LF}), all in litres per hectare:

$$ETH_{SJ} = (SUC \cdot STKDM \cdot k_{c1} + GF \cdot STKDM \cdot k_{c2}) \cdot k_{ps} \cdot \rho \quad (4)$$

$$ETH_{SF} = SUG_{CW} \cdot STKDM \cdot k_{pc} \cdot k_{c2} \cdot k_{ps} \cdot \rho \quad (5)$$

$$ETH_{LF} = SUG_{LF} \cdot LFDM \cdot k_{pc} \cdot k_{c2} \cdot k_{ps} \cdot \rho \quad (6)$$

where SUG_{CW} is sugars from cell wall hemicellulose and cellulose in stalk dry matter (%), SUC is soluble sucrose in stalk dry matter (%), GF is soluble glucose and fructose in stalk dry matter (%), SUG_{LF} is sugars present in leaf dry matter (%), $STKDM$ is dry stalk biomass ($t \text{ ha}^{-1}$), $LFDM$ is dry dead and green leaf biomass ($t \text{ ha}^{-1}$), k_{pc} is process efficiency of sugar from cellulose and hemicellulose (0.85), k_c is the conversion efficiency of ethanol from sugar ($k_{c1} = 0.537$ for sucrose and $k_{c2} = 0.51$ for monosaccharide sugars), k_{ps} is process efficiency of ethanol from sugar (0.85) and ρ is the specific gravity of ethanol (1270 L t^{-1}). Measured SUG_{CW} was only available for the W100 treatments of sugarcane, Napier grass and sorghum, and these values were also applied to W50 treatments for ethanol calculations. For sugar beet we assumed a SUG_{CW} value of 66% based on Bertin et al. (1988). SUG_{LF} was not measured and a value of 57% was assumed for sugarcane and sorghum (Murray

et al. 2008, Krishan et al. 2010, Chandel et al. 2012). It should be noted that this approach to calculating ethanol yield is a very generalised one. Actual ethanol yield may differ significantly depending on the technology and species used.

Water use efficiency

Biomass (and bioethanol) water use efficiency (BWUE and EWUE) was defined as aboveground dry biomass (and theoretical first- and second-generation ethanol volume) produced per unit of evapotranspiration (kg m^{-3} and L m^{-3}).

Radiation use efficiency

Biomass (and bioethanol) radiation use efficiency (BRUE and ERUE) was defined as aboveground dry biomass (and theoretical first- and second-generation ethanol volume) produced per unit of intercepted radiation (g MJ^{-1} and mL MJ^{-1}).

Drought sensitivity

Yield response factors (K_y) were calculated according to Doorenbos and Kassam (1979) for each of the crops as a means to quantify drought sensitivity.

$$K_y = [(Y_{\text{pot}} - Y_{\text{act}}) / Y_{\text{pot}}] / [(ET_{\text{pot}} - ET_{\text{act}}) / ET_{\text{pot}}] \quad (7)$$

where Y and ET refer to aboveground dry biomass yield (t ha^{-1}) and evapotranspiration (mm) at harvest, and subscripts *act* and *pot* refer to values for the W50 and W100 treatments, respectively.

Data were analysed using two-way analysis of variance (ANOVA) with GenStat 14th Edition (VSN International, Hemel, Hempstead) where possible (stalk fibre fraction, sucrose fraction, hexose fraction, green leaf fraction, dead leaf fraction and dry biomass yield). The ANOVA for biomass- and bioethanol WUE was based on replicated cane yield data only, since replicated CWU data was not available. Statistical significance of main and interaction effects were calculated for the 5% ($P \leq 0.05$) and 1% ($P \leq 0.01$) confidence levels and least significant differences (LSD) determined for $P \leq 0.05$. ANOVA could not be conducted on TT50 and TT80 PAR interception as well as seasonal average FI_{PAR} and radiation capture data because the data were based on curves fitted to average values. High costs associated with stalk fibre component analysis prevented any ANOVA being conducted on this and consequently also the theoretical ethanol yield data.

Results and discussion

Climate

Climatic conditions during the experiment are shown in Figure 1. The summer rainfall region is characterised by very hot summers and mild winters with a seasonal total ET_{ref} equal to 1779 mm, while rainfall was abnormally high during December 2011 and January 2012 (148% and 139% of long-term mean, respectively) as well as September and October 2012 (598% and 360% of the long-term mean,

