Genetic coefficients of sugarcane phenology traits for crop model refinement

by

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Declaration

"I declare that the thesis hereby submitted by **Immaculate Nontokozo Hlengiwe Ngobese** for the degree MSc. Agric. at the University of the Free State is my own independent work and has not previously been submitted by me at another University/Faculty. I further more cede copyright of the thesis in favour of the University of the Free State."

Dedication

This dissertation is dedicated to God Almighty who has been with me, guided me and comforted me every step of the way and for blessing me with wonderful people in my life, including my husband

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LIST OF ACRONYMS AND ABBREVIATIONS

%	percentage
^{o}C	Degree Celsius
^{o}Cd	Degree day
AK	Amatikulu
AMMI	Additive Main Effects and Multiplicative Interaction effects
ANOVA	Analysis of variance
BH	Bruynshill
C	Cultivar/genetic variance
cm	centimetre
d	day
DAP	Days after planting
DTOT	Total biomass
E	Environmental variance
ERC	Estimated recoverable crystal
FAS	Fertilizer advisory services
FiPAR	Fractionally intercepted photosynthetically-active radiation
FPOP	Final tiller population
G	gram
G x E	Genotype by environment interaction
GLA	Green leaf area
H^2	Broad sense heritability
Ha^{-1}	Per hectare
K	Canopy light extinction coefficient
Kg	kilogram
LAI	Leaf area index
LAmax	Maximum leaf area
LAR	Leaf appearance rate
LAR_1	Phyllochron interval 1 (for leaf numbers below LAR_{SWITCH})
LAR_2	Phyllochron interval 2 (for leaf numbers above LAR_{SWITCH})
LAR_{SWITCH}	Leaf number at which the phyllochron changes
LER	Leaf elongation rate
LFmax	Maximum number of green leaves
LI	Light interception
LN	Leaf number above which leaf area is limited to LA_{MAX}
LSD	Least significant difference
M1	Error mean square
M2	Mean square of cultivar, site and ratoon interaction
M3	Mean square of interaction of ratoon with site
M4	Mean square of interaction of cultivar with site
M5	Mean square of cultivar
Max	maximum
MAX POP	Maximum population
MET	multi-environment trial

Min	minimum
MJ	Megajoule
MJ^{-1}	Per megajoule
Mm	Millimetre
m^2	Square metre
MS	Mean square
P	Plant crop
PAR	Photosynthetically active radiation
PARCE	Photosynthetic active radiation conversion efficiency
PCA	Principal component analysis
PG	Pongola
POPTT16	Stalk population at/after 1600 degree days
PTP	Peak tiller population
R	Ratoon/Ratoon crop
R	Correlation
RA	Ratooning ability
REML	Restricted maximum likelihood
Rep	replication
RUE	Radiation use efficiency
RV	Recoverable value
S	Site
SASRI	South African Sugarcane research institute
SER	Stalk elongation rate
$SWDF_i$	Levels of water stress
TAR	Tiller appearance rate
Ton	Tonne
TOT	Crop size
TSP	Tiller survival percentage
TT	Thermal time
TTEMP	Thermal time to emergence for a plant crop
TTEMR	Thermal time to emergence for a ratoon crop
TTMTP	Thermal time from emergence to peak tiller population
TTPP	Thermal time to peak tiller population
TTSSE	Thermal time from emergence to start of stalk growth
TU	Cumulative thermal units
TU^{-1}	Rate of leaf appearance
TVD	Top visible dewlap
V_A	Additive genetic variance
V_G	Total genetic variance
V_P	Phenotypic variance
σ^2g	Genetic variance
σ^2p	Phenotypic variance
σ^2gs	cultivar x site interaction
σ^2gr	cultivar x ratoon interaction
σ^2gsr	cultivar x site x ratoon interaction

$\sigma^2 e$	environmental variance

Abstract

Crop models provide a simulation of crop growth and development through the use of mathematical equations and have substantial potential as research tools. They can assist breeding by predicting complex traits (e.g. sucrose yield) through simulating interactions between simple genetic traits (e.g. leaf elongation rate per unit thermal time) and environmental factors (e.g. temperature). The Canegro sugarcane model uses cultivar coefficients to simulate the effects of genotype, environment, and management on crop performance. The current coefficients in the Canegro model are limited to data from the cultivar NCo376 and estimates for a wider range of cultivars are not available for key growth parameters. The primary objective of this study was to quantify the cultivar coefficient values for some tillering and stalk elongation, leaf phenology, and biomass production traits for a diverse range of sugarcane cultivars. An additional objective was to determine the stability and heritability of these traits across environments and crop stages to determine their potential contribution to future model-assisted breeding.

Cultivar trials were established at three separate sites on South African Sugarcane Research Institute (SASRI) research farms; Amatikulu (AK), Pongola (PG), and Bruynshill (BH). The same set of 12 cultivars was tested at the three sites. The trials were planted in randomized complete block designs with four replications. The following cultivar traits were determined from within-season growth measurements: peak tiller population (PTP); thermal time to peak tiller population (TTPP); final population (FPOP); tiller survival percentage (TSP); stalk elongation rate (SER); leaf appearance rate (LAR); maximum leaf area (LAmax); thermal time to maximum leaf area (TTLAmax); maximum leaf number (LFmax); and leaf area index (LAI). Cane yield, estimated recoverable crystal percent (ERC%), ERC yield, total biomass, and brown (dead), and green leaf material were determined at each harvest. Plant and first ratoon crops were harvested at AK and PG, while only the plant crop was harvested at BH. The data were analysed using GENSTAT to estimate the variance components associated with cultivar, site, crop, and their interactions. Broad-sense heritability was calculated for each trait. Cultivar rank correlations across sites and across crops within sites were evaluated as a measure of trait stability.

The highly significant ($P<0.01$) effect of cultivar (C) was larger than the cultivar x ratoon (C x R) and cultivar x site (C x S) effects for most traits. Mean trait values for most traits differed

significantly between sites and ratoons within sites. Cultivars generally showed consistent rankings for PTP, TSP, SER, LAR, LAmax, LAI, and ERC% across sites for individual crops. Cultivars also showed consistent rankings across ratoons within a site for PTP, FPOP, SER, LAmax, LAR, LFmax, LAI, ERC%, cane yield, and ERC yield. This suggests that some traits are stable and can therefore be used for model-wise exploration of genotype by environment (G x E) interactions in sugarcane. Also, it may be feasible to characterise cultivars for some traits from single-site and single-ratoon experiments in the future. Some cultivars were identified as ideal indicator cultivars for future characterisation studies. Broad sense heritability estimates ranged from 0 to 0.99 for all traits studied. The FPOP, PTP, SER, LAR, LFmax, LAmax, LAI, cane yield, ERC%, and total biomass had high broad sense heritability estimates. These traits are therefore largely genetically controlled and can be selected for in a breeding programme.

The cultivar coefficient values determined here will be incorporated into the Canegro crop model and help refine the model's ability to simulate cultivar growth differences across environments. The range of values determined for these traits will also contribute to model-wise exploration of G x E interactions and future model-assisted breeding efforts for sugarcane.

Opsomming

Gewasmodelle verskaf ‘n simulasie van gewasgroei en ontwikkeling deur die gebruik van wiskundige vergelykings en het groot potensiaal as navorsingshulpmiddel. Dit kan teling ondersteun deur komplekse eienskappe te voorspel (bv. sukrose opbrengs) vanaf interaksies tussen eenvoudige genetiese eienskappe (bv. blaar verlengingstempo per eenheid hitte tyd) en omgewingsfaktore (bv. temperatuur). Die Canegro suikerriet model gebruik cultivar koeffisiente om die effek van genotipe, omgewing en bestuur op gewasproduktiwiteit te simuleer. Die huidige koeffisiente in die Canegro model word beperk tot data van die cultivar NCo376 en skattings vir ‘n wyer reeks van cultivars is nie beskikbaar vir sleutel groei parameters nie. Die primêre doel van hierdie studie was om cultivar koeffisiënt waardes te kwantifiseer vir sekere stoel en stam verlengings eienskappe, blaar fenologie, en biomassa produksie eienskappe vir ‘n diverse reeks suikerriet cultivars. ‘n Addisionele doel was om die stabiliteit en oorerflikheid van hierdie eienskappe oor omgewings en gewasstadiums te bepaal om hulle potensiële bydrae tot toekomstige model-ondersteunde teling vas te stel.

Drie cultivarproewe is gevestig by drie omgewings op SASRI navorsingsplase; Amatikulu (AK), Pongola (PG) en Bruynshill (BH). Dieselfde stel van 12 cultivars is getoets by die drie omgewings. Die proewe is geplant in gerandomiseerde blokontwerpe met vier herhalings. Die volgende cultivar eienskappe is bepaal van binne-seisoen groei metings: piek stam populasie (PTP); hittetyd tot piek stam populasie (TTPP); finale populasie (FPOP); stam oorlewings persentasie (TSP); stam verlengingstempo (SER); blaar verskyningstempo (LAR); maksimum blaararea (LAmax); hittetyd tot maksimum blaararea (TTLAmax); maksimum getal blare (LFmax); en blaararea indeks (LAI). Rietopbrengs, geskatte herwinbare kristal persentasie (ERC%), ERC opbrengs, totale biomassa, en bruin (dooie) en groen blaarmateriaal is is bepaal by elke oes. Plant en eerste ratoen gewasse is geoes by AK en PG, terwyl net plant gewas geoes is by BH. Die data is geanaliseer met GENSTAT om die variansie komponente geassosieer met cultivar, omgewing, gewas en hulle interaksies te bepaal. Breë sin oorerflikheid is vir elke eienskape bereken. Cultivar rangorde korrelasies oor omgewings en gewasse is geëvalueer as ‘n meting van eienskap stabiliteit.

Die hoogs betekenisvolle ($p<0.01$) effekte van cultivar (C) was groter as die van cultivar x ratoen (C x R) en cultivar x omgewings (C x S) effekte vir meeste eienskappe. Gemiddelde eienskap waardes vir meeste eienskappe het betekenisvol verskil tussen omgewings en ratoene

binne omgewings. Cultivars het oor die algemeen konstante rangordes getoon vir PTP, TSP, SER, LAR, LAmax, LAI en ERC% oor omgewings en individuele gewasse. Cultivars het ook konstante rangordes oor ratoens binne ‘n omgewing vir PTP, FPOP, SER, LAmax, LAR, LFmax, LAI, ERC%, riet opbrengs en ERC opbrengs getoon. Dit dui aan dat sommige eienskappe stabiel is en dat hulle gebruik kan word vir model-wyse ondersoeke van genotipe by omgewing (G x E) interaksie in suikerriet. Cultivars mag ook gekarakteriseer word vanaf enkel omgewing en enkel-ratoen proewe in die toekoms. Sommige cultivars is geïdentifiseer as ideaal vir toekomstige karakteriserings studies. Breë sin oorerflikheidsskattings het gewissel van 0 tot 0.99 vir alle eienksappe. Die FPOP, PTP, SER, LAR, LFmax, LAmax, LAI, rietopbrengs, ERC% en totale biomassa het hoë breë sin oorerflikheid getoon. Hierdie eienskappe word daarom grootliks geneties beheer en kan geselekteer word in telings programme.

Die cultivar koefissiënte wat hier bepaal is sal in die Canegro gewasmodel geïnkorporeer word en sal help om die model se vermoë te verfyn om cultivar groeiverskille te simuleer oor omgewings. Die waardes wat bepaal is vir die gemeette eienskappe sal ook bydra tot model gebasseerde ondersoek van G x E interaksies en toekomstige model-ondersteunde teling in suikerriet.

CHAPTER 1

General introduction

Sugarcane (*Saccharum officinarum)* is a large, perennial grass that is grown in tropical or subtropical areas which are within 30^{o} of the equator (Ming et al., 2006). Sugarcane is the largest member of the Poaceae family and is responsible for approximately 70% of sugar production worldwide with a world average yield of 68 ton ha^{-1} (Contreras et al., 2009). South Africa is one of the top nine leading countries in sugar production (Fischer et al., 2009). The South African industry continues to be one of the worlds' most cost competitive producers of high quality sugar producing an estimated average of 2.2 million tons of sugar per season.

Sugarcane productivity is generally measured in terms of cane yield (biomass) and sucrose content (quality) (Donaldson et al., 2008). These traits form the basis for cultivar selection. Factors which limit sugarcane yields include climatic variation, insect and disease pressure, marginalisation of sugarcane growing areas by other competitive crops and absence of break-throughs in breeding programmes (Mnisi and Dlamini, 2012). There are records of yield decline in the South African sugar industry (Singels et al., 2005; 2011). Crop models have been developed and applied in many areas of research, including estimating the sensitivity of crop production to climate change (Williams et al., 1988), evaluating cultivar performance (Boote et al., 2003), assessing the adaptation of a new cultivar to a region (Muchow et al., 1991), studying the nature of genotype x environment interaction (White, 1998), forecasting crop yield before harvest (Yun, 2003) and evaluating improved management options (Paz et al., 2007). Crop model simulations can be beneficial in overcoming the challenges in productivity (Zhou, 2003; Bezuidenhout, 2005).

Crop growth models help analyse and predict the effects of genotype, environment and management on crop performance and resource dynamics. Bannayan and Crout (1999) emphasised that crop forecasting is one of the most important potential applications of crop modelling. The ability of the models to simulate cultivar differences will allow growers to choose suitable cultivars for specific growing environments, which will enhance sucrose production (Zhou, 2003).

The Canegro model is a detailed research model of SASRI (van den Berg and Smit, 2005) which was constructed by Inman-Bamber (1991) from the CERES-Maize model (Jones and

Kiniry, 1986).Canegro is one of the leading sugarcane crop growth models worldwide (O'Leary, 2000) and has been shown to accurately simulate sugarcane yield when compared with South African sugar industry data (Bezuidenhout and Singels, 2007a; 2007b). It was developed primarily as a tool to direct and assist research (Inman-Bamber, 1995). The model uses the concept of cultivar coefficients, which are crop characters that define the development, vegetative growth, and reproductive growth of individual genotypes by summarising quantitatively how a genotype responds to environmental factors.

Crop simulation modelling can assist plant breeding through the predictions of complex trait values using simple traits. The model is able to quantify and integrate crop responses to genetic, environmental, and management factors and can, therefore, be used as a tool to study G x E interactions. Most G x E studies do not investigate the contribution of lower level traits on yield. Future research needs to focus on quantifying lower-level cultivar traits for different genotypes in different environments and evaluating the G x E interactions for such traits. Zhou et al. (2003) showed that some lower-level cultivar traits could be used to quantify cultivar coefficients used in crop models. The traits also offer potential as selection criteria that will improve identification of superior sugarcane genotypes (Zhou, 2005). There is limited use of lower level traits in crop modelling. Various studies in other crops such as soybean, cassava and other grains have been conducted to determine genetic coefficients. Studies on soybean by Mavromatis et al. (2001) and Irmak et al. (2000) have used typical information such as final seed yield, seed size, canopy height and anthesis to develop genetic coefficients. However, no tests have been done on detailed growth information from different regions.

Currently the cultivar trait coefficients in the Canegro model are limited to data from one cultivar only (NCo376) (O'Leary and Kiker, 2000). Examples of such traits include LAR, TAR (tiller appearance rate), PTP, TTTP and SER. More research is therefore needed to determine the trait values for a wider range of contrasting sugarcane cultivars. Additionally, information on the stability of these traits across sites and across ratoon crops is needed to evaluate the ease of cultivar characterisation in the future, and the potential use of the trait for model-assisted breeding efforts. For example, by varying individual trait values during simulations, it will be possible to evaluate the importance of that trait on final sucrose yields. If such traits are highly stable with high heritability, breeders could potentially select for the trait with greater confidence of its influence on final yields.

These quantified trait values will be incorporated into the Canegro crop model to refine its ability to simulate cultivar growth differences across environments. The realistic range values for the selected lower level traits can be used as a selection criterion and to help understand G x E interactions. An additional objective was to determine the stability and heritability of these traits across environments and crop stages to determine their potential contributions to future model-assisted breeding. Evaluating the stability of cultivars in relation to lower level traits used in modelling will help us understand how crop models can be used in breeding. If the lower level traits are found to be heritable, they could assist in ideotype development. Incorporating the determined values into crop models (such as the Canegro crop model) will speed up the process and reduce the cost of conducting multi-environment trials (MET's).

The specific objectives of this study were to:

- Determine trait coefficient values for a diverse range of cultivars to improve the Canegro model's ability to simulate cultivar differences
- Determine realistic range values for all traits to assist with model-wise exploration of trait suitability to different environments.
- Evaluate the stability of traits across environments (sites and ratoons) i.e. effects of G x E
- Determine the heritability of traits to evaluate their potential use in breeding programmes.

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CHAPTER 2

Literature review

2.1 Sugarcane origin

Sugarcane is a large, perennial, tropical or subtropical grass which is widely grown in zones within 30º of the equator (Ming et al., 2006). The origin of sugarcane is believed to be the Pacific islands from where it spread to other regions like the Asiatic islands and India, before spreading to other areas of the world (Barnes, 1953). The botanical classification of sugarcane is *Saccharum officinarum* and it is the largest member of the Poaceae family. The other widely recognised species of the genus *Saccharum* are *S. barberi*, *S. robustum*, *S. sinense* and *S. spontaneum* (Barnes, 1974). According to Grivet et al. (2004) sugarcane genetic resources can be divided into three groups:

(i) Traditional cultivars: these are the noble cultivars which have brightly coloured stalks and are rich in sugar e.g. *S. officinarum* L. and the North Indian and Chinese cultivars which have thinner stalks, flatter colours and lower sugar content, e.g. *S. barberi;*

(ii) Wild relatives: related to the traditional cultivars, they are informally grouped into the 'Saccharum complex', have little or no sugar and have diverse morphological and ecological adaptations, e.g. *S. spontaneum* L.;

(iii) Modern cultivars: created by Dutch breeders in Java in the early 1900s (Burnquist, 2001); these are hybrids of traditional cultivars and *S. spontaneum* L. and replaced the traditional cultivars during the 20th century. Due to the unusually large number of chromosomes in species of *Saccharum* there is great variability in hybrids (Barnes, 1953).

2.2 The South African sugar industry

Sugarcane is responsible for approximately 70% of sugar production worldwide with a world average yield of 68 ton ha^{-1} (Contreras et al., 2009). Although 100 countries cultivate sugarcane, the majority of its production occurs in a few countries, one of which is South Africa (Fischer et al., 2009). The South African industry started in the mid 1800's and currently ranks approximately ninth as the world's largest sugarcane producing industry (Gopinathan, 2010).

Sugarcane cultivation in South Africa occurs along the east coast (Figure 2.1), extending from 25º33'S to 30º93'S and between 29º92'E and 32º32'E (Ramburan, 2012).

The South African industry continues to be one of the worlds' most cost competitive producers of high quality sugar, producing an estimated average of 2.2 million tons of sugar per season; which is produced by 14 mill supply areas, extending from Northern Pondoland in the Eastern Cape to the Mpumalanga Lowveld. These areas cover a total of 432 000 ha which has remained constant since 2000. The industry is currently made up of about 50 000 growers of which 48 000 are small scaled registered sugarcane growers (Meyer, 2006).

The South African payment system is based on the Recoverable Value (RV%) formula:

$RV\% = S - d*N - c*F$, where (1)

S = sucrose % in cane

N = non sucrose % in cane

F = fibre % in cane

d = coefficient to cater for losses of sucrose through molasses during processing

c = coefficient to cater for losses of sucrose from bagasse during processing

The d and c factors are approximately 0.42 and 0.02 respectively. The c factor is calculated annually based on a three-season rolling average (Meyer and Clowes, 2011) while the d factor is calculated monthly based on sucrose and molasses prices.

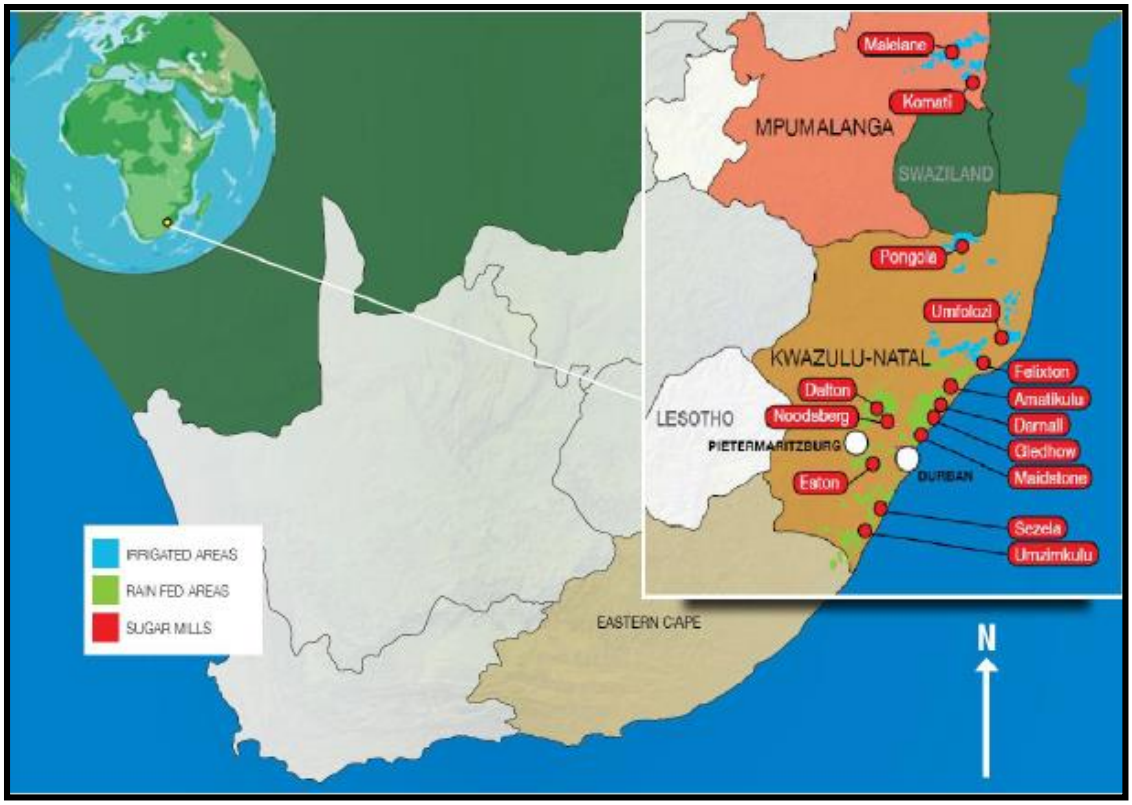

Figure 2.1 A map representing the sugarcane industry in South Africa (From SA Sugar Industry Directory 2010-2011)

2.3 Sugarcane growth and development in South Africa

The sugarcane plant is basically a stalk that is divided into nodes and internodes. Nodes are where lateral buds appear which are used for propagation. The stalk is the part which is of most interest to the grower because it is where the commercial product, sucrose, is stored (Barnes, 1974). The height and diameter of the stalk vary with cultivar and conditions of growth. The rate of stalk growth is affected by climatic and cultural factors (Ramburan et al., 2010). The stalk is also made up of fibre (bagasse) which is used for making cattle food, or paper. The baggase is also used in many sugarcane industries to cogenerate electricity for their own consumption (Mbhowa, 2013). During planting the stalk is sectioned in pieces called setts.

Conventionally, setts consisting of two to three buds are planted in furrows and new shoots emerge from lateral buds. The plant crop is generally harvested 12 to 24 months after planting, leaving behind a portion of the stem underground (Ming et al., 2006). It is this which gives rise to the succeeding growth of the cane known as ratoons (Barnes, 1974). The mass of roots and

underground parts of the stem from the previous crop quickly die off, leaving the new growth to survive and develop on nutrients absorbed through its own roots. Although several ratoons are possible from single plantings, continual stool damage from harvesting and weed control operations and the impact of pests and diseases eventually lead to a decline in yield with subsequent ratoons (Bull, 2000).

2.3.1 Germination and establishment

Sugarcane is generally propagated by cuttings of the stalk containing one or more buds (van Dillewijn, 1952). These are known as setts, seed cane or seed pieces. Germination begins with the development of organs already in the buds of these setts. During the initial stages of germination, root primordia around the nodes of the sett produce roots and the buds give rise to primary shoots. The roots, known as sett roots, are thin and fibrous and are important in maintaining the primary shoot until it has developed its own set of roots (Barnes, 1964). Germination starts from 7 to 10 days after planting and lasts for about 30 to 35 days, depending on environmental conditions.

Germination is influenced by many factors such as cultivar (Moreira and Cardoso, 1998), origin of the seed stock, age of seed cane (Das, 1981), nutrient supply in the cutting (Verma and Sudama, 1965), depth of planting (Humbert, 1968), orientation of the buds at planting, soil moisture (McMartin, 1957), temperature (Whiteman et al., 1963) and aeration (Singh and Ali, 1983). Limitations in one or more of these factors may result in microbial attack on the sett, causing it to decay. Among the various factors that influence the germination of sugarcane setts under field conditions, the water content of both soil and sett is the most important (Panje and Rao, 1963). Under excessive moisture conditions, major causes of germination failure are seed rots caused by attack of micro-organisms such as pineapple disease *(C. paradoxa)*. Excessive moisture may also suffocate the shoot roots, making it impossible for the shoot to emerge (Bakker, 1999). Jin-lan et al. (2010) showed that germination and emergence were best when the soil water content was 60 to 80%, whereas drought or excessive water had an adverse effect on germination. The germinating bud is initially dependent on sett nutrients and water (Bull and Glasziou, 1975).

Cultivars differ in their germination capacity and also their temperature sensitivity (Bull, 2000). Afghan et al. (2010) studied qualitative and quantitative characteristics of 13 sugarcane

cultivars and reported differences in germination percentage between different cultivars. Similarly, Sattar et al. (2010) showed a 16% difference in germination percentage between the highest and lowest germinating cultivars. Variation among sugarcane cultivars in respect to germination has also been reported by Ricaud and Domaingue (1991), Robertson et al. (1996) and Yadav (1981). Germination percentage has a direct bearing on plant population per unit area and is thus an important yield determinant of any sugarcane crop.

While the primary shoot forms, the lower buds near the bottom germinate and develop secondary shoots, which become tillers. This process continues, resulting in a number of tillers, which eventually form mature stalks of a stool. Tillering is normally completed four months after planting (Peng, 1984), but this varies depending on growing conditions and cultivar. This process continues until it is limited by factors such as lack of space (Shih and Gascho, 1980), light (Casagrande, 1991) (tillering stops when 70% light is intercepted by leaves), restriction of root development, or shortage of nutrients (Casagrande, 1991). Light is the most important external factor influencing tillering. Adequate light reaching the base of the sugarcane plant during the tillering period is of paramount importance. Temperatures around 30^{o}C are considered optimum for tillering, while temperatures below 20^{o}C retard tillering (van Dillewijn, 1952).

Maximum tiller population is normally reached around 90 to 120 days after planting (Diola and Santos, 2010). Only a portion of the tillers formed actually develop into mature canes. Some cultivars tiller early and profusely but most of the tillers may not survive; mortality of 30-60% of the total tillers may be due to moisture stress (Gosnell, 1968), increased competition (Ramesh and Mahadevaswamy, 2000) as well as crop husbandry and cultural practices (Kanyaiyalal et al., 1987). Although 6 to 8 tillers are produced per bud, only 1.5 to 2 tillers per bud remain to form mature cane stalks, and by about 150 to 180 days a stable population is established. Tillering provides the crop with the appropriate number of stalks required for a good yield. According to Raman et al. (1985) and Javed et al. (2000) stalk number is the major contributing factor to cane yield. Quebedeadux and Martin (1986) proposed that both stalk number and stalk weight should be assessed to get an accurate yield potential of any cultivar. Singh et al. (1985) also reported that stalk number was the most important character contributing directly to higher yield.

The rate of tillering, peak tiller number and final stalk population are largely genetically controlled. As many as 350 000 tillers per hectare have been recorded in South Africa on ratoon crops of cultivar NCo376 before full canopy (Meyer and Clowes, 2011). However, only about 155 000 tillers survived due to tiller mortality. Munir et al. (2009) compared yield and quality of cultivars from Faisalabad and revealed significant cultivar differences in tiller numbers. Khan et al. (2013) studied genetic and phenotypic diversity of ten elite sugarcane clones and showed significant differences in tiller number, stalk weight, cane girth, cane and sugar yields, and fibre content. Once tillers are established, elongation of the stalk follows (Barnes, 1964).

2.3.2 Stalk elongation

During stalk growth, each internode (joint) tends to function as a single unit. While it has a leaf attached, the internode completes cell elongation, cell-wall thickening and filling its storage volume with sugars, most of which are sucrose. Hence, the internodes complete their cycle by the time the attached leaf dies, and the lower internodes are essentially ripe while the upper part of the stalk is still growing (van Dillewijn, 1952). Stalk elongation is initially quick, and during this phase the fibre content of the stalk is very high. Stalk elongation takes place for about 270-300 days (Srivastava and Rai, 2012) and is affected by factors such as temperature (Edwards and Paxtan, 1979), moisture (Gosnell, 1968), age, and cultivar (Babu, 1990). Stalk elongation is very sensitive to both temperature and soil moisture. Rapid stalk elongation occurs when daily mean temperatures reach about 18.5°C and will continue to grow rapidly under warmer conditions at between 1 to 2 cm d^{-1}, and can almost cease when temperatures drop in the cool winter months. Water deficit during stalk elongation causes a lower rate of stalk elongation (Meyer and Clowes, 2011). Too much water will slow or stop elongation due to waterlogging in the root zone. Yield decline occurs in sugarcane if the meristem and uppermost leaves of the plant are below the water level. According to sugarcane farmers at Mfolozi the minimum period of inundation (flooding) before sugar-cane is completely destroyed, varies between approximately three days during warm months and six days if the flood occurs during cold months (Berning et al., 2000).

During early growth the rate of stalk elongation responds rapidly to rainfall when temperatures are relatively high. In fact, high growth at high temperatures is dependent upon adequate soil moisture. However, as the average temperature declines, growth also declines, even after significant rainfall.