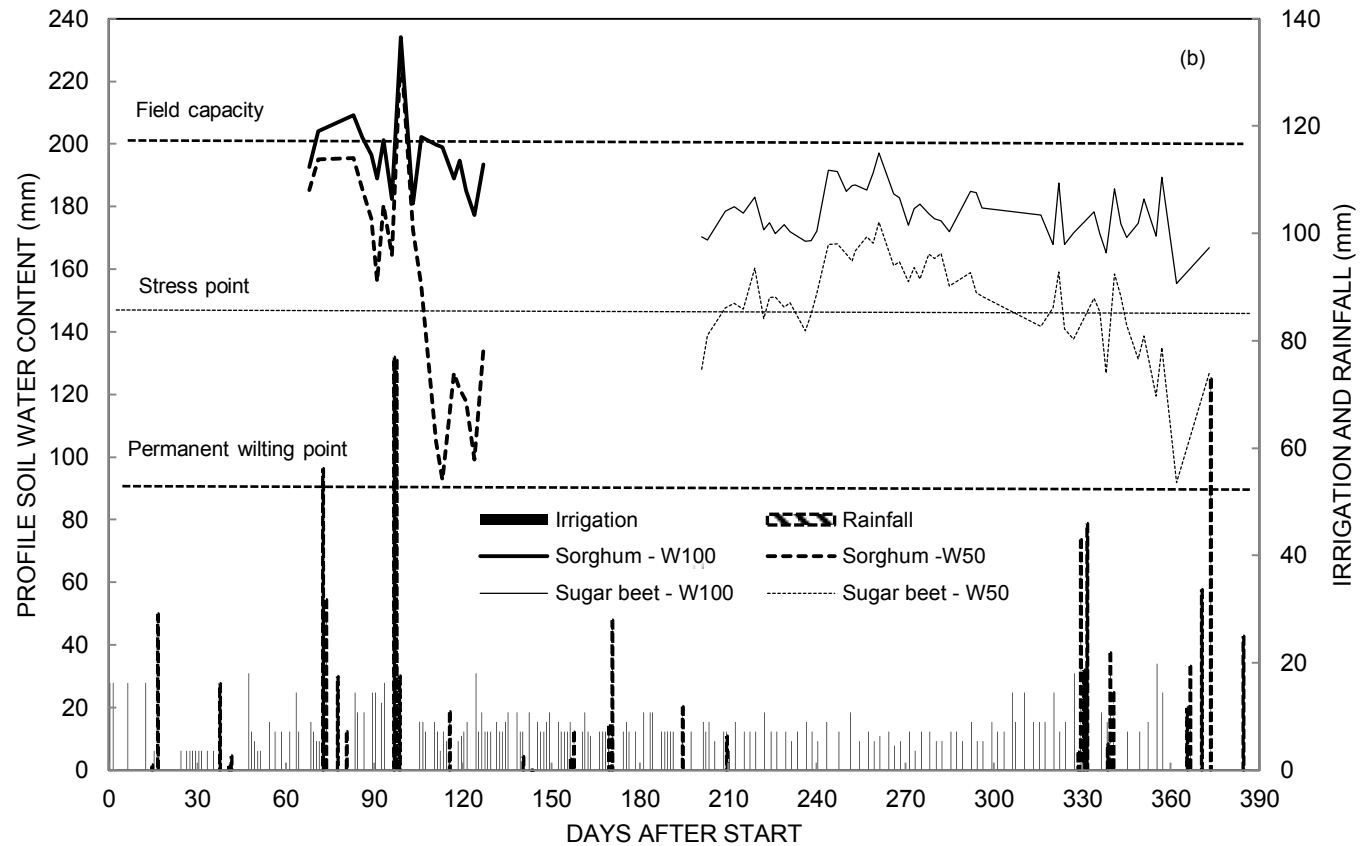


Figure 2: Daily water balance of the well-watered (W100, solid lines) and the water stress (W50, dotted lines) treatments of selected genotypes, a) N19 and 04G0073 and b) Sorghum and Sugar beet. Rainfall and irrigation events are indicated as hashed and solid bars, respectively. Stress point was taken as 50% of the plant- available water capacity

respectively). Total rainfall during the growing season (October 2011 to October 2012) was 888 mm compared with a long term mean of 641 mm.

Soil water status

Well-watered treatments (W100) were kept within the target range without exposing the crop to any water stress (Figure 2). Due to the high rainfall recorded during the first part of the growing season (October 2011 to January 2012), stress could only be imposed in the W50 treatments from the end of January 2012 onwards. Water stress was relieved by rainfall in March 2012 and again at the end of the season (September and October 2012). In total, 04G0073 endured 221 stress days, followed by IK76-63, N31, N19 and Napier grass with 214, 189, 180 and 157 stress days respectively. Sorghum experienced only 19 stress days which occurred just before flowering. Sugar beet endured 44 stress days which also occurred in the latter part of the growing season.

Canopy cover

In the W100 treatment, sugar beet and sorghum had the quickest canopy development, requiring 426 and 509 °Cd less thermal time, respectively to reach 50% PAR interception (TT50) than conventional sugarcane (average TT50 value of 701°Cd) (Table 2). The canopy development rate of the high-fibre sugarcane hybrid (04G0073) was slightly faster than that of conventional sugarcane, while that of IK76-63 required an additional 206°Cd to reach 50% canopy due to its very slow germination (Table 2). Napier grass and sorghum achieved slightly higher seasonal

average FI than conventional sugarcane, because of their quicker canopy development. Radiation capture by 04G0073 and Napier grass over the first six months of growth was substantially higher than that of sugarcane, indicating that these crops are more efficient in radiation capture when harvested at six months rather than 12 months.

Table 2: Thermal time to 50% (TT50) and 80% (TT80) PAR interception (time in days given in parentheses), seasonal average fractional interception (FI_{PAR}) and radiation captured for the well-watered (W100) and water stress (W50) treatments

Variable	Irrigation treatment	Crop and Cultivar						
		Sugar-cane	Sugar-cane	Sugarcane hybrid	Erianthus	Napier grass	Sorghum	Sugar beet
		N31	N19	04G0073	IK76-63	Landrace	Big Kahuna	EB0809
TT50 (°Cd)	W100	696 (79)	705 (82)	688 (81)	906 (108)	482 (60)	275 (33)	192 (54)
	W50	716 (83)	716 (86)	723 (85)	941 (115)	491 (65)	295 (35)	197 (61)
TT80 (°Cd)	W100	898 (96)	998 (108)	916 (97)	1228 (129)	716 (77)	395 (40)	228 (65)
	W50	914 (98)	1015 (110)	998 (108)	1305 (136)	847 (91)	416 (42)	283 (76)
FI (%)	W100	80.9	77.9	79.9	73.5	82.5	83.9	72.9
	W50	78.8	77.2	78.4	71.6	80.3	83.0	66.6
Radiation captured (MJ m ⁻²)	W100	4388	4087	4290	3861	4526	2168	1629
	W50	4168	4020	4164	3716	4283	2149	1438

The water stress experienced in the experiment had little effect on canopy development of all the genotypes as there was no water stress during the first 110 d of the experiment. As a result, TT50 values of the W50 treatments were only reduced by between 5 °Cd (sugar beet) and 35 °Cd (IK76-63) relative to the W100 control. Averaged across all crops, water stress reduced the seasonal average fractional interception by 3% and radiation capture by 4% (Table 2).

Biomass yield

The potential of conventional sugarcane as a biomass crop was illustrated by the very high dry biomass yields obtained in N19 and N31 in both the W100 and W50 treatments at 12 months (Table 3). Unexpectedly, the high-fibre sugarcane hybrid 04G0073 and *Erianthus* clone IK76-63 produced significantly ($P < 0.001$) less dry biomass (17% and 14% respectively) than the conventional sugarcane cultivars N19 and N31 in the W100 treatment; however, growth was affected by flowering in 04G0073 and very poor germination in IK76-63.

Conventional sugarcane also outperformed the combination of sorghum and sugar beet, which was only able to produce 47 t ha⁻¹ dry material in 12 months (Table 3). Napier grass (63 t ha⁻¹) was the only crop that could compete with conventional sugarcane in terms of total dry biomass produced.

Water stress in the W50 treatment significantly reduced ($P < 0.001$) dry biomass yields of all the crops, except sorghum (Table 3).

At eight months 04G0073 and Napier grass were able to achieve higher biomass yields than conventional sugarcane (data not presented). This reflects the relative slow initial growth and establishment of sugarcane, a phenomenon also highlighted by Alexander (1985) and Allison et al. (2007). Once the canopy and stalk population is established, conventional sugarcane varieties N19 and N31 were able to maintain higher growth rates, partly due to the fact that they were not limited by flowering and self-trashing as with 04G0073 and Napier grass. Production strategies of high-fibre types could possibly be adjusted by harvesting at a younger age to maximise production per unit time.

Dry biomass components fractions (stalk fibre, sucrose and hexose from stalk juice; green and dead leaf) differed significantly ($P < 0.001$) between the crops that were evaluated (Table 3). Napier grass, 04G0073 and IK76-63 for example had the highest fibre contents and N19, sorghum and sugar beet had the highest sucrose contents (Table 3). Biomass component fractions were not significantly affected by water stress.

Stalk fibre component fractions are provided in Table 4. Cellulose and hemi-cellulose contents were mostly very similar for the different crops (about 35% and 30%, respectively), although IK76-63 appeared to have higher cellulose and lower hemi-cellulose content than the other crops. Conventional sugarcane and sorghum had lower lignin content than the other crops (about 21% compared to about 25%), making it more suitable for second-generation ethanol production. Stalk fibre composition values of the sugarcane crop compared favourably with typical values

Table 3: Biomass component fractions (stalk fibre; sucrose and hexose from stalk juice; green and dead leaf) and total above-ground dry biomass yield at harvest for the well-watered (W100) and water stress (W50) treatments. For statistical significance see footnote *

Variable	Irrigation treatment	Crop and Cultivar							Average
		Sugar-cane	Sugar-cane	Sugarcane hybrid	Erianthus	Napier grass	Sorghum	Sugar beet	
		N31	N19	04G0073	IK76-63	Landrace	Big Kahuna	EB0809	
Stalk fibre fraction	W100	0.27	0.29	0.37	0.47	0.48	0.24	0.08	0.31
	W50	0.34	0.25	0.36	0.48	0.45	0.23	0.08	0.31
	Mean	0.31	0.27	0.36	0.47	0.46	0.24	0.08	0.31
Sucrose fraction	W100	0.29	0.33	0.11	0.02	0.02	0.37	0.59	0.25
	W50	0.24	0.36	0.11	0.02	0.03	0.37	0.61	0.25
	Mean	0.27	0.35	0.11	0.02	0.03	0.37	0.60	0.25
Hexose fraction	W100	0.06	0.04	0.06	0.04	0.06	0.09	0.11	0.06
	W50	0.04	0.04	0.06	0.05	0.06	0.09	0.11	0.06
	Mean	0.05	0.04	0.06	0.05	0.06	0.09	0.11	0.06
Green leaf fraction	W100	0.12	0.18	0.18	0.25	0.16	0.20	0.25	0.19
	W50	0.14	0.15	0.19	0.25	0.18	0.22	0.22	0.19
	Mean	0.13	0.17	0.19	0.25	0.17	0.21	0.24	0.19
Dead leaf fraction	W100	0.26	0.16	0.27	0.21	0.28	0.09	-	0.21
	W50	0.23	0.19	0.28	0.21	0.29	0.08	-	0.21
	Mean	0.25	0.18	0.28	0.21	0.29	0.09		0.21
Dry biomass yield (t ha ⁻¹)	W100	68.3	76.1	59.6	61.8	63.1	23.8	23.1	53.6
	W50	58.7	65.9	56.3	51.8	49.4	24.7	18.4	46.4
	Mean	63.5	71.0	57.9	56.8	56.2	24.2	20.7	50.0

*Stalk fibre fraction, sucrose fraction, hexose fraction, green leaf fraction and dead leaf fraction: crop effects were highly significant ($P < 0.001$) with least significant difference (LSD) values of 0.04, 0.03, 0.01, 0.03 and 0.02, respectively. Dry biomass yield: crop and irrigation effects were highly significant ($P < 0.001$) with LSD values of 5.46 and 2.92 t ha⁻¹, respectively

reported in the literature. Chandel et al. (2012) reported typical ranges for cellulose, hemicellulose and lignin contents of sugarcane bagasse of 38-45%, 26-36% and 11-25%, respectively. Corresponding values reported by O'Shea et al. (2013) were 34-43%, 21-27% and 17-22% respectively, while Rao (1997) as quoted by Seebaluck and Sobhanbabu (2012), reported values of 26-43%, 17-23% and 13-22%, respectively. Variation is ascribed to genetic and environmental factors as well as crop development stage (Chandel et al. 2012).