Cultivars differ in their rate of stalk elongation (Smith, 1983). In sugarcane, it has been shown that stalk elongation rate per unit thermal time is genetically determined to a large extent (Smit and Singels, 2007). There is also evidence that stalk number, stalk height, stalk diameter, stalk weight, and cane yield show large and consistent genotypic variation (Silva et al., 2008). According to Lingle et al. (2009) the difference between stalk length of cultivars was primarily due to the increase in the number of internodes rather than the length of individual internodes.

2.3.3 Leaf growth and development

Leaves are the photosynthetic 'engines' of the plant, producing the sugar that is stored in the stalks. The leaves of sugarcane are arranged alternately with a single leaf arising from each node. Each leaf consists of a lower part (sheath) and an upper part (blade). The leaves are continually renewed; mature leaves die and young ones are added. The number of green leaves present on a stalk is governed by the rate at which the leaves are produced and the longevity of individual leaves (van Dillewijn, 1952). Leaf appearance is often represented by its inverse, the phyllochron, defined as the time interval between the appearance of successive leaves on a stem (Xue et al., 2004).

One of the most important characteristics of leaves with regard to their function is their total area. The leaves become both longer and wider as the plant develops, until a stable leaf size is established. Leaf area depends on the number of leaves and average surface per leaf (van Dillewijn, 1952) and gives an idea of the plants' photosynthetic capacity (Patil et al., 2009). The area of the individual leaf blades is smallest at the base of the plant and gradually increases toward the top until a maximum is reached. As new leaves appear and individual leaves expand, leaf area index increases. Leaf area index (LAI) estimates the crop's ability to capture light energy and is broadly defined as the amount of leaf area (m^2) in a canopy per unit ground area (m^2) (Watson, 1947). Radiation interception is a function of LAI; this therefore means that an increase in leaf area can improve interception of solar radiation (Shoko et al., 2009), which therefore influences biomass accumulation. Shih and Gascho (1980) reported a positive correlation between LAI and sugarcane biomass yield. In general, maximum LAI is achieved about six months from planting and then slowly declines. This may be affected by both cultivar of sugarcane and growing conditions (Rahman et al., 2001). LAI determines and controls canopy water interception, radiation interception, and water and carbon gas exchange (Sandhu et al., 2012).

Leaf area development is critical in the establishment of full leaf canopy for maximum radiation interception (Sinclair et al., 2004). Factors that affect tillering also affect the number of leaves and LAI (Shoko et al., 2007). These factors include crop nutrition (Moberly, 1971), trash management (Thompson, 1965), crop rotation (Garside et al., 2001), water use (Thompson and du Toit, 1965), genotype (Zhou, 2003), crop management (Braunak and Hurney, 1988), seasons (Inman-Bamber, 1994) and climatic conditions (Barnes, 1964). Leaf area index differs greatly between cultivars (Iqbal et al., 2011). Generally, cultivars having high population density possess a larger LAI (Iqbal et al., 2011). Rafiq et al. (2007) showed that early maturing cultivars like CPF-237 attain maximum LAI earlier than late or medium late maturing cultivars like SPF-213. In contrast, Robertson et al. (1996) investigated early growth between plant crops of two cultivars and found no difference for leaf area per stalk.

2.3.4 Maturity and ripening

The final growth stage in sugarcane is the maturity and ripening phase. Growth rate decreases, resulting in an increase in sucrose content (Bull, 2000). This phase commences eight months after planting and continues through to harvest (Binbol et al., 2006). During ripening, simple sugars (monosaccharides such as fructose and glucose) are converted into cane sugar (sucrose, a disaccharide). The sugarcane ripening proceeds from the bottom of the stalk to the top. Climatic factors such as rainfall, solar radiation, water availability and temperature are major factors that influence sugarcane maturation and the increase in sucrose content (Keating et al., 1999).

Clowes and Breakwell (1998) revealed that high temperatures, particularly at night, usually result in increased flowering of sugarcane. Flowering in sugarcane results in reduced cane and sucrose yields; achieved by stopping the growth of leaves and internodes. High temperatures are also known to negatively affect sprouting and sugarcane emergence (Rasheed et al., 2011). Poor emergence results in a significantly lower tiller population (Chandiposha, 2013). Under high temperatures (above 32°C) sugarcane cultivars limit internode growth, resulting in reduced sucrose content (Bonnett et al., 2006). In humid tropical and subtropical regions the dry season and low temperatures towards harvest are known to slow down growth, causing an increase in sucrose content (Clements, 1962). Water stress, caused by low soil water availability, reduces carbohydrate synthesis, leaf expansion, and internode elongation, which is followed by an increase in sucrose content (Alexander, 1973).

Sucrose content differences are evident between different sugarcane cultivars (Rohwer and Botha, 2001). Chohan et al. (2007) and Keerio et al. (2003) reported genetic differences among newly developed sugarcane genotypes for cane yield and yield contributing traits. Singh and Venkatarama (1983) and Lingle and Irvine (1994) observed the highest relative growth rates and net sucrose accumulation during stalk elongation and ripening of early cultivars when compared with late ones. Sharma and Kohi (1980) carried out investigations on various cane cultivars and found that cultivar COJ-64 produced the highest amount of sugar per hectare and was closely followed by the cultivars B0-70, CO-l148, C0-6239 and COS-687.

2.4 Crop modelling (Canegro model)

2.4.1 Introduction

A crop model is a simple representation of a crop. Models represent crop growth and growth responses to the environment, through the use of mathematical equations. Crop growth models help analyse and predict the effect of genotype, environment, and management on crop performance and resource dynamics. Crop models have been developed and applied in many areas of research, including estimating the sensitivity of crop production to climate change (Williams et al., 1988), evaluating cultivar performance (Boote et al., 2003), assessing the adaptation of a new cultivar to a region (Muchow et al., 1991), studying the nature of G x E interaction (White, 1998), forecasting crop yield before harvest (Yun, 2003) and evaluating improved management options (Paz et al., 2007). Bannayan and Crout (1999) emphasised that crop forecasting is one of the most important potential applications of crop modelling.

A wide range of crop models exist for various crops, including sugarcane. Canegro is one of the leading sugarcane crop growth models worldwide (O'Leary, 2000) and was shown to accurately simulate sugarcane yield when compared to the South African sugar industry data (Bezuidenhout and Singels, 2007a; 2007b). The Canegro model is a detailed research model of SASRI (van den Berg and Smit, 2005) which was constructed by Inman-Bamber (1991) from the CERES-Maize model (Jones and Kiniry, 1986). It was developed primarily as a tool to direct and assist research (Inman-Bamber, 1995).

The Canegro model uses daily weather data, cultivar and soil properties, as well as management input data to simulate sugarcane crop growth to predict cane yield, sucrose yield, crop biomass, nitrogen, and water use (Lisson et al., 2005). Canegro uses the concept of cultivar coefficients,

which are crop characters that define the development, vegetative growth and reproductive growth of individual genotypes by summarising quantitatively how a genotype responds to environmental factors. The cultivar trait coefficients are normally determined from field experiments through sampling of growth and development data for each cultivar at regular intervals. Cultivar properties simulated in the Canegro are whole plant, stalk and root biomass, sucrose concentration, plant phenology, and other variables (Singels et al., 2010).

2.4.2 Overview of the model

Figure 2.2 shows the effect of environmental factors on yield components of sugarcane. Daily meteorological observations define the atmosphere in the Canegro model. A daily observation typically includes temperature (^{o}C) and solar radiation. Temperature is regarded as one of the main driving forces for some physiological and physical processes in the sugarcane plant and its environment (Bezuidenhout, 2000). Daily heat units, also known as thermal time, are a common temperature property which affects respiration, photosynthesis, leaf properties (such as leaf elongation, leaf appearance rate and width) and shoot population (Figure 2.2).

Two parameters, number of tillers (shoot population) and total leaf area per tiller, determine the light interception ability of the canopy. Fractional intercepted photosynthetically active radiation (FiPAR) and solar radiation play an active role in gross photosynthesis (Spitters et al., 1986). The model calculates the gross photosynthesis on a crop level and respiration is divided into two processes; maintenance respiration and growth respiration. Respiration and photosynthesis processes are calculated as a fraction of biomass. During these growth processes, new dry matter is allocated to different organs of the plant (i.e. leaves and roots). In the model, root development is simulated first, while leaves and stalk growth accelerates at a later stage.

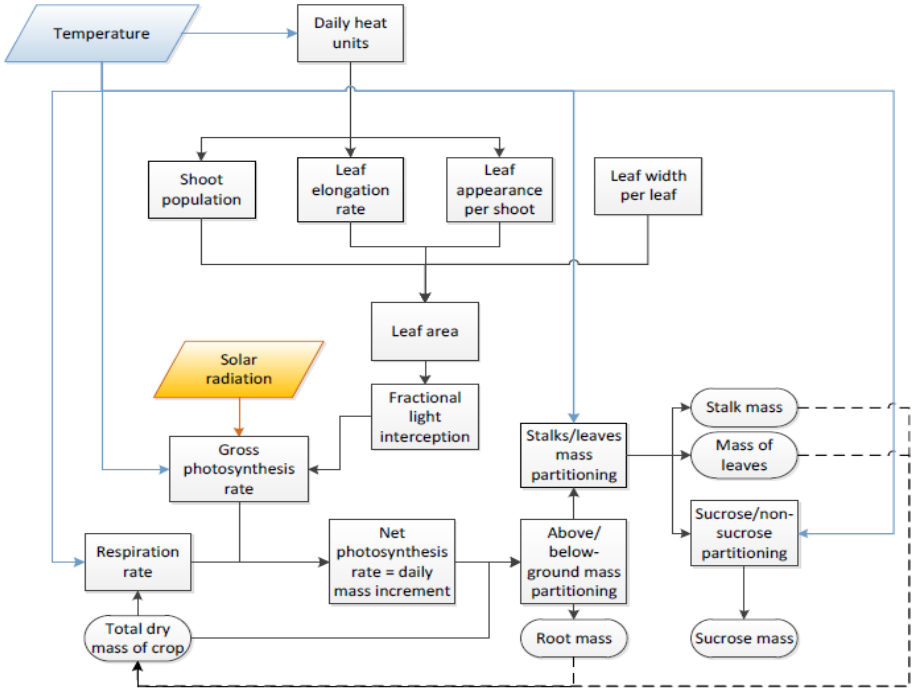

Figure 2.2 Schematic diagram of SASRI Canegro model plant growth and development processes (Jones, 2013)
(Square boxes are model processes and round boxes are state variables. Solid blue parallelograms and the solid shaded parallelograms are sources of weather data Solid black lines indicate data flow from calculations made from the current day's value, while dashed black line calculations are based from the previous day's values. Solid blue lines and the red line represent weather data flow. For the sake of simplicity, this diagram excludes the water balance and water stress impacts).

2.4.3 Cultivar traits used in the Canegro crop model

Plant properties are modelled in the Canegro model through the use of genetic coefficients. Genetic coefficients (or cultivar trait coefficients) are simple, cultivar specific traits that interact with the environment to express a more complex trait like yield. Examples of such traits include LAR, TAR, PTP, TTTP, SER and radiation use efficiency (RUE). Some of these traits are shown in Table 2.1 and are described briefly below.

Table 2.1 Selected phenological cultivar traits used in the Canegro crop model

Parameter	Units	Description
LAR_1	^oCd	Phyllochron interval 1 (for leaf numbers below LAR_{SWITCH}) - 14 days
LAR_2	^oCd	Phyllochron interval 2 (for leaf numbers above LAR_{SWITCH}) - 14 days
LAR_{SWITCH}	Leaf	Leaf number at which the phyllochron changes
MAX POP	stalks m^{-2}	Maximum tiller population
TTEMP	^oCd	Thermal time to emergence for a plant crop
TTEMR	^oCd	Thermal time to emergence for a ratoon crop
TTSSE	^oCd	Thermal time from emergence to start of stalk growth
TTMTP	^oCd	Thermal time from emergence to peak tiller population.
LN	Leaf	Leaf number above which leaf area is limited to LA_{MAX}
LA_{MAX}	cm^2	Max leaf area assigned to all leaves above leaf number LN
K		Canopy light extinction coefficient
SER	mm $(^oC\ h)^{-1}$	Change in plant extension rate per unit change in temperature
POPTT16	Stalks m^{-2}	Stalk population at/after 1600 degree days

The selected phenological traits are measured either in glasshouse or field conditions with high measurement intensity. Cane yield and its components (selected traits) is cultivar dependant. The difference in traits within a genotype grown in different environments is due to the response of the genotype to the environment. Currently in the Canegro model the number of traits that describe different cultivars is very low, as major trait values were determined from experimental data of the cultivar NCo376 only. More experimental work is therefore needed to determine the trait values for a wider range of contrasting sugarcane cultivars.

2.4.3.1 Tiller characteristics

Tiller population is an important crop variable that is simulated by the Canegro model. Germination and emergence of primary tillers, underground branching and emergence of secondary tillers and tiller senescence are three biological processes influencing tiller population. Primary tiller emergence is simulated when a specific period of thermal time has accumulated from planting (TTEMP) or ratooning (TTEMR). Underground branching will reach peak tiller population once a cultivar-specific amount of thermal time has been reached since emergence (TTMTP).

Donaldson et al. (2011) showed significant differences in maximum tiller population between cultivars NCo376 and N26 in a study at Pongola. TTPP was 461 and 655°Cd for the May and December crop starts, respectively. Tiller population maximums of 111 964 ha^{-1} and 55 000 ha^{-1} were produced by NCo376 and N26 in the December ratoons, respectively. Tiller population maximum was significantly lower in the May ratoons at 68 214 ha^{-1} and 28 750 ha^{-1} for NCo376 and N26, respectively. According to Zhou (2003) there was a significant difference in peak tiller population among the cultivars ZN6, ZN7, N14 and NCo376. Peak tiller population values were 150 700 ha^{-1}, 182 700 ha^{-1}, 248 000 ha^{-1} and 265 300 ha^{-1} for cultivars ZN6, ZN7, N14, and NCo376, respectively.

Shukla and Singh (2011) reported variable behaviour of three different cultivars (CoS 96269, CoPant 97222 and CoLk 9616). During the spring planting sugarcane genotype CoLk 9616 had the highest final tiller population compared with CoS 96269 and CoPant 97222 at all crop stages. Significantly higher numbers of millable canes (119 400 ha^{-1}) were produced by genotype CoLk 9616 compared with CoPant 97222 (93 940 ha^{-1}) and CoS 96269 (100 250 ha^{-1}). Similarly in summer, CoLk 9616 had the highest number of millable canes (98 900 ha^{-1}) while genotype CoS 96269 had the lowest number of millable canes (61 390 ha^{-1}).

Rafiq and Sattar (2013) reported differences in millable canes of different genotypes. The widely adapted cultivar HSF-240 produced significantly higher cane number in both years with an average of 147 650 stalks ha^{-1}. The lowest number of millable canes was 44 350 stalks ha^{-1} produced by cultivar Q-88.

The literature shows that there are differences in tillering characteristics between cultivars and that those differences vary depending on crop start date. Peak tiller population and final tiller population have been shown to be influenced by the planting season.