Table 4: Stalk fibre component fractions at harvest for the well-watered treatment of the different crops

Variable	Crop and Cultivar					
	Sugar- cane	Sugar- cane	Sugarcane hybrid	Erianthus	Napier grass	Sorghum
	N31	N19	04G0073	IK76-63	Landrace	Big Kahuna
Lignin	0.212	0.207	0.241	0.261	0.267	0.215
Hemi-cellulose	0.316	0.316	0.308	0.273	0.297	0.299
Cellulose	0.366	0.346	0.371	0.403	0.362	0.348
Ash extractives	0.104	0.129	0.078	0.061	0.072	0.135

Ethanol yield

The highest theoretical ethanol yield was obtained for N19 (29.6 kL ha⁻¹), which was 32% and 36% more than that for 04G0073 or IK76-63, respectively. In 04G0073 and IK76-63, sucrose contributed very little (18% and 4%) to total ethanol yield compared with 48% and 43% in N19 and N31, respectively (Table 5). The combination of sorghum and sugar beet yielded only 19.2 kL ha⁻¹ of ethanol. Ethanol yields were reduced under water stress conditions in proportion to the reduction in biomass yield.

Table 5: Theoretical first-generation (produced from stalk sucrose and hexose), second-generation (produced from stalk fibre (lignin and cellulose) and leaf fibre (green and dead)) and total ethanol yield for each treatment

Variable	Irrigation treatment	Crop and Cultivar						
		Sugar-cane	Sugar-cane	Sugarcane hybrid	Erianthus	Napier grass	Sorghum	Sugar beet
		N31	N19	04G0073	IK76-63	Landrace	Big Kahuna	EB0809
Sucrose	W100	11.36	14.16	3.66	0.70	0.79	1.19	7.91
(kL ha ⁻¹)	W50	8.18	13.66	3.60	0.61	0.73	1.32	6.64
Hexose	W100	2.09	1.58	1.99	1.36	2.09	1.11	1.40
(kL ha ⁻¹)	W50	1.34	1.58	1.81	1.28	1.62	1.25	1.14
Total first-generation	W100	13.45	15.74	5.65	2.06	2.88	2.30	9.31
(kL ha ⁻¹)	W50	9.52	15.24	5.41	1.89	2.35	2.57	7.78
Stalk fibre	W100	5.95	6.91	7.12	9.21	9.33	3.81	0.36
(kl ha ⁻¹)	W50	6.39	5.18	6.37	7.83	6.88	3.84	0.18
Leaf fibre	W100	7.00	6.99	7.24	7.73	7.39	1.90	1.52
(kL ha ⁻¹)	W50	5.90	6.08	7.13	6.29	6.11	1.99	1.11
Total second-generation	W100	12.95	13.90	14.36	16.94	16.72	5.71	1.89
(kL ha ⁻¹)	W50	12.29	11.26	13.50	14.12	12.99	5.83	1.29
Total ethanol yield	W100	26.4	29.6	20.0	19.0	19.6	8.0	11.2
(kL ha ⁻¹)	W50	21.8	26.5	18.9	16.0	15.3	8.4	9.0

For conventional sugarcane cultivars second-generation ethanol yield contributed roughly half of total ethanol yield, with a 50:50 split between ethanol from bagasse

and leaf material. The contribution of second-generation ethanol to total ethanol in the high-fibre crops was expectedly higher, while that for sugar beet was much lower, than that of conventional sugarcane.

Chandel et al. (2012) and Hatti-Kaul et al. (2007) reported maximum ethanol yields of 300 L t⁻¹ of sugarcane bagasse, which corresponds to the ethanol yields from sugarcane stalk fibre calculated in this study. Chong and O'Shea (2012), however found that actual ethanol yield can vary from 110 to 270 L t⁻¹, depending on type of fermentation process and reaction conditions. De Souza et al. (2013) reported typical 1st generation ethanol yields from sugarcane, sorghum and sugar beet of 6.5, 1.4 and 5.4 kL ha⁻¹ respectively. These yields are much lower (about half) than achieved in our study, presumably because of much lower sugar yields due to less favourable growing conditions. De Souza et al. (2013) also reported second-generation ethanol yield for *Miscanthus* of up to 12.4 kL ha⁻¹, which compares well with the yield of Napier grass in our study. Zhao et al. (2009) reported that theoretical ethanol yields from sweet sorghum varied from 4.5 to 5.4 kL ha⁻¹ from sugar, and from 4.5 to 6.5 kL ha⁻¹ from cellulosic material, with totals varying from 4.8 to 11.4 kL ha⁻¹, depending on genotypes. Mengistu et al. (2013) reported sugar beet yields of up to 53 tons of fresh tubers achieved in six months near Pietermaritzburg, with an ethanol yield from sugar of 4.0 kL ha⁻¹. Maximum dry stalk mass for sweet sorghum varied between 9.4 and 10.4 t ha⁻¹ with first-generation ethanol yields of between 2.3 and 2.6 kL ha⁻¹ for the Pietermaritzburg and Pretoria trial sites of the same study (Mengistu et al. 2013). In the present study first-generation ethanol yield for forage sorghum was approximately 2.5 kL ha⁻¹, even though it had much lower sugar content in the stalk compared to sweet sorghum in the study of Mengistu et al. (2013).

Seasonal crop water use

The high-fibre sugarcane hybrid (04G0073) and *Erianthus* clone (IK76-63) used slightly more water (204 mm and 70 mm, respectively) than the conventional sugarcane cultivars N31 and N19 (with average water use of 1425 mm), while Napier grass used about 241 mm less water (Table 6). Due to their relatively short growing seasons (117 and 186 days, respectively) the combination of sorghum and sugar beet used substantially less water (461 mm) than conventional sugarcane. Crops in the W50 treatment used on average 28% less water compared to the W100 treatments (Table 6).

Water use efficiency

N19 and sorghum had significantly ($P < 0.001$) higher biomass and ethanol water use efficiency (BWUE = 6.3 kg m^{-3} and EWUE = 2.5 L m^{-3}) and (6.4 kg m^{-3} and 2.2 L m^{-3}), respectively (Table 6) compared to the other crops. Both also outperformed a combination of sorghum and sugar beet (5.1 kg m^{-3} and 2.0 L m^{-3}). Water use efficiency (BWUE and EWUE) of the commercial sugarcane cultivars were on average 33% and 28% higher than that of the high-fibre sugarcane hybrid 04G0073 and *Erianthus* clone IK76-63, respectively (Table 6). These results are in agreement with reported values of BWUE for sugarcane (assuming a dry matter content of 25% and stalk fraction of 0.75) that varied between 2.6 and 5.3 kg m^{-3} (Kingston 1994; Olivier and Singels 2012). For sugar beet, BWUE reported values were between 4.6 and 5.6 kg m^{-3} (Brown et al. 1987), but can be as high as 10 kg m^{-3} (Dunham 1983). High BWUE values obtained by sorghum were in the range of those commonly reported for sorghum that ranged between 2.8 and 12.6 kg m^{-3} (Narayanan et al.

2013). Water stress resulted in an average increase of 19% in BWUE and EWUE (Table 6). Rinaldi and Vonella (2006) found that BWUE was increased by 21% and 24% for spring and autumn sown sugar beet when irrigation was reduced.

Table 6: Water use efficiency for biomass (BWUE) and biofuel (EWUE, first- and second-generation) for the well-watered (W100) and water stress (W50) treatments. Crop water use (CWU) for treatments are also shown. For statistical significance see footnote *

Variable	Irrigation treatment	Crop and Cultivar							Average
		Sugar-cane	Sugar-cane	Sugarcane hybrid	Erianthus	Napier grass	Sorghum	Sugar beet	
		N31	N19	04G0073	IK76-63	Landrace	Big Kahuna	EB0809	
CWU	W100	1554	1296	1629	1495	1184	385	579	1160
(mm)	W50	975	962	1038	1110	962	371	405	832
BWUE	W100	4.39	5.85	3.65	4.13	5.33	6.19	3.99	4.79
(kg m ⁻³)	W50	6.02	6.86	5.42	4.66	5.14	6.67	4.53	5.61
	Mean	5.21	6.36	4.54	4.40	5.24	6.43	4.26	5.21
EWUEfirst-generation	W100	0.87	1.22	0.35	0.14	0.24	0.60	1.61	0.72
(L m ⁻³)	W50	0.98	1.59	0.52	0.17	0.25	0.70	1.93	0.88
EWUE	W100	0.83	1.07	0.88	1.13	1.41	1.49	0.33	1.02
second-generation	W50	1.26	1.17	1.30	1.27	1.35	1.58	0.32	1.18
(L m ⁻³)									
EWUE total	W100	1.70	2.29	1.23	1.27	1.66	2.09	1.94	1.74
(L m ⁻³)	W50	2.24	2.76	1.82	1.44	1.60	2.27	2.25	2.05
	Mean	1.97	2.53	1.53	1.36	1.63	2.18	2.10	1.90

*Biomass water use efficiency: crop and irrigation effects were highly significant ($P < 0.001$) with least significant difference (LSD) values of 0.70 and 0.37, respectively

Conventional sugarcane had higher first-generation EWUE (EWUE1) than high-fibre sugarcane, Napier grass and sorghum, as can be expected because of higher sugar contents in the stalk juice. Sugar beet had the highest EWUE1 because of the very high sugar content of the tubers. Conversely, the high-fibre crops had the highest second-generation EWUE (EWUE2) because of their higher fibre yields. EWUE of the water stress treatments were higher than well-watered treatments, following the trends in BWUE, because the primary driver of total ethanol yield is biomass.

Maximum EWUE1 values reported by Mengistu et al. (2013) for sorghum and sugar beet were lower (0.47 and 0.72 L m⁻³ respectively) than values shown in Table 6 for this study. This difference is attributed to the higher dry matter yields obtained in the current study.

Radiation use efficiency

N19 had significantly ($P < 0.001$) higher biomass and ethanol radiation use efficiency (BRUE, ERUE) compared to the other crops (1.75 g MJ⁻¹ and 0.69 mL MJ⁻¹), which was more than the sorghum and sugar beet combination (1.23 g MJ⁻¹ and 0.53 mL MJ⁻¹), respectively (Table 7). Sinclair and Muchow (1999) suggested a RUE for sugarcane of 2 g MJ⁻¹, although there is sufficient evidence that RUE varies considerably because of variation in temperature, soil fertility (leaf nitrogen content), crop age, crop class, lodging, and culm death (Muchow et al. 1994, Robertson et al. 1996, Park et al. 2005, Donaldson et al. 2008).