2.4.3.2 Leaf emergence and development

Leaf appearance is the number of leaves that become visible on a stem per unit time (Streck et al., 2003). Leaves will emerge based on a phyllochron interval. Phyllochron is the time interval between the appearance of successive leaves. This interval can be expressed on a calendar basis or in thermal time (TT), measured in units of degree-days (°C day). Two phyllocron intervals (°C day) are specified in Canegro: the first phyllocron interval (LAR_1) is used for calculating the timing of appearance of the first 14 leaves. For leaves emerging thereafter, LAR_2 is used. LAR_1 applies to leaves below a certain threshold, while LAR_2 applies to leaves above the threshold (LAR_{SWITCH}).

Leaf size and area is driven by leaf elongation rate (LER, cm d^{-1}) which is dependent on air-temperature as well as water availability. Leaf area expansion is calculated from increases in leaf length and width. Leaves stop expanding once a maximum allowable blade area is reached. This value increases for successive leaves (Inman-Bamber and Kiker, 1997) until a specific number of leaves LF_{MAX} has formed. After this, the maximum allowable blade area remains at a constant value LA_{MAX}.

It has been shown that these leaf development traits are influenced by genotype. Sinclair et al. (2004) studied leaf appearance of four cultivars and showed values for leaf appearance rate ranging from 0.0085 to 0.0115 leaf TU^{-1}[a], or a phyllochron interval of 118–87 TU[b]. Values of leaf appearance rates reported by Inman-Bamber (1994) for cultivars NCo376 and N12 were 0.0092 and 0.0085 leaf TU^{-1}, respectively (109 and 118 TU phyllochron interval, respectively).

Marin (2011) studied two cultivars in four environments and compared their performance to NCo376. The maximum leaf size differed little among the Brazilian cultivars, ranging between 796 and 733 cm^2. Both these cultivars reached maximum leaf size at leaf number 25, which is

[a] **TU^{-1} – rate of leaf appearance**

[b] **TU -cumulative thermal units**

similar to results of Sinclair et al. (2004) for cultivar CP72-2086 in Florida. The range of LAR_1 values (104-113^{o}C days^{-1} leaf^{-1}) was higher for both Brazilian cultivars than cultivar NCo376. However, at 116-122^{o}C days^{-1} leaf^{-1}, LAR_2 was smaller than observed for cultivar NCo376. Zhou (2003) evaluated four cultivars in one environment. The cultivars had different leaf sizes and youngest leaves that attained maximum leaf area. Values ranged from 457.8 cm^2 (N14) to 355.3 cm^2 (NCo376).

In sugarcane, it has been shown that leaf size and leaf elongation rate show consistent genotypic variation (Bonnet, 1998; Robertson et al., 1998). However, little genetic and phenotypic information on these traits is available for a wider range of genotypes in different locations.

2.4.3.3 Leaf area index and light interception

The fraction of photosynthetically active radiation (PAR) intercepted by the crop is determined by green leaf area index (LAI). LAI is the total one-sided area of leaf tissue per unit ground surface area. In Canegro, LAI is often simulated as the product of green leaf area (GLA) per plant and plant population (Muchow et al., 1990). Green leaf area per plant is determined by the number of green leaves per plant and the sum of their area.

By making use of LAI the fraction of PAR that is intercepted by the crop is calculated. This is known as the fractionally intercepted photosynthetically-active radiation (FiPAR) and is calculated by making use of Beer's law:

FI = 1 - exp (-Kc*GLAI)

Where *k* is a PAR extinction coefficient that changes as the crop develops. In Canegro, this is calculated as a function of the total number of leaves per tiller.

LAI differs greatly with cultivars. Generally, a cultivar with a high population density will have more LAI (Iqbal et al., 2011). Early maturing cultivars attain maximum LAI earlier than late maturing cultivars (Rafiq et al., 2007). Gomathi et al. (2011) reported significant variation in LAI between the genotypes Co 99004 and Co 99006. LAI under normal irrigated conditions was 3.89 and 3.86, for cultivars Co 99006 and Co 99004, respectively. Drought treatment caused an average reduction of 17.46% in LAI and values of 3.53 and 3.06 were recorded for genotypes Co 99004 and Co 99006 respectively.

Zhou (2003) plotted LAI against days after planting and accumulated thermal time. There was a marked difference among the cultivars in LAI towards the peak LAI. Cultivar N14 had the highest LAI while ZN7 had the lowest, with values that ranged from 0.082-1,833 and 0.062-1,229 respectively. Low stalk population cultivars (ZN6 and ZN7) reached their peak LAI earlier than high stalk population cultivars (N14 and NCo376). Cultivars achieved peak LAI around 2500^{o}C days from planting.

2.4.3.4 Stalk height and stalk elongation traits

Stalk elongation is of interest because stalk length is a major component of cane and sucrose yield. Stalk elongation starts when a genotype-specific thermal time has elapsed (TTSSE) after emergence. Shukla and Singh (2011) showed that individual cane length of genotype Col k 9616 (161.3 cm) was significantly higher than that of CoPant 97222 (156.7 cm) and CoS 96269 (147.2 cm). Similarly a study by Ethan et al. (2013) on three cultivars in Nigeria, showed significant differences in the stalk lengths of the sugarcane cultivars with values ranging from 86.30 cm to 98.02 cm. Shah et al. (2008) reported a range of 111 to 132 cm in stalk length for different genotypes.

Pedrozo (2010) reported that stalk height was significantly affected by genotype. Muturi and Wawire (2006) showed differences in stalk elongation rates of three sugarcane cultivars. Average stalk elongation rates under irrigation were 29.3, 19.3 and 18.2 cm month^{-1} for EAK70-97, CO1148 and CB38-22, respectively. These reduced to 13.3, 15.5, and 15.0 cm month^{-1} for rain fed crop (same cultivars). Maximum stalk elongation rate of up to 48 mm d^{-1} were recorded for Q127, 44 mm d^{-1} for Q96, and 38 mm d^{-1} for Ql17 by Shannon and Holden (1996).

Stalk characteristics are genetically and environmentally controlled. When stalk height data was plotted against days after planting and accumulated thermal time, Zhou (2003) reported that during the peak stalk elongation period, ZN7 had the tallest stalks while N14 had the shortest. Stalk elongation of December ratoons was reduced due to low winter temperatures experienced when they were six to eight months old (Donaldson et al., 2011).

2.4.3.5 Biomass accumulation and partitioning

Biomass accumulation and partitioning are some of the processes simulated in the Canegro model (Singels et al., 2013). Simulation of dry matter partitioning and distribution of a crop is an important means of predicting yield (Arvin et al., 2014). The Canegro model uses Beers law of radiation extinction to calculate photosynthetically active radiation (PAR). In the Canegro model total biomass (DTOT in ton ha^{-1} day^{-1}) is calculated by converting intercepted PAR (IPAR in MJ ha^{-1}) using photosynthetic active radiation conversion efficiency (PARCE in g MJ^{-1}) (Ngxaliwe, 2014). This conversion efficiency is affected by crop size (TOT in ton ha^{-1}) and the level of water stress ($SWDF_i$) (Rossler, 2013).

The biomass produced is partitioned to roots, stalks and leaves according to sink demand. Sink demand is determined from cultivar specific parameters, physiological age and current environmental conditions such as temperature and water status. The remainder is stored as sucrose in the stalk. The partitioning fraction to each component changes as the crop develops. Biomass is only partitioned into stalks after stalk elongation has commenced (Rossler, 2013).

2.4.4 Sugarcane breeding in South Africa (SASRI breeding programme)

Sugarcane breeding in South Africa started at an experiment station in Mount Edgecombe. One of the core functions of SASRI is to develop improved cultivars for the South African sugar industry (Parfitt, 2005). This is achieved through breeding, selection, and release of sugarcane cultivars that are adapted to the major agro-climatic regions of the industry (Parfitt, 2005). The five agro-climatic regions are the northern irrigated areas, coastal high potential, coastal average potential, coastal hinterland, and the midlands humic and sandy soils areas.

Parents used for crossing are selected from local or imported germplasm (Ramburan, 2010). Approximately 1500 crosses are made annually in glasshouses at SASRI during the crossing season (Parfitt, 2005). The resultant seed are germinated to produce potential new cultivars that are distributed to one of the SASRI selection stations in each agro-climatic region.

The next five selection stages of the breeding process are conducted on six research stations. Each research station receives clones that have been produced from parents that are adapted to that region. Cultivars are selected at each stage for their performance in a specific environment in terms of sucrose content, resistance to pests, yield stability, and cane yield (Anonymous,

2012). At the last stage, the top clones from each region are exchanged between research stations and also tested in multi-environment trials at off-station sites. This enables researchers at SASRI to evaluate their performance over a number of environments i.e. to evaluate G x E interaction. The entire breeding and selection process involves between 11 and 15 years of testing before a new cultivar can be released (Parfitt, 2005). Cultivars are released primarily based on their yield responses to soil and climatic conditions experienced on selection stations.

Crop simulation modelling can assist plant breeding, through the predictions of the selected traits (Table 2.1). The model is able to quantify and integrate crop responses to genetic, environmental, and management factors, and can, therefore, be used as a tool to study G x E interactions.

2.4.5 Role of crop models in breeding

One application of models in plant breeding is through the design of ideotypes for target environments (Boote and Tollenaar, 1994). An ideotype is a theoretical cultivar which has ideal traits for target environments. Crop models have been used to identify ideotypes for soybean and maize (Boote and Tollenaar, 1994), wheat (Aggarwal et al., 1997), peanut (Boote and Jones, 1986), and rice (Kropff et al., 1995). Crop models such as Canegro are able to identify traits that contribute to increases in potential yield. By varying trait values (cultivar coefficients) in the model, one can determine the optimal combination of traits (the ideotype) for specific environments. For example, in soybean, 10% genetic gain was simulated by adjusting growth habits, seed fill duration, and maximum leaf photosynthesis (Boote et al., 2001). The reliability of these estimates relies heavily on accurate and realistic ranges of the different traits within a crop. These simulations may be influenced by cultivar differences and/or environmental changes, thus information on the heritability and stability of the trait across environments is also required.

Currently, breeders make use of MET's to evaluate genotype performance and make selections of superior genotypes. However, METs are relatively expensive and time consuming. Muchow et al. (1996) suggested that crop growth models are powerful tools that can be used in conjunction with MET data to help interpret G x E interactions. Crop models are able to quantify and integrate genetic responses to environmental and management factors, and therefore, can be used as a tool to study G x E interactions. By incorporating genetic variation

into the model and varying these coefficients, MET's could be simulated for many more environments than are possible using conventional trial-based approaches. This could help understand the type of genotypes that are suited to different environments, and hence complement traditional breeding approaches. Aggarwal et al. (1996) found that total variation between genotype, environment, and G x E interactions in wheat, produced by simulation (4%, 75% and 15% respectively) was similar to the total variation observed in the field.

2.5 Genotype by environment interaction

Variation in the relative performance of cultivars across environments is termed G x E interaction. Genotypic (G), environmental (E), and genotype by environment (G x E) interaction are the three main components of variability in any population. G x E interactions are an old universal issue, in all living organisms, which has been poorly addressed (Kang, 2002). These interactions often complicate testing and selection of superior genotypes. The presence of G x E interactions causes differences in stability between genotypes, thus reducing the genetic progress in many breeding programmes. To assess the performance of genotypes under different environments, the mean yield of those genotypes can be used. When cultivars are grown at several locations for testing their performance, their relative rankings do not usually remain the same. This indicates the presence of G x E interaction (Issa, 2009). In plant breeding, the most important type of G x E interaction is crossover or qualitative (Figure 2.3c), which implies changes in the rankings of genotypes across environments (Baker, 1988). With non-crossover interactions (Figure 2.3b), genotypes with superior means can be recommended for all environments. Crossover G x E interaction results in complications when it comes to breeding, selection and testing of superior genotypes. Crossover interactions show that different genotypes are adapted to different environments. This existence of G x E interactions complicates the identification of superior genotypes for a range of environments (Issa, 2009).

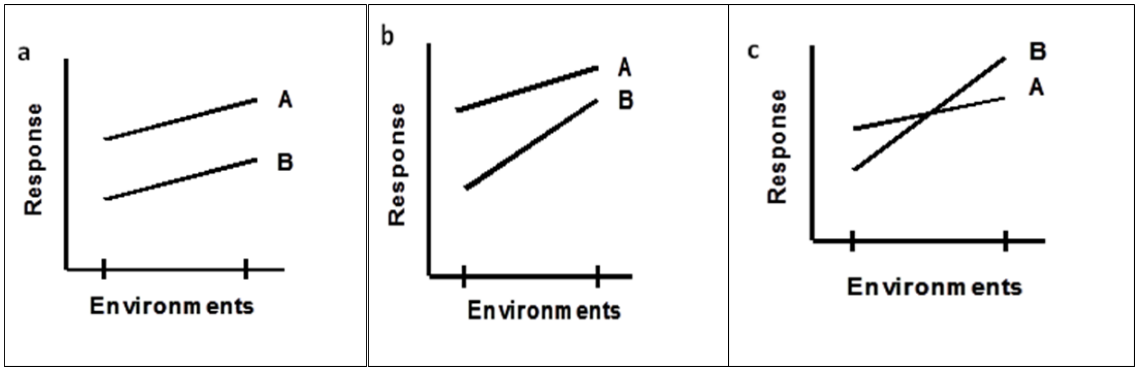

Figure 2.3 Graphical representation of types of G x E interaction: (a) no interaction - A and B responses parallel in the two environments; (b) non-crossover G x E interaction; (c) crossover interaction

2.5.1 Significance of G x E interactions

Different stresses imposed by environments result in inconsistent performance of cultivars across environments. Cultivars which are preferred by farmers are those which perform well and consistently under different conditions. Studying of G x E interaction is important to plant breeders because this interaction can limit the progress in the selection process. Understanding the cause of G x E interaction is important to help in selecting cultivars with high yields that are stable across pertinent environments (Masindeni, 2006).

The G x E interaction determines if a genotype is widely adapted for an entire range of environmental conditions or whether separate genotypes must be selected for different sub environments. Information on the structure and nature of G x E interaction is useful to breeders as it can help determine the need to develop new cultivars for all environments of interest (Bridges, 1989). G x E interaction studies have been carried out on many crops worldwide. Gilbert et al. (2006) indicated that significant G x E interactions persist beyond cultivar release in commercially grown genotypes in Florida. Cultivar (genotype), environment, time of harvest and their interactions had significant effects on sugar per ton, tons of cane per hectare, and tons of sugar per hectare (of three data sets).

In South Africa, Laubscher et al. (2000) found that minimum night temperature was a major contributor to G x E in maize. G x E interactions have been found in other crops including soybean (Smit and de Beer, 1991), lucerne (Smith and Smith, 1992) and potatoes (Steyn et al., 1993). Research on causes of G x E interactions is limited, and is usually focused on yield performance over environments. Most of these studies do not account for the key influences of

lower level traits (such as those traits used in crop models) on yield. Information is therefore needed on the G x E interactions associated with the key cultivar traits used in crop models.