Table 7: Radiation use efficiency for biomass (BRUE) and biofuel (ERUE first- and second-generation) for the well-watered (W100) and water stress (W50) treatment. For statistical significance see footnote *

Variable	Irrigation treatment	Crop and Cultivar							Average
		Sugar-cane	Sugar-cane	Sugarcane hybrid	Erianthus	Napier grass	Sorghum	Sugar beet	
		N31	N19	04G0073	IK76-63	Landrace	Big Kahuna	EB0809	
BRUE	W100	1.55	1.85	1.38	1.60	1.39	1.09	1.41	1.47
(g MJ ⁻¹)	W50	1.40	1.64	1.35	1.39	1.15	1.15	1.27	1.34
	Mean	1.48	1.75	1.37	1.50	1.27	1.12	1.34	1.40
ERUE first-generation	W100	0.31	0.38	0.13	0.05	0.06	0.11	0.57	0.23
(mL MJ ⁻¹)	W50	0.23	0.37	0.13	0.05	0.05	0.12	0.54	0.21
ERUE second-generation	W100	0.29	0.34	0.33	0.44	0.37	0.26	0.11	0.31
(mL MJ ⁻¹)	W50	0.29	0.28	0.32	0.38	0.30	0.27	0.09	0.28
ERUE total	W100	0.60	0.72	0.46	0.49	0.43	0.37	0.68	0.54
(mL MJ ⁻¹)	W50	0.52	0.65	0.45	0.43	0.35	0.39	0.63	0.49
	Mean	0.56	0.69	0.46	0.46	0.39	0.38	0.67	0.51

*Biomass radiation use efficiency: crop and irrigation effects were highly significant ($P < 0.001$) with least significant difference (LSD) values of 0.17 and 0.09, respectively

Sugar beet is considered a crop that is relatively efficient in transforming solar radiation into dry matter, higher than durum wheat, but lower than maize and sorghum (Tanner and Sinclair 1983). Variable values are reported for sugar beet, varying from 1.66 g MJ⁻¹ in irrigated and 1.44 g MJ⁻¹ in non-irrigated conditions (Olivier and Singels 2012) but up to larger values ranging from 2.96 g MJ⁻¹ to 3.76 g

MJ⁻¹ (Damay and Le Gouis 1993). RUE values for sorghum are within the range of published values that vary from 1.2 to 4.3 g MJ⁻¹ (Muchow 1989, Rosenthal et al. 1993). Water stress resulted in a significant ($P < 0.001$) reduction in RUE (average of 8%), except for 04G0073 and sorghum (Table 7).

Drought sensitivity

Under conditions of water stress, Napier grass experienced a greater yield loss than any of the other crops due to its high yield response factor of 1.16. The hybrid 04G0073 was least sensitive to stress ($k_y = 0.15$) followed by N19, N31, IK76-63 and sugar beet (k_y of 0.38, 0.50, 0.63 and 0.69, respectively). The sorghum crop experienced very little, if any, water stress, because of high rainfall that occurred early during the relatively short growing season. Sorghum is well known for its drought tolerance and is widely grown in hot and dry subtropical and tropical regions of the world (Rooney et al. 2007).

Conclusion

Data collected in this study may only apply to situations with similar soil and climatic conditions, for the genotypes tested, and for plant crops. In a broad sense, however, some of the information gathered could be applicable to additional climate-soil situations, and therefore contributes to the body of information required for planning bioethanol feedstock production strategies.

Key findings

Although conventional sugarcane initially grew slower than sorghum and Napier grass, it produced very high biomass (about 70 t ha⁻¹) and theoretical ethanol yields (about 27 kL ha⁻¹) at 12 months. It also outperformed the combination of sorghum and sugar beet. This trend was also applicable for the water deficit treatments.

Biomass and ethanol water use efficiency were also relatively high for the conventional sugarcane cultivars (about 6 kg m⁻³ and 2 kL m⁻³), outperforming all other crops except sorghum. Their suitability for cultivation in marginal areas has yet to be tested thoroughly though, with annual crop water use in the stress treatments varying from 975 to 1110 mm. Marginal areas, with rainfall of less than 800 mm, will have much lower water use and more severe water stress than what was endured in this experiment. Assuming a rainfall efficiency of 70%, a BWUE of 5 kg m⁻³, a EWUE of 2.25 kL m⁻³ and a EWUE2 of 1.2 kL m⁻³ (Table 6), this implies estimated biomass and ethanol yields of 28 t ha⁻¹ and 12.6 kL ha⁻¹, with second-generation ethanol yields of 6.7 kL ha⁻¹, assuming optimal process efficiencies.

The high-fibre sugarcane hybrid showed promise in terms of growing faster initially and producing more biomass at eight months (56 t ha⁻¹ vs. 45 t ha⁻¹) than the conventional sugarcane cultivars, but then flowered, reducing its growth rate markedly thereafter. It also seemed less sensitive to mild drought conditions than the conventional sugarcane cultivars. This particular genotype may not have been the ideal selection to represent type II sugarcane, and further research is recommended to explore high-fibre germplasm for higher biomass yields and more efficient use of water.

Theoretical (assuming optimal process efficiencies) ethanol yields achieved in this study was very high (29 kL ha^{-1} for well-watered N19) compared to previously quoted values, primarily because of the high biomass achieved. Although there were marked differences in biomass component fractions (mainly stalk sucrose and stalk fibre), ethanol yields of the different treatments were largely determined by biomass yield, explaining why conventional sugarcane types out-yielded other crops in terms of ethanol yield. Stalk fibre composition varied little between the grass crops, although lignin content was significantly lower in conventional sugarcane and sorghum (21%), than in the other crops (26%). This had little influence on theoretical second-generation ethanol yields. The contribution of cellulosic ethanol to total ethanol yield varied hugely, from 89% for the high-fibre sugarcane hybrid to about 48% for conventional sugarcane, to as low as 14% for sugar beet. Results suggest that cellulosic ethanol production could be an attractive option that could be incorporated into conventional or biomass sugarcane production systems, particularly because of the comparative ease of cultivation and processing versatility of sugarcane.