2.5.2 Methods to analyse G x E interactions

Various statistical methods are available to quantitatively evaluate and describe G x E interactions. These methods include analysis of variance (e.g., least squares, restricted maximum likelihood=REML), stability analysis (e.g. regression), multivariate methods (e.g. principal component analysis (PCA), cluster analysis, factor analysis, and Additive Main Effects and Multiplicative Interaction effects (AMMI model).

2.5.2.1 Analysis of variance (ANOVA)

An ANOVA is the most widely used and usually the first step in any statistical analysis of G x E interaction data (Skroppa, 1984). Multi-environment trials of genotypes are considered as factorial designs (Rafii et al., 2012) that investigate effects of genotypes, environments, and their interaction (i.e G x E interaction). The ANOVA model may be expressed by a linear equation:

$$Xij = \mu + Gi + Ej + GEij + eij \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

Where:

Xij is the expected yield of the i^{th} genotype in the j^{th} environment

μ is the general mean value of the population (trial)

Gi, *Ej* and *GEij* represent the effects of the genotype or cultivar, environment, and G x E interaction respectively

e*ij* represents the residual error

In ANOVA, magnitudes of sums of squares of relevant terms as well as variance components are used to quantify sources of variation. ANOVA allows the determination of the components of variance arising from the different factors. If the G x E interaction variance is found to be significant, one or more of the various methods for measuring the stability of genotypes can be used to identify the stable genotype/s. If there is a significant G x E interaction the superiority

of a genotype depends on the environment in which it is grown. This means that the breeder should consider developing different cultivars for different locations.

ANOVA has been used in many studies, and most of these studies have evaluated G x E of cane and sugar yields. Kennedy (1978) reported that G x E effects for cane and sugar yields in Barbados and Jamaica were small. In contrast, Tai et al. (1982) showed significant cultivar, cultivar x locations and cultivar x year interactions for yield. Similarly Parfitt (2005) reported significant site x genotype interaction for TCANE (304.2), ERC% (2.6) and TERC (8.6) of 28 genotypes.

Tahir et al. (2014) reported highly significant variances for environments, genotypes, and G x E interaction. The effect of environment was high for all the characters. Mean square differences were also significant for genotype, showing that none of the genotypes were the same across the three environments for all characters. Similar results have also been reported by Singh and Singh (1987), where they found significant mean squares for environments, genotypes and their interaction for various sugarcane characters.

Most G x E studies do not investigate the effects of lower level traits used in models on yield. Often the genotypes and environments used are too limited. Future research needs to focus on quantifying lower-level cultivar traits for different genotypes in different environments and evaluating the G x E interactions for such traits. Zhou et al. (2003) showed that these cultivar traits could be used to quantify coefficients used in crop models. The traits also offer potential as selection criteria that will improve identification of superior genotypes (Zhou, 2005).

2.5.2.2 Heritability

The knowledge of the extent to which sugarcane traits are heritable is of great importance for sugarcane breeding programmes. Determining the magnitude of parameters such as phenotypic variance, genetic variance, and heritability allows for the identification of traits that are effective for selection. Heritability is the ratio of the genetic variance to the phenotypic variance (Allard, 1960). There are two types of heritability; broad and narrow sense heritability. Narrow sense is the degree to which a trait is passed from parent to offspring expressed as the ratio of the additive genetic variance to the total phenotypic variance (V_A/V_P). Broad sense (H^2) heritability is the degree to which a trait is genetically determined, expressed as the ratio of the

total genetic variance to the phenotypic variance (V_G/V_P) (Bokmeyer, 2009). Broad sense heritability is thus relevant for individual genotype selection in sugarcane.

High broad sense heritability estimates were reported by Jamoza et al. (2014) for stalk diameter (92.8%), number of millable canes (91.2%), single stalk weight (90.7%), and number of internodes (90.7%). These results indicate that a large portion of total variance of these traits is heritable and trait selection would be effective. Kang et al. (1983) also reported high heritability estimates for plant height (84%), stalk diameter (94%), stalk weight (93%), and sucrose content (91%). However, Chavanne and Mariotti (1989) found moderate heritability estimates for stalk length (41%), stalk diameter (51%), and number of millable stalks (53%). Jamoza et al. (2014) further reported that the genetic variance exceeded the G x E variance components for all traits except cane yield and stalk height.

The most important function of heritability is its ability to predict how reliable a particular trait is for selection. The higher the heritability of a given trait, the less that trait is influenced by the environment. Thus, the higher the heritability of a trait, the greater will be its response to selection and genotypes can be selected based on the phenotype. To date, there have been no studies investigating the heritability of lower level phonological traits commonly used in crop growth models. Information on the heritability of these traits will help determine their future role in model-assisted breeding for sugarcane.

2.6 Summary

The Canegro sugarcane model (Singels and Bezuidenhout, 2002) simulates sugarcane crop growth and development from daily weather data, cultivar and soil properties, as well as management input data. Simulations in the Canegro model are based on the use of genetic (cultivar) coefficients. These are cultivar specific traits that interact with the environment to express a more complex trait like yield. Cultivar coefficients are cultivar specific values for a particular trait, and these vary from model to model.

The cultivar trait coefficients used in Canegro are based exclusively on research conducted on the cultivar NCo376. Values of trait coefficients for other contrasting cultivars are not available, and as a result, the Canegro model is unable to simulate performance of other contrasting cultivars. Being able to model cultivar differences will aid growers in choosing appropriate cultivars for specific growing conditions. The ability of crop models to quantify

and integrate responses to environmental and management factors can help study G x E interactions as sugarcane breeders often face significant G x E interactions in their trials grown under multiple environments. The presence of large G x E interaction has a negative effect on trait heritability. Hence, heritability needs to be determined for the different traits.

Limited studies have been conducted on various lower level traits used in crop models. The trait values studied are for a limited number of genotypes and studies were not conducted under contrasting environments. While the studies have examined various sugarcane lower level traits, G x E interactions of these traits has not been evaluated. The examination of the significance of G x E interaction in recently released genotypes is important both for grower choice of cultivars and for verification of breeding programme results.

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CHAPTER 3

General materials and methods

3.1 Cultivar and site information

Three cultivar trials were established in 2011 in areas representing the general growing conditions of the South African industry. The trials were conducted on SASRI research farms or commercial fields at Amatikulu (AK), Pongola (PG), and Bruynshill (BH). The Pongola site represented the irrigated region, the Amatikulu site represented the warmer coastal rainfed region, and the Bruynshill site represented the cooler rainfed midlands region. Table 3.1 shows the agro-ecological and meteorological data for the three sites.

The same set of 12 cultivars was tested at each of the three sites. These cultivars represented genotypes released in South Africa over the past 30 years, and which differed from each other in general growth and development characteristics (Table 3.2). Cultivar NCo376 is a standard cultivar used in the breeding programme in South Africa, for which there is extensive information on crop modelling traits.

Table 3.1 Agro-ecological and meteorological data for Amatikulu, Bruynshill and Pongola

	Year	Region		
		Amatikulu	**Bruynshill**	**Pongola**
Coordinates		29º1'0" S, 31º36'0" E	29º25'0" S, 30º41'0" E	27º24'0"S, 31º35'35" E
Elevation		93 m	990 m	308 m
Planting date		22 September 2011	25 October 2011	8 November 2011
Irrigation regime		Rainfed	Rainfed	Irrigated
Annual rainfall* (mm)	**2012**	1113.5	1000.3	851.7
	2013	1009.4	927.1	935.7
Annual temperature(max/min) (ºC)	**2012**	27.1/16.6	23.3/11.5	28/15.1
	2013	27.4/16.1	23.6/11.1	27.7/14.7
Mean annual rainfall (mm annum^{-1})**		933.8	760	731.6

*Total precipitation/rainfall received in the country

**Mean annual rainfall is the long term mean in depth (over space and time) of the annual total

Table 3.2 Characteristics of the 12 cultivars tested in cultivar trials at Amatikulu, Pongola, and Bruynshill

Cultivar	Characteristic
NCo376, N12	Established cultivars, good ratooning ability (RA*), high stalk population, average sucrose content
N31, N52	High biomass cultivars, low sucrose content, high stalk population, high fiber content
N25	Irrigated, high cane yield, low sucrose content, low fiber content
N19	Irrigated, low cane yield, high sucrose content, average fiber content
N36, N48	Low population, thick stalked cultivars, high sucrose content, lodging prone
N40, N35	Very high sucrose content, very low cane yield.
N41, N51	Average population, poor canopy, average sucrose content

* RA: The ability to maintain yields as the number of ratoon crops increase

3.2 Trial details

Trials were planted using randomised complete block designs with four replications and a total of 48 experimental units (Figure 3.1). Experimental plots comprised six rows of sugarcane spaced 1.4 m apart, eight rows spaced 1.2 m apart, and six rows spaced 1 m apart for the PG, AK and BH trials, respectively. Row length for the trials ranged from 8 to 10 m. Row spacing in each trial was according to the cultural practices of each location.

Plots were demarcated and sign-posted for clear identification of cultivar and plot number. Fertiliser application was according to the SASRI fertiliser advisory services recommendations. Fertiliser was applied twice, at planting and approximately 90 days after planting as topdressings, while hand weeding was done as required to control weeds.

Disease-free stalks produced in SASRI's farm nurseries were stripped of green and dead leaf material, topped and then laid in pairs (head to end) in the planting furrow and covered with

soil. Two crops, namely plant cane and first ratoon were harvested at approximately 12 months of age in the Pongola and Amatikulu trials. At Bruynshill (midlands), only the plant crop was harvested at 24 months of age. This is due to the slow growth rates brought about by low temperatures in the midlands region, where harvest maturity is only reached around 24 months of age.

41 **N40**	40 **N12**	25 **N48**	24 **N52**	9 **N31**	8 **N51**
42 **N48**	39 **N19**	26 **N12**	23 **N36**	10 **N41**	7 **N52**
43 **N25**	38 **NCo376**	27 **N40**	22 **N41**	11 **N36**	6 **N35**
44 **N52**	37 **N36**	28 **N35**	21 **N19**	12 **N40**	5 **N25**
45 **N41**	36 **N19**	29 **NCo376**	20 **N31**	13 **N12**	4 **N19**
46 **N31**	35 **N41**	30 **N52**	19 **N25**	14 **N51**	3 **NCo376**
47 **N35**	34 **N51**	31 **N31**	18 **N48**	15 **N35**	2 **N12**
48 **N51**	33 **N36**	32 **N25**	17 **N40**	16 **NCo376**	1 **N48**

Figure 3.1 The Amatikulu trial plan. Each colour represents a block (replicate) in the field and each block consists of the same 12 cultivars randomised within the block. Trials at other sites had similar trial plans

3.3 Measurements during the growing season

A total of six plants in 2 x 2 m sections were randomly selected and tagged from the net rows (Figure 3.2) as soon as possible after emergence. A series of measurements was done every two to three weeks on the tagged plants. We were hopeful that this frequency of measurements would be sufficient to accurately determine the crop's response in relation to thermal time and for use in crop modelling.

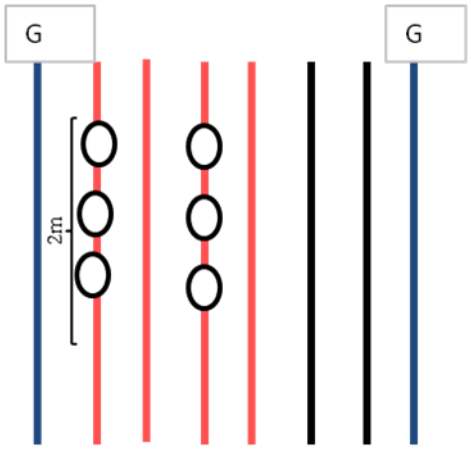

Figure 3.2 An illustration of a plot from the cultivar trials

Red lines show the net plot with the six selected stalks (white circles); navy lines represent the guard rows and the black lines represent the two rows used for destructive sampling

3.3.1 Tiller and leaf phenology

From the tagged plants in each plot, fully expanded leaves were numbered chronologically, from the bottom of the tiller upwards to the top visible dewlap (TVD) (Figure 3.3). Leaf width and length were measured with a tape measure for each of the numbered leaves on the selected plants on each occasion (Figure 3.4). The leaf width was measured at the midpoint of the leaf blade (Figure 3.4a). The leaf length was measured from the beginning of the leaf blade to the leaf tip (Figure 3.4b). The height of each numbered leaf was measured from the soil surface to each visible leaf dewlap on each occasion (Figure 3.4c). Stalk height was determined from the mean height of the six tagged plants in each plot. Tiller population was determined by counting the number of tillers in a previously identified 2-m section in each plot at two-week intervals from emergence until harvest.

3.3.2 Radiation interception by the canopy

A ceptometer (LP-80 AccuPAR - Decagon Devices) was used to measure PAR intercepted by green leaves between 10h00 and 12h00 on cloudless days. These measurements were taken every two to three weeks. At each plot, one reference reading was taken above the leaf canopy followed by 10 readings below the canopy. Readings were taken at 45 degree angles within the inter-row. The instrument was levelled before making a recording. Fractional intercepted PAR (FiPAR) was calculated as follows:

FiPAR = (above canopy PAR - below canopy PAR x I00)/above canopy PAR (Eq. 3.1)

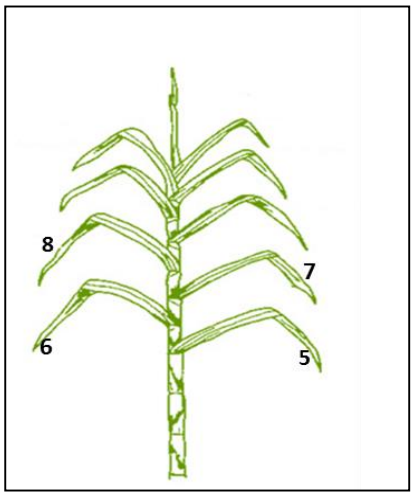

Figure 3.3 Leaf numbering of tagged plants (numbered chronologically, from the bottom up) (leaves 1-4 have senesced, therefore numbering starts st five)

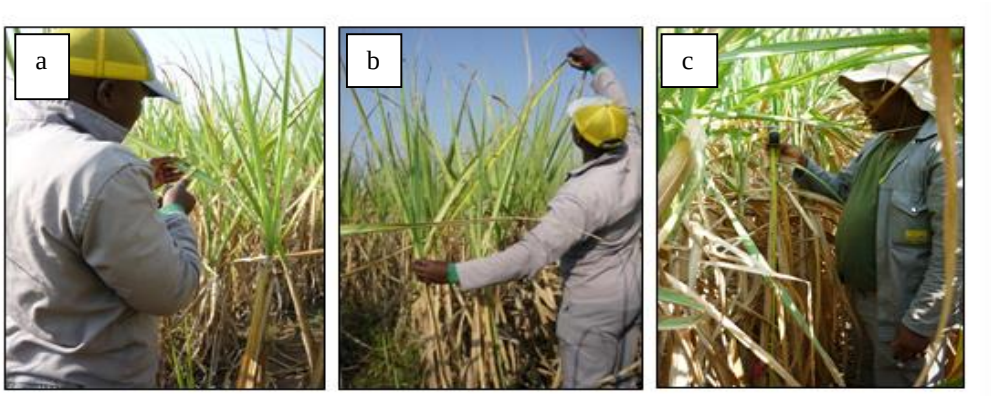

Figure 3.4 Illustrations of leaf width (a), leaf length (b) and stalk height (c) measurements respectively

3.3.3 Biomass accumulation

Destructive measurements of aboveground biomass were conducted at 3, 5, 7, 9 and 12 months after planting. These measurements were done by manually cutting cane stalks at the soil surface. In each plot, 12 (Amatikulu and Bruynshill) or 16 (Pongola) stalks were randomly selected from the rows reserved for destructive sampling (Figure 3.2). These stalks were partitioned into green leaves, trash (dead leaves), meristem and stalk (Figure 3.5). These

partitions were weighed to determine fresh mass. Each stalk sample was evaluated for stalk diameter (mm), height (cm), and mass (kg). In order to estimate biomass yields at each destructive sampling date, stalk population was also determined by counting millable stalks over the 8 or 10 m rows in each plot.