The information gathered in this study is useful for deriving important crop model parameters (e.g. thermal time requirements for crop development, water and radiation conversion coefficients, drought sensitivity factors and biomass partitioning fractions) that are required for accurate prediction of yield and water use of conventional and high-fibre sugarcane types.

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References

- Alexander A G. 1985. *The energy cane alternative*. Amsterdam: Elsevier.
- Allison JCS, Pammenter NW, Haslam RJ. 2007. Why does sugarcane (*Saccharum sp.* hybrid) grow slowly? *South African Journal of Botany* 73: 546-551.
- Bertin C, Rouau X, Thibault JF. 1988. Structure and properties of sugar beet fibres. *Journal of the Science of Food and Agriculture* 44: 15-29.
- Brown KF, Messemer AB, Dunham RJ, Biscoe PV. 1987. Effect of drought on growth and water use of sugar beet. *Journal of Agricultural Science* 109: 421-435.
- Ceotto E, Di Candiloa M, Castelli F, Badeckc FW, Rizzad F, Soavee G, Voltaf A, Villani G, Marlettoh V. 2013. Comparing solar radiation interception and use efficiency for the energy crops giant reed (*Arundo donax L.*) and sweet sorghum (*Sorghum bicolor L. Moench*). *Field Crops Research* 149: 159–166.
- Chandel AK, da Silva SS, Carvalho W, Singh OV. 2012. Sugarcane bagasse and leaves: Foreseeable biomass of biofuel and bio-products. *Journal of Chemical Technology and Biotechnology* 87: 11- 20.
- Chong BF, O’Shea MG. 2012. Developing sugarcane lignocellulosic biorefineries: opportunities and challenges. *Biofuels* 3: 307- 319

- Damay N, Le Gouis J. 1993. Radiation use efficiency of sugar beet in Northern France. *European Journal of Agronomy* 2: 179 -184.
- de Souza AP, Grandis A, Leite DCC, Buckeridge MS. 2013. Sugarcane as a Bioenergy Source: History, Performance, and Perspectives for Second-Generation. *Bioethanol Bioenergy Research* Published online, doi:10.1007/s12155-01309366-8.
- Donaldson RA, Redshaw KA, Rhodes R. 2008. Season effects on productivity of some commercial South African sugarcane cultivars, I: Biomass and radiation use efficiency. *Proceedings of South African Sugar Technologists Association* 81: 517–527.
- Doorenbos J, Kassam AH. 1979. Yield Response to Water, FAO Irrigation and Drainage Paper No. 33, Rome: Food and Agriculture Organization of the United Nations.
- Dunham RJ. 1993. The Sugar Beet Crop: Science into Practice. In: Cook DA, Scott RK (eds), *Water Use and Irrigation*, London: Chapman Hall. pp 279–309.
- Hatti-Kaul R, Törnvall U, Gustafsson L. 2007. Industrial biotechnology for the production of bio-based chemicals: a cradle to grave perspective. *Trends in Biotechnology* 25: 119-124
- Keating BA, Robertson MJ, Muchow RC, Huth NI. 1999. Modelling sugarcane production systems 1: Development and performance of the sugarcane module. *Field Crops Research* 61: 253–271.
- Kingston G. 1994. Benchmarking yield of sugarcane from estimates of water use. *Proceedings of Australian Society of Sugarcane Technologists* 16: 253-271.
- Krishnan C, da Costa Sousa L, Jin M, Chang L, Dale B, Balan V. 2010. Alkali-based AFEX pre-treatment for the conversion of sugarcane bagasse and cane leaf residues to ethanol. *Biotechnology and Bioengineering* 107: 441–450.
- McGlinchey MG, Inman-Bamber NG. 1996. Predicting sugarcane water use with the Penman-Monteith equation. In: Champ CR, Sadler CREJ, Yoder REJ (eds), *Evapotranspiration and Irrigation scheduling, Proceedings of the International Conference*, 3-6 November, San Antonio, Texas, USA. pp 592-598.
- Mcintyre RK, Nuss KJ. 1996. An evaluation of sugarcane variety N19 under rainfed conditions in the South African sugar industry. *Proceedings of the South African Sugar Technologists Association*, 70:120-124.

- Mengistu M, Kunz R, Gush M, Steyn M, du Toit E, Everson C, Jewitt G, Naiken V, Doidge I, Clulow A. 2013. Interim report—Estimation and quantification of WUE of selected bio-fuel crops, WRC progress report for project K5/1874//4, Water use of cropping systems adapted to bio-climatic regions in South Africa and suitable for biofuel production. Pretoria, Water Research Commission.
- Muchow RC. 1989. Comparative productivity of maize, sorghum and pearl millet in a semi-arid tropical environment: I. Yield potential. *Field Crops Research* 20: 191-205.
- Muchow RC, Spillman MF, Wood AW. 1994. Radiation interception and biomass accumulation in a sugarcane crop grown under irrigated tropical conditions. *Australian Journal of Agricultural Research* 45 (1): 37-49.
- Murray SC, Rooney WL, Mitchell SE, Sharma A, Klein PE, Mullet JE, Kresovich S. 2008. Genetic improvement of sorghum as a biofuel feedstock: II QTL for stem and leaf structural carbohydrates. *Crop Science* 48: 2180–2193.
- Nair SS, Kang S, Zhang X, Miguez FE, Izaurralde RC, Post WM, Dietze MC, Lynd LR, Wullschleger SD. 2012. Bioenergy crop models: descriptions, data requirements and future challenges. *GCB Bioenergy* 4: 620–633.
- Narayanan S, Aiken RM, Vara Prasad PV, Xin Z, Yu J. 2013. Water and Radiation Use Efficiencies in Sorghum. *Agronomy Journal* 105 (3): 649–656.
- O’Shea MG, Keeffe EC, Burns EM, Staunton SP. 2013. An analysis of lignocellulosic diversity of sugarcane. *Proceedings of International Society Sugar Cane Technologists* 28: 215.
- Olivier FO, Singels A. 2012. The effect of crop residue layers on evapotranspiration, growth and yield of irrigated sugarcane. *Water SA* 38 (1): 77-86.
- Park SE, Robertson M, Inman-Bamber NG. 2005. Decline in the growth of a sugarcane crop with age under high input conditions. *Field Crops Research* 92 (2/3): 305-320.
- Ragauskas AJ, Williams CK, Davidson BH, Britovsek G, Cairney J, Eckert CA, Frederick WJ, Hallett JP, Leak DJ, Liotta CL, Mielenz JR, Murphy R, Templer R, Tschaplinski T. 2006. The path forward for biofuels and biomaterials. *Science* 311: 484-489.