Figure 3.5 Destructive components during biomass partitioning (left to right: trash (dead leaves), stalks, meristem, and green leaves)

3.3.4 Cane and sucrose yield determinations

During harvest, three or four net rows were harvested by hand and weighed using a mechanical grab fitted with a load cell (Figure 3.6). A 12 or 16-stalk sample was taken from each plot at each harvest. Quality analysis was done on sub-samples using standard procedures developed at SASRI to give an indication of the percentage sucrose and other quality parameters such as the juice Brix and purity (Meyer and Clowes, 2011). Estimated recoverable crystal (clumps of sugar formed on a base after evaporation of a sugar-saturated solution) percent (ERC%) of cane expressed as a percentage on a fresh mass basis was chosen as the measure of cane quality. The ERC% was calculated as follows:

$$\text{ERC\% cane} = aS - bN - cF \qquad \text{(Eq. 3.2)}$$

Where:

- ERC% cane is the estimated quantity of crystal which can be recovered from the incoming cane supply (expressed as crystal % cane),
- S is the sucrose % cane,
- N is the non-sucrose % cane (calculated as brix % cane minus sucrose % cane),

- F is the fibre % cane and
- a, b, and c are constant parameters related to the sucrose losses within the factory. The values of the three constant parameters in the ERC formula represent the sucrose losses in the form of product sugar, bagasse or final molasses.

Figure 3.6 A mechanical grab fitted with a load, used to determine cane yields during harvest

3.4 Traits calculated from measurements

The ability of simulation models to predict growth and development is dependent on the use of genetic (cultivar) traits. These crop characters summarise quantitatively how a genotype responds to environmental factors. The high frequency of measurements was used to calculate some of these specific traits used in the Canegro model. Some of these traits are shown in Table 3.3 and described briefly below.

The cultivar traits used in Canegro are simulated once a certain amount of thermal time units have accumulated. Thermal time is a common temperature property affecting respiration, photosynthesis, leaf properties (such as leaf elongation, leaf appearance rate, and width), and shoot population. Thermal time is defined as the sum of all the positive values of the mean daily temperatures minus the base temperature (Equation 3.3). Base temperature is the temperature below which the plant process of interest does not progress.

$$TT = (\Sigma(Tmax + Tmin)/2) - Tbase \qquad \text{(Eq. 3.3)}$$

Table 3.3 Description of cultivar traits used in the Canegro model

Parameter name	Description	Units
PTP	Peak tiller population	Stalks ha^{-1}
TTPP	Thermal time required to reach peak tiller population	^{o}C days
FPOP	Final tiller population at harvest	Stalks ha^{-1}
TSP	Tiller survival percentage. The number of millable tillers at harvest expressed as a percentage of the peak tiller population	percentage
SER	Stalk elongation rate	mm $(^{o}Cd)^{-1}$
LAmax	Max leaf area assigned to all leaves	cm^2
TTLAmax	Thermal time required to reach max leaf area	^{o}C days
LFmax	Maximum number of green leaves	Number
GLAI	Green leaf area index	Number
LAR	Leaf appearance rate	Leaves $(^{o}Cd^{-1})$
Total biomass yield	Total biomass	g unit $area^{-1}$
RUE	Radiation use efficiency	g MJ^{*-1}
FiPAR	Fractional interception of photosynthetically active radiation	Number

*MJ = megajoule

3.4.1 Tiller and stalk traits

The PTP was determined as the maximum number of tillers produced when tiller numbers (per hectare) were plotted against thermal time for the 12 cultivars. The PTP was determined as the peak of the tiller population curve. The TTPP was determined as the thermal time in degree days required to reach PTP. The FPOP was determined as the tiller population at harvest, and the TSP was calculated as the FPOP divided by the PTP. The SER was determined from the fitted straight line-equation of the graph of accumulated thermal time on the x-axis and stalk height (mm) on the y-axis. The SER was the coefficient of *x*.

3.4.2 Leaf traits

Leaf area per stalk was determined by summing the product of leaf width, length and a shape factor of 0.7. LAmax was determined as the maximum leaf area when the sum of leaf area per plant was plotted against thermal time. The TTLAmax was determined as the degree days required to reach LAmax. Leaf area was critical in determining leaf area index; which was calculated from the measured mean leaf area divided by the plot area. The LAR was determined from the fitted straight line-equation of the graph of accumulated thermal time versus leaf number. The LAR was the coefficient of *x*. LFmax was determined as the maximum leaf number from the graph.

3.4.3 Biomass accumulation and partitioning

The above ground biomass in sugarcane mainly consists of the stalk and the leaf components. At each sampling date, the biomass partitioning of each cultivar was determined. The cane plant was partitioned into green leaves, trash, meristem, and stalks. The samples were weighed (to determine fresh mass) and dried for 24 hours before weighing again (to determine the dry mass). Total biomass was determined as the sum of green leaf material, brown leaf material, meristem and stalk weight (g stalk^{-1}). The biomass determined from fresh mass (g stalk^{-1}) at each sampling date was then plotted against accumulated thermal time to study the biomass accumulation patterns of the different cultivars.

3.4.4 Radiation use efficiency

The PAR measurements from the ceptometer taken below and above the canopy were used to calculate the fraction of PAR intercepted. The fractional intercepted radiation (FiPAR) was calculated from Equation 3.1. RUE was determined for each sampling date by dividing FiPAR by the biomass yield for that date.

3.5 Data processing and statistical analysis

Traits (Table 3.3) used for statistical analyses were obtained from the mean of six plants. A combined ANOVA was performed using Genstat for AK and PG where main and interaction effects were tested. The main effects were cultivar, site, and crop cycle. The trial at BH was only harvested for the plant crop. Therefore, a separate ANOVA was also conducted for BH, AK and PG to test the main effects of site and cultivar for the plant crop only.

A comparison of means for the different traits was performed using Duncan's protected least significant difference (LSD) test at 95% significance. Broad sense heritability estimates and variance components were calculated for all studied traits. Cultivar rank correlations across sites and ratoons were determined using Spearmans correlation in Genstat to see if cultivars were consistent across sites or crops and to see if cultivars can be characterised from single-site and single-crop experiments in the future.

3.6 Heritability estimates

Broad-sense heritability for each trait was calculated from the components of variances which were determined from the mean squares from the ANOVA (Table 3.4).

Table 3.4 Analysis of variance with mean squares to calculate heritability

Source	**Mean Square**
Cultivar (C)	M5
Site (S)	
Crop cycle (R)	
(C x S)	M4
(C x R)	
(R x S)	M3
(C x S x R)	M2
Residual	M1

Broad sense heritability was determined from the equation: $h^2 = \sigma^2 g / \sigma^2 p$:

Where $\sigma^2 g$ = (M5 + M2 –M3 – M4)/replication (rep) x sites (s) x ratoons (r)

And $\sigma^2 p = \sigma^2 g + \sigma^2 gs/s + \sigma^2 gr/r + \sigma^2 gsr/sr + \sigma^2 e$/rep x s x r

- $\sigma^2 gs$ = M4 –M2/rep x r
- $\sigma^2 gr$ = M3- M2/rep x s
- $\sigma^2 gsr$ =M2 –M1/rep
- $\sigma^2 e$ = M1

$\sigma^2 g$ = the genetic variance

$\sigma^2 p$ = the phenotypic variance

$\sigma^2 gs$ = variance of cultivar x site interaction

$\sigma^2 gr$ = variance of cultivar x ratoon interaction

$\sigma^2 gsr$ = variance of cultivar x site x ratoon interaction

$\sigma^2 e$ = the environmental variance

replications = number of reps (4)

s = number of sites (2)

r = number of crop cycles (2)

CHAPTER 4

Quantifying sugarcane cultivar differences and genotype x environment interactions for tiller and stalk phenology traits

Abstract

The cultivar trait values in the Canegro sugarcane model are normally determined from field experiments through sampling of growth and development data for each cultivar at regular intervals. There have not been comprehensive characterisations of South African cultivars for tillering and stalk elongation traits across multiple sites and seasons. Incorporating these quantified trait values into the Canegro crop model would refine its ability to simulate cultivar growth differences across environments and thereby contribute to model-assisted breeding. The primary objective of this study was to quantify the tillering and stalk elongation traits for a diverse range of South African sugarcane cultivars across contrasting environments and over different crop cycles (plant and ratoon crops). A secondary objective was to determine the general stability and heritability of the traits to assess their future use in model-assisted breeding.

Three cultivar trials were established in 2011 at three sites on SASRI research farms and commercial fields; AK, PG and BH. The tiller population and stalk elongation traits determined for each cultivar were PTP, TTPP, FPOP, TSP and SER. The stability of each trait was assessed through cultivar rank correlations between sites and crops. Broad sense heritability (H^2) was determined for these traits from the variance components obtained from the analysis of variance.

The trait PTP was more stable across sites in the ratoon crop compared with the plant crop. The trait was also more stable across crops under irrigated conditions compared with rainfed conditions. Except for cultivar N12, which showed consistently high TTPP (slow growth rate), this trait was generally not stable across sites and crop cycles. The trait FPOP showed strong rank correlations between sites and crop cycles, suggesting that it is a stable trait with potential for use in model-assisted breeding. The cultivars N12 and NCo376 (high FPOP), and N36 and N48 (low FPOP) were identified as ideal indicator cultivars for future studies. The trait TSP was relatively stable across sites for the plant and ratoon crop. This trait was more stable across crops under rainfed conditions compared with irrigated conditions. The trait SER was the most

stable trait of all, showing strong rank correlations between sites and across ratoons within sites. Cultivars NCo376 and N12 were identified as ideal indicator cultivars for low SER, while cultivars N31 and N52 were identified has high SER cultivars.

Heritability (H^2) estimates ranged from 0.49 for TTPP to 0.97 for FPOP. The cultivar x ratoon, cultivar x site, and cultivar x site x ratoon interaction components had less influence on all traits than the genotypic and environmental variances. Environmental variances were higher than genotypic variances for all traits. The high H^2 suggests that selection for PTP, SER and FPOP would be most effective. The information gathered on trait stability, trait range values, heritability, and indicator cultivars will be used to design future trials involving cultivar characterisation. The results also provide insight into the use of different traits for model-assisted breeding.

4.1 Introduction

Following planting of sugarcane setts, the buds emerge to form primary stalks from which secondary and tertiary tillers develop. This process is called tillering and results in the formation of a number of tillers which eventually form mature stalks of a stool (van Dillewijn, 1952). Tillering is normally completed four months after planting (Peng, 1984), but varies depending on growing conditions and cultivar. In the initial stages of tillering, tiller number increases exponentially, reaching a maximum tiller population between 90-120 days after planting. Only a portion of those tillers formed actually develop into mature cane stalks that contribute to final yield. Competition between tillers for water, nutrients and light results in substantial tiller mortality. Tiller mortality usually stabilizes 150 days after planting to form millable cane (final tiller population).

The rate of tillering, peak tiller number, and final tiller number (collectively known as population dynamics) are largely genetically controlled. These dynamics can also be influenced by other factors such as light, temperature, soil moisture (irrigation), row spacing, and practices of fertilization. Tillering provides the crop with an appropriate number of stalks required for a good yield. According to Raman et al. (1985) and Javed et al. (2000), stalk number is the major contributing factor to cane yield. In crop growth models, tiller population dynamics are used to model leaf area index (LAI) and light interception (LI). In the Canegro model, tillering is defined as the period from shoot emergence to the occurrence of peak tiller population and start

of tiller senescence (Singels et al., 2008). This phase is completed when a cultivar specific thermal time (average daily temperature minus a base temperature below which no growth occurs) has accumulated. Genetic traits used to simulate tiller population dynamics in the Canegro model include the TTPP, PTP, FPOP and TSP.

A handful of studies have investigated cultivar differences in population dynamics. Significant differences in PTP were observed between cultivars NCo376 and N26 under irrigation in Pongola (Donaldson, 2011). Also, in cane grown in the south east lowveld of Zimbabwe, Zhou (2003) observed significant differences in PTP among cultivars ZN6 (150 700 stalks ha^{-1}), ZN7 (182 700 stalks ha^{-1}), N14 (248 000 stalks ha^{-1}), and NCo376 (265 300 stalks ha^{-1}). This shows that PTP is cultivar dependent.

Shukla and Singh (2011) reported variable behaviour in FPOP of three different cultivars (CoS 96269, CoPant 97222, and CoLk 9616) grown in subtropical India. They found that cultivar CoLk 9616 produced a significantly higher FPOP of 119 400 stalks ha^{-1} compared with cultivars CoPant 97222 (93 940 stalks ha^{-1}) and CoS 96269 (100 250 stalks ha^{-1}). Rafiq and Sattar (2013) showed that the widely adapted cultivar HSF-240 produced significantly higher FPOP (147 650 stalks ha^{-1}) in both years compared with cultivar Q-88 (44 350 stalks ha^{-1}). Zhou (2003), Shukla and Singh (2011), Donaldson (2011), and Rafiq and Sattar (2013) showed that although there were differences between cultivars in tillering characteristics and traits such as PTP and FPOP, these traits are also influenced by growing season and environmental conditions. Despite past studies, there has not been a comprehensive characterisation of South African cultivars for tillering traits across multiple sites and seasons.

Stalk elongation follows soon after tiller emergence and continues until harvest. Tillers generally elongate more rapidly during the early stages of growth, slowing down as the crop canopy closes and cane reaches maturity. Stalk elongation varies with cultivar and growing conditions. Some sugarcane cultivars have a short-lived tillering stage which allows stalk elongation to commence early, while other cultivars tiller for longer periods. The former usually have fewer stalks at harvest than the latter. Factors that affect stalk elongation include temperature and soil moisture. Several authors including Inman-Bamber and Smith, (2005) reported sugarcane stalk elongation to be negatively and strongly affected by drought. Some studies have reported a strong relationship between air temperature and stalk elongation (Inman-Bamber, 1991; 1994; Koonjah et al., 2006).

Due to this strong correlation between stalk elongation and air temperature, crop models simulate SER as a function of thermal time (Inman-Bamber, 1991; 1994). In the Canegro model, stalk elongation is completed once a given amount of thermal time units have accumulated (^{o}Cd) (Singels et al., 2008). In sugarcane, it has been shown that SER per unit thermal time is genetically determined to a large extent (Smit and Singels, 2007). When stalk height data was plotted against days after planting and accumulated thermal time, Khan et al. (2013) reported highly significant differences amongst cultivars; cultivars NIA-2010 and NIA0819/P5 produced significantly higher stalk height followed by NIA-2004. Smit and Singels (2007) also reported that cultivar N31 had a significantly higher SER of 1.82 mm (^{o}Cd)$^{-1}$ than N37 (1.48 mm (^{o}Cd)$^{-1}$) and NCo376 (1.36 mm (^{o}Cd)$^{-1}$). In Nigeria, Ethan et al. (2013) compared growth of three sugarcane cultivars, NCS 002, NCS 003 and Co 957 under irrigated conditions. They found that cultivar NCS 002 had taller stalks (99.9 cm) than cultivars Co 957 (91.61 cm) and NCS 003 (81.97 cm). As with stalk population dynamics, quantifying differences in SER for different cultivars across sites and environments has not been attempted.

Stalk number, stalk length, stalk diameter, and stalk weight have been found to be highly correlated to cane yield (Hooda et al., 1979; Punia et al., 1983. The use of these traits for indirect selection for higher yields has been suggested (Yahaya et al., 2009). The knowledge of heritability estimates of these yield components allow for genetic improvement and for selection of the best genotypes. For instance, Kang et al. (1983) estimated high broad sense heritability in two bi-parental crosses for plant height (84%), stalk diameter (94%), stalk weight (93%) and sucrose content (91%). Ghura (2005) also found that broad sense heritability ranged from 90% for stalk diameter and sucrose percentage to 78% for purity. These results showed that the traits studied can be improved through selection. Despite the discussion of H^2 for these traits, little attention has been devoted to lower level traits (such as TTPP, TSP, FPOP and SER). H^2 information for lower level traits might help contribute to model-assisted breeding in future. Broad-sense heritability (sometimes referred to as genetic repeatability) is the degree to which the phenotype is determined by its genotype (Toghiani, 2012).

The primary objective of this study was to quantify for a diverse group of South African sugarcane cultivars their FPOP, TTPP, PTP, TSP, and SER, traits related to tillering and stalk elongation. These quantified trait values will be submitted to the Canegro crop model for evaluation to determine if their incorporation into the model will refine its ability to simulate cultivar growth differences across environments. The determination of realistic range values

for these traits will also contribute to the ability to conduct a model-wise exploration of G x E interactions. An additional objective was to determine the cultivar stability and heritability for these traits across environments and crop cycles to determine their potential contribution to future model-assisted breeding. Higher stability of cultivars across sites and crop cycles would suggest that these traits could be accurately evaluated in experiments conducted across fewer site and crop cycles in the future.

4.2 Material and methods

Full details of all trials, including site characteristics, cultivars, experimental designs, plot dimensions, and crop characteristics were described in detail in Chapter 3. For brevity, only the details of the data collection and analysis relevant to this chapter are briefly mentioned below.

From emergence stalk height and tiller population measurements were conducted every two to three weeks on the tagged plants. Stalk height was determined from the mean height of the six tagged plants in each plot. Tiller population was determined by counting the number of tillers in the 2-m sections in each plot at two-week intervals from emergence until harvest. This high frequency of measurements was needed to help ensure accurate determination of the cultivars' responses in relation to thermal time and for use in crop modelling.

The tiller population and stalk elongation parameters were determined for each cultivar. The PTP was determined as the maximum number of tillers produced when tiller numbers (per hectare) were plotted against thermal time. The TTPP was determined as the thermal time in degree days required to reach PTP. The FPOP was determined as the tiller population at harvest, and the TSP was calculated as the FPOP divided by the PTP. The SER was determined from the fitted straight line-equation of the graph of accumulated thermal time on the x-axis and stalk height (mm) on the y-axis. The SER was the coefficient of *x*.

The data for the parameters was analysed using Genstat version 14 (VSN International Ltd, 2011). A combined ANOVA was performed for AK and PG where main and interaction effects were tested. The main effects were cultivar, site, and crop cycle. The trial at BH was harvested on a 24-month cutting cycle, and as a result, was only harvested for the plant crop. A separate ANOVA was therefore also conducted for BH, AK, and PG to test the main effects of site and cultivar for the plant crop only. Treatment mean comparisons were made using Duncan's LSD

test at P=0.05. The stability of each trait was assessed through cultivar rank correlations between sites and crops. The variance components were calculated using the mean squares from the ANOVA. Estimates of broad sense heritability for the different traits were calculated using the variance components method. Broad sense heritability was determined from the equation: $h^2 = \sigma^2 g / \sigma^2 p$, as shown in Chapter 3.

4.3 Results and discussion

4.3.1 Tiller population dynamics

When tiller population was plotted against accumulated thermal time, similar trends were observed for all cultivars in all three sites. Examples of these tillering patterns are shown for the plant crops of cultivars NCo376 (Figure 4.1), N36 (Figure 4.2), and N48 (Figure 4.3). In general, the tillers appeared rapidly in the early part of the season and reached a peak during the season. Thereafter, tillers died, and numbers decline to a relatively stable population that is maintained until harvest. A distinct trend was that higher PTP and FPOP was reached at BH compared with the PG and AK sites, respectively.

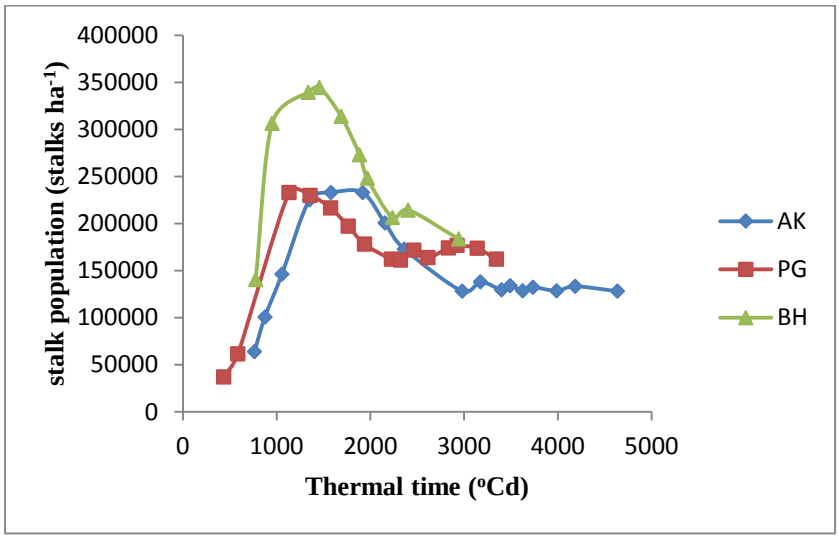

Figure 4.1 Tillering patterns of cultivar NCo376 in BH, AK and PG as a function of accumulated thermal time (base temperature 10°C)

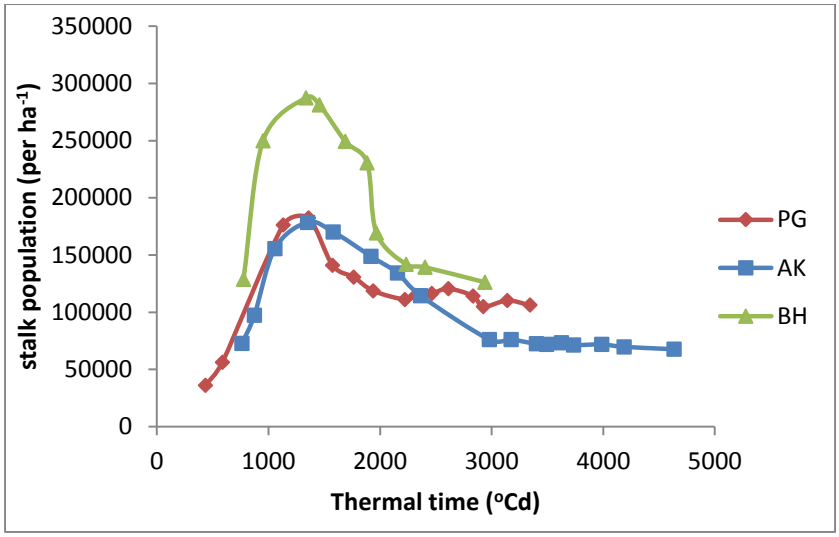

Figure 4.2 Tillering patterns of cultivar N36 in BH, AK and PG as a function of accumulated thermal time (base temperature 10°C)

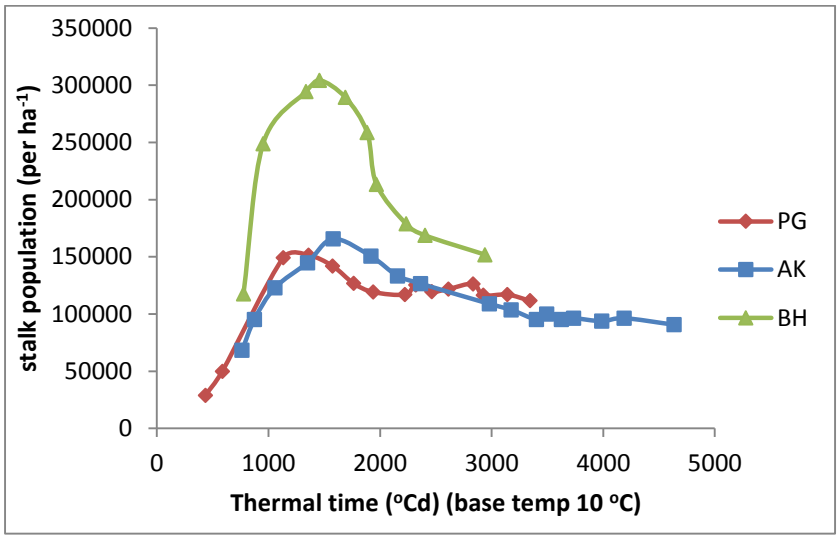

Figure 4.3 Tillering patterns of cultivar N48 in BH, AK and PG as a function of accumulated thermal time (base temperature 10°C)

Table 4.1 shows mean square values from the combined ANOVA of the PG and AK trials for both the plant and first ratoon crops. The mean square values were highly significant ($P<0.01$) for the main effects of site (S), crop cycle (R), and cultivar (C) for most traits (Table 4.1). The effect of ratoon on FPOP was significant at $P<0.05$ and the effect of site on TTPP was non-significant. The mean square for R was higher than the mean square for S and C for PTP, TTPP, and TSP. Therefore R had the largest effect on variation for these traits. The mean square for

S was high for FPOP. These results show that the main effects of R and S contributed most to total variance while C had a lower contribution for all traits.

The cultivar x site (C x S) interaction was significant (P<0.01) for PTP and significant at P<0.05 for TTPP and TSP (Table 4.1). This indicated that varying relative responses of cultivars to site occurred for these three traits. The crop cycle x site (R x S) interaction was highly significant for all traits analysed. The variance due to cultivar x crop cycle (C x R) was not significant for PTP, TTPP, and FPOP, showing that the relative cultivar values for these traits did not vary across crop cycles. The three-way C x S x R interactions were significant (P<0.05) for PTP and FPOP, showing that the relative cultivar values for these traits varied from one site to the next, and from the plant to the ratoon crop. The order of importance of sources of variation was:

R x S >R > S > C > C x S> C x S x R and C x R for PTP;

R > R x S > C > C x S > C x S x R > C x R > S for TTPP;

S > R x S > C > R > C x S x R > C x S > C x R for FPOP;

and R x S > R > S > C > C x R > C x S = C x S x R for TSP.

Table 4.1 Mean square values from analysis of variance, combined across the plant and first ratoon crop cycles and sites (Pongola and Amatikulu) for peak tiller population (PTP), thermal time to peak tiller population (TTPP), final tiller population (FPOP), and tiller survival percentage (TSP)

Source	d.f.	Mean square values			
		PTP ($x10^{6}$)	TTPP	FPOP (x 10^{5})	TSP
		Stalks ha^{-1}	^{o}Cd	Stalks ha^{-1}	%
Crop cycle (R)	1	176300**	11554861**	14210*	0.61**
Site (S)	1	55300**	66507	543800**	0.50**
Cultivar (C)	11	16650**	521098**	33900**	0.06**
R x S	1	328600**	1801059**	45520**	0.89**
C x R	11	2928	145931	2907	0.02**
C x S	11	5720**	282991*	3964	0.01*
C x S x R	11	3572*	161861	5716*	0.01

**, *; Significant at P<0.01 and P<0.05, respectively.

Table 4.2 shows the mean square values from ANOVA of the PG, AK, and BH plant crop only. The main effects (C and S) mean squares were highly significant (P<0.01) for all traits (Table 4.2). The mean square values were highly significant for the C x S interaction for PTP (P<0.01)

and significant for TTPP, FPOP and TSP (P<0.05). Some cultivars were affected by site for the plant crop and more analysis is needed to determine which specific cultivars were most affected. The main effects (C and S) mean squares exceeded the C x S interaction mean squares; indicating that the main effects of C and S each accounted for more variation than the C x S interaction.

Table 4.2 Mean square values from analysis of variance for the plant crop, combined across sites (Bruynshill, Pongola, and Amatikulu) for peak tiller population (PTP), thermal time to peak tiller population (TTPP), final tiller population (FPOP), and tiller survival percentage (TSP)

Source	d.f.	Mean square values			
		PTP (x 10^6)	TTPP	FPOP (x 10^6)	TSP
		Stalks ha^{-1}	^oCd	Stalks ha^{-1}	%
Cultivar (C)	11	11830**	199732**	4825**	0.04**
Site (S)	2	267100**	854231**	51090**	0.75**
C x S	22	3889**	88252*	581*	0.01*

**, *; Significant at P<0.01 and P<0.05, respectively.

4.3.1.1 Peak tiller population (PTP)

In the plant crop, the mean PTP was highest at BH, followed by AK and PG (Table 4.3a). The crop at BH had a longer growth period (24 months) compared with the crops at AK and PG (12 months) and this could be the reason for the higher numbers of tillers produced. These results highlight the influence of environment on sugarcane population. The cultivars NCo376, and N12 had the highest PTP in PG. Similarly, at BH, N12, and N52 also had the highest PTP, and at AK, N35, N31 and N40 also had the highest PTP. Average PTP ranged from 153 571 to 238 839 stalks ha^{-1} for PG, 272 500 to 431 250 stalks ha^{-1} for BH and 177 083 to 335 417 stalks ha^{-1} for AK. The differences in PTP across cultivars in all three sites for the plant crop was significant (P<0.01). The cultivar N12 had high PTP at PG and BH but had its PTP was low at AK. In contrast, cultivar N19 ranked high in AK and had a low PTP in PG and BH. Although there was switching of ranks for some cultivars across the AK and PG sites in the plant crop, the moderate (r=0.50) correlation between the two sites (Table 4.3b) meant that the general cultivar ranking was fairly consistent. Despite the significant C x S interaction (Table 4.2) in the plant crop, the relative rankings of some cultivars were fairly consistent across sites e.g. N31, N36, N48, and N52. This suggests that ranking of cultivars into low, moderate, and high

range values for PTP may be a possibility, but researchers should expect that PTP of some cultivars will be affected by location.