- Rao PJM. 1997. *Industrial utilisation of sugar cane and its co-products*. New Delhi: ISPCK Publishers.
- Renouf MA, Wegener MK, Nielsen LK. 2008. An environmental life cycle assessment comparing Australian sugarcane with US corn and UK sugar beet as producers of sugars for fermentation. *Biomass and Bioenergy* 32 (12): 1144-1155.
- Rinaldi M, Vonella AV. 2006. The response of autumn and spring sown sugar beet (*Beta vulgaris* L.) to irrigation in Southern Italy: Water and radiation use efficiency. *Field Crops Research* 95: 103–114.
- Robertson MJ, Wood AW, Muchow RC. 1996. Growth of sugarcane under high input conditions in tropical Australia. I. Radiation use, biomass accumulation and partitioning. *Field Crops Research* 48: 11-25.
- Rooney WL, Blumenthal J, Bean B, Mullet JE. 2007. Designing sorghum as a dedicated bioenergy feedstock. *Biofuels, Bioproducts and Biorefining* 1: 147-157.
- Rosenthal WD, Gerek TJ, Wade LJ. 1993. Radiation use efficiency among grain sorghum cultivars and plant densities. *Agronomy Journal* 85: 703-703.
- Schoonees-Muir B, Ronaldson N, Naidoo G, Schörn PM. 2009. SASTA laboratory manual. 5th Edition. South African Sugar Technologists' Association, Mount Edgecombe. ISBN 978-0-620-43586-4.
- Seebaluck V, Sobhanbabu PRK. 2012. Sugar cane processing and energy generation from fibre resources. In: Johnson FX, Seebaluck V (eds), *Bioenergy for sustainable development and international competitiveness: The role of sugarcane in Africa*. Oxon: Routledge. pp 113.
- Sinclair TR, Muchow RC. 1999. Radiation use efficiency. *Advances in Agronomy* 65: 215-265.
- Singels A, Bezuidenhout CN. 2002. A new method of simulating dry matter partitioning in the Canegro sugarcane model. *Field Crops Research* 78: 151–164.
- Sluiter A, Hames B, Ruiz R, Scarlata C, Sluiter J, Tempelton D. 2005a. *Determination of Ash in Biomass*. Technical report NREL/TP-510-42622, National Renewable Energy Laboratory, U.S. Department of Energy, Office of Energy Efficiency & Renewable Energy.

- Sluiter A, Ruiz R, Scarlata C, Sluiter J, Tempelton D. 2005b. *Determination of Extractives in Biomass*. Technical report NREL/TP-510-42619, National Renewable Energy Laboratory, U.S. Department of Energy, Office of Energy Efficiency & Renewable Energy.
- Sluiter A, Hames B, Ruiz R, Scarlata C, Sluiter J, Tempelton D, Crocker D. 2011. *Determination of Structural Carbohydrates and Lignin in Biomass*. Technical report NREL/TP-510-42618. National Renewable Energy Laboratory, U.S. Department of Energy, Office of Energy Efficiency & Renewable Energy.
- Soil Classification Working Group. 1991. *Soil classification: A taxonomic system for South Africa*. Pretoria, Agricultural Research Council – Institute for Soil, Climate and Water.
- SASRI (South African Sugarcane Research Institute). 2006. Variety information sheets. South Africa. Available at: <http://www.sasa.org.za/divisions/SASugarCaneResearchInstitute/VarietyInformation/VarietyInfoSheets.aspx> [accessed January 2015].
- SASRI. 2013. SASRI mill room manual. Mount Edgecombe. South Africa.
- Tanner CB, Sinclair TR. 1983. Efficient water use in crop production: research or re-search? In: Taylor HM, Jordan WR, Sinclair TR (eds), *Limitations to Efficient Water Use in Crop Production*. Madison, ASA. pp 1-27.
- Tew LT, Cobill MR. 2008 (eds). *Genetic Improvement of Sugarcane (Saccharum spp.) as an Energy Crop: Genetic improvement of Bioenergy Crops*. New York: Springerlink Science and Business Media.
- Waclawovsky AJ, Sato PM, Lembke CG, Moore PH, Souza GM. 2010. Sugarcane for bioenergy production: an assessment of yield and regulation of sucrose content. *Plant Biotechnology Journal* 8:1-14.
- Zhao YL, Dolat A, Steinberger Y, Wang X, Osman A, Xie GH. 2009. Biomass yield and changes in chemical composition of sweet sorghum cultivars grown for biofuel. *Field Crops Research* 111: 55–64.