In the ratoon crop, PG had a higher mean PTP than AK, which was in contrast to the response observed in the plant crop. The mean PTP varied from 153 646 to 289 063 stalks ha^{-1} in AK and 247 321 to 441 964 stalks ha^{-1} in PG. The cultivar N41 ranked high in PG but had a low ranking in AK, while cultivar N40 ranked high in AK and had a low PTP in PG. Cultivars NCo376 and N35 ranked high in AK and PG, while cultivars N36 and N48 were ranked low at both sites. The correlation coefficient was moderate ($r=0.58$) and significant ($P<0.05$) across sites for the ratoon crop (Table 4.3b), showing that PTP may be more stable in the ratoon crop than in the plant crop.

Within a site, it was generally found that cultivar rank correlations were strong across crops at PG, but not as strong at AK. For example, cultivars N19, N25, N40, N48, N51, and NCo376 were ranked similarly in the plant and ratoon crops at PG. This is further enhanced by the moderately significant ($P<0.05$) correlation coefficient of 0.57 at PG across crop cycles (Table 4.3b). This was in contrast to the weak and non-significant correlation value of 0.41 at AK across ratoons. The weak relationship between cultivar rankings across crops at AK meant that cultivar characterisation for PTP in ratoons cannot be adequately represented from information gathered in the plant crop only. In contrast, under irrigated conditions (PG), cultivar characterisation in the plant crop could represent the ratoon crop as well for future experiments.

Table 4.3a) Mean peak tiller population (PTP) (stalks ha^{-1}) of 12 cultivars at Bruynshill (BH), Amatikulu (AK) and Pongola (PG) for the plant and first ratoon crop cycles

	Plant crop			First ratoon	
Cultivar	**AK**	**PG**	**BH**	**AK**	**PG**
N12	209896 abc (8)	206250 cd (2)	431250 f (1)	222396 bcd (5)	312500 ab (7)
N19	248438 c (4)	176786 abc (8)	306250 abc (8)	181250 ab (11)	294643 ab (9)
N25	221354 abc (7)	163839 ab (10)	323750 abc (6)	204688 abcd (7)	285268 ab (11)
N31	335417 e (1)	201339 bc (3)	347500 cde (3)	289063 e (1)	328571 abc (5)
N35	241146 bc (6)	198661 bc (4)	323125 abc (7)	244271 de (2)	411161 cd (2)
N36	178646 a (11)	183482 abc (6)	290625 ab (11)	153646 a (12)	286607 ab (10)
N40	190104 ab (10)	172768 abc (9)	272500 a (12)	238542 cde (3)	299554 ab (8)
N41	202604 abc (9)	153571 a (12)	299375 abc (10)	185938 abc (9)	350000 bc (4)
N48	177083 a (12)	154464 a (11)	305625 abc (9)	184375 abc (10)	247321 a (12)
N51	255729 cd (3)	180357 abc (7)	331875 bc (5)	196354 abcd (8)	321429 ab (6)
N52	299479 de (2)	186161 abc (5)	395625 df (2)	208333 abcd (6)	357589 bc (3)
NCo376	242187 bc (5)	238839 d (1)	346875 cd (4)	227604 bcd (4)	441964 d (1)
Mean	**233507**	**184710**	**331198**	**211372**	**328051**
§	**b**	**a**	**c**		
§§	**c**	**a**		**b**	**d**

Means followed by the same letters do not differ significantly within a site and crop cycle. Numbers in brackets indicate ranking of cultivars (1 representing a cultivar with the highest PTP, while 12 represents a cultivar with the lowest PTP)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 4.3b) Cultivar rank correlation coefficients for PTP at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.57*	1		
AK P	0.50	0.55	1	
AK R	0.55	0.58*	0.41	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, *; Significant at P<0.01 and P<0.05, respectively.

4.3.1.2 Thermal time to peak population (TTPP)

In the plant crop, the mean TTPP in BH and PG was lower than at AK (Table 4.4a). The TTTP was probably higher at AK due to the rainfed conditions; the lack of water may have caused slower tiller development. A range of cultivars including the cultivar N31 (1333 ^{o}Cd) and

NCo376 (1189 ^{o}Cd), in AK and PG respectively, reached their peak more rapidly than the other cultivars, i.e. they had low TTPP values. The TTPP varied from 2099 to 1333 ^{o}Cd at AK, 1713 to 1189 ^{o}Cd at PG and 1747 to 1201 ^{o}Cd at BH. A low (r=-0.04) and non-significant rank correlation was observed between AK and PG for the plant crop (Table 4.4b).

In the ratoon crop, PG had a higher mean TTPP than AK, which was in contrast to the response observed in the plant crop. In the ratoon crop, cultivar N12 recorded the highest TTPP at AK. The TTPP values ranged from 486 to 2106 ^{o}Cd in AK and 994 to 1251 ^{o}Cd in PG. The mean TTPP obtained by the ratoon crop was lower than the plant crop in both sites. This shows that ratoon crops generally reach PTP quicker than plant crops. The rank correlation coefficient between AK and PG for the ratoon crop was 0.36, indicating a weak relationship between the cultivars rankings (Table 4.4b). This shows that TTPP may not be stable across sites in the ratoon crop as well.

Within sites, the TTPP values were similar for cultivars N12, N19, N25, N31, N41, N48, N51, and NCo376 between the plant and ratoon crop at AK (Table 4.4a). Similarly cultivars N12, N31, N35, N40, N41, and N48 had similar ranking in PG in the plant and ratoon crop (Table 4.4a). Despite these similar rankings, non-significant and weak rank correlations at AK (r=0.34) and PG (r=0.28) were observed. This means that a single trait value per cultivar may not be adequate to represent TTPP across ratoons. This weak correlation may be due to the response of cultivars such as N52, which showed very different rankings between the plant and ratoon crop at both AK and PG. The difference in responses may be linked to the cultivar's differential reaction to environmental conditions at the two sites and shows that the rate of tillering of N52 may be dependent on seasonal or crop (plant or ratoon) effects. On the other hand, cultivar N12 was consistent and stable across sites and crop cycles suggesting that breeding programs should be able to identify stable cultivars for TTPP if this is a high priority.

Table 4.4a) Means for thermal time to peak population (TTPP) (ºCd) for 12 cultivars at Amatikulu (AK), Pongola (PG) and Bruynshill (BH) for plant crop and first ratoon

	Plant crop			First ratoon	
Cultivar	**AK**	**PG**	**BH**	**AK**	**PG**
N12	2099 $^{c\ (1)}$	1713 $^{c\ (1)}$	1573 $^{bc\ (2)}$	2106 $^{b\ (1)}$	1251 $^{b\ (1)}$
N19	1464 $^{ab\ (10)}$	1669 $^{bc\ (2)}$	1366 $^{ab\ (8)}$	486 $^{a\ (12)}$	1065 $^{ab\ (9)}$
N25	1633 $^{ab\ (5)}$	1302 $^{ab\ (7)}$	1239 $^{a\ (11)}$	782 $^{a\ (6)}$	1149 $^{ab\ (2)}$
N31	1333 $^{a\ (12)}$	1302 $^{ab\ (7)}$	1201 $^{a\ (12)}$	510 $^{a\ (11)}$	1097 $^{ab\ (5)}$
N35	1749 $^{abc\ (3)}$	1245 $^{a\ (9)}$	1424 $^{ab\ (3)}$	620 $^{a\ (10)}$	994 $^{a\ (12)}$
N36	1406 $^{ab\ (11)}$	1245 $^{a\ (9)}$	1396 $^{ab\ (4)}$	1182 $^{a\ (3)}$	1149 $^{ab\ (2)}$
N40	1834 $^{bc\ (2)}$	1245 $^{a\ (9)}$	1270 $^{a\ (10)}$	782 $^{a\ (6)}$	1014 $^{a\ (11)}$
N41	1607 $^{ab\ (6)}$	1464 $^{abc\ (5)}$	1396 $^{ab\ (4)}$	722 $^{a\ (8)}$	1097 $^{ab\ (5)}$
N48	1534 $^{ab\ (8)}$	1467 $^{abc\ (3)}$	1388 $^{ab\ (7)}$	722 $^{a\ (8)}$	1097 $^{ab\ (5)}$
N51	1607 $^{ab\ (6)}$	1457 $^{abc\ (6)}$	1747 $^{c\ (1)}$	1049 $^{a\ (5)}$	1149 $^{ab\ (2)}$
N52	1522 $^{ab\ (9)}$	1467 $^{abc\ (3)}$	1366 $^{ab\ (8)}$	1285 $^{a\ (2)}$	1045 $^{a\ (10)}$
NCo376	1749 $^{abc\ (3)}$	1189 $^{a\ (12)}$	1396 $^{ab\ (4)}$	1078 $^{a\ (4)}$	1097 $^{ab\ (5)}$
Mean	**1628**	**1397**	**1397**	**944**	**1100**
§	**b**	**a**	**a**		
§§	**d**	**c**		**a**	**b**

Means followed by the same letters do not differ significantly within a site and crop cycle. Numbers in brackets indicate ranking of cultivars (1 representing the cultivar with the highest TTPP, while 12 represents the cultivar with the lowest TTPP)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 4.4b) Cultivar rank correlation coefficients for TTPP at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.28	1		
AK P	-0.04	-0.11	1	
AK R	0.11	0.36	0.34	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, *; Significant at P<0.01 and P<0.05, respectively.

4.3.1.3 Final tiller population (FPOP)

In the plant crop, BH produced the highest mean FPOP, followed by PG and AK, respectively (Table 4.5a). The higher FPOP at PG was attributed to the ample water supply compared to AK, where there was moisture stress. The FPOP ranges for AK, PG and BH were 67 708 to 133 333, 104 911 to 162 946 and 126 250 to 234 375 stalks ha^{-1}, respectively. Cultivar N12 had the highest FPOP at PG and BH in the plant crop. There was a moderate (r=0.61) and significant (P<0.05) cultivar rank correlation for the AK plant and PG plant crops. The consistent ranking of cultivars across the three sites and the strong correlation suggests that cultivar FPOP performance was stable across sites in the plant crop. The consistent high rankings of cultivars such as N12 and NCo376 and the consistent low ranking of N36 suggests that these cultivars may be candidates for use as group representatives (high and low FPOP groups) in further studies.

In the ratoon crop, mean tiller production in PG was significantly higher than at AK (Table 4.5a). The FPOP varied from 73 958 to 127 083 stalks ha^{-1} at AK and 115 625 to 166 964 stalks ha^{-1} at PG. Similar to the plant crop, cultivar NCo376 ranked amongst the top in PG and AK, while cultivar N36 ranked at the bottom across the two sites (Table 4.5a).The FPOP mean for the ratoon crop at PG was higher than the plant crop while the ratoon crop in AK had a lower FPOP mean than the plant crop. Despite some similar rankings, the rank correlation across the sites was low (r=0.37) and non-significant (Table 4.5b). This shows that FPOP may be stable across sites in the plant crop but not in the ratoon.

Cultivar ranking was similar across crop cycles, within a site. For example cultivars N12, N36, N40, N41, N48, N51, N52, and NCo376 had similar rankings between the plant and ratoon crop at AK. Cultivars N12, N25, N35, N36, N48, and NCo376 ranked similarly in the plant and ratoon crop at PG. The correlation coefficient was moderate and significant between crop cycles for the same location at AK (r=0.60) and moderate and non-significant at PG (r=0.50) (Table 4.5b). In general, the FPOP values of the cultivars were similar across ratoons, meaning that cultivar trait values for FPOP in ratoons can be adequately represented from information gathered in the plant crop.

Table 4.5a) Means for final tiller population (FPOP) (stalks ha^{-1}) for 12 cultivars at Amatikulu (AK), Bruynshill (BH), and Pongola (PG) in the plant and first ratoon crop cycles

	Plant crop			First ratoon	
Cultivar	**AK**	**PG**	**BH**	**AK**	**PG**
N12	133333 $^{e\ (1)}$	162946 $^{d\ (1)}$	234375 $^{e\ (1)}$	127083 $^{eg\ (1)}$	147768 $^{ab\ (4)}$
N19	112500 $^{cd\ (3)}$	124107 $^{abc\ (5)}$	148125 $^{ab\ (9)}$	96875 $^{bc\ (7)}$	150000 $^{ab\ (2)}$
N25	103125 $^{bc\ (6)}$	110714 $^{a\ (10)}$	159375 $^{abcd\ (7)}$	108333 $^{cdef\ (3)}$	130804 $^{a\ (9)}$
N31	97396 $^{bc\ (9)}$	134375 $^{c\ (3)}$	188125 $^{d\ (2)}$	117708 $^{defg\ (2)}$	143750 $^{ab\ (8)}$
N35	105729 $^{c\ (5)}$	124107 $^{abc\ (5)}$	144375 $^{ab\ (11)}$	81250 $^{ab\ (10)}$	146875 $^{ab\ (5)}$
N36	67708 $^{a\ (12)}$	106250 $^{a\ (11)}$	126250 $^{a\ (12)}$	73958 $^{a\ (12)}$	130357 $^{a\ (10)}$
N40	81771 $^{ab\ (11)}$	120536 $^{abc\ (7)}$	147500 $^{ab\ (10)}$	89062 $^{abc\ (9)}$	125000 $^{a\ (11)}$
N41	106250 $^{c\ (4)}$	120536 $^{abc\ (7)}$	162500 $^{bcd\ (6)}$	107292 $^{cd\ (5)}$	149107 $^{ab\ (3)}$
N48	90625 $^{bc\ (10)}$	111607 $^{ab\ (9)}$	151875 $^{abc\ (8)}$	93750 $^{bc\ (8)}$	115625 $^{a\ (12)}$
N51	101042 $^{bc\ (7)}$	133036 $^{bc\ (4)}$	183750 $^{cd\ (3)}$	98437 $^{bcd\ (6)}$	144643 $^{ab\ (7)}$
N52	100521 $^{bc\ (8)}$	104911 $^{a\ (12)}$	172500 $^{bcd\ (5)}$	74479 $^{a\ (11)}$	146429 $^{ab\ (6)}$
NCo376	128125 $^{de\ (2)}$	162054 $^{d\ (2)}$	183750 $^{cd\ (3)}$	108333 $^{cde\ (3)}$	166964 $^{b\ (1)}$
Mean	**102344**	**126265**	**166875**	**98047**	**141443**
§	**a**	**b**	**c**		
§§	**a**	**b**		**a**	**c**

Means followed by the same letters are not significantly different within a site and crop cycle. Numbers in brackets indicate ranking of cultivars (1 representing the cultivar with the highest FPOP, while 12 represents the cultivar with the lowest FPOP)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 4.5b) Cultivar rank correlation coefficients for FPOP at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.50	1		
AK P	0.61*	0.88**	1	
AK R	0.73*	0.37	0.60*	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, *; Significant at P<0.01 and P<0.05, respectively.

4.3.1.4 Tiller survival percentage (TSP)

In the plant crop PG had the highest TSP, followed by BH and AK. The higher TSP could be linked to more favourable irrigated conditions at PG compared to harsher rainfed conditions at

the other two sites. The cultivar N12 had the highest TSP at AK (64%) and relatively high TSP at PG (79%) and BH (56%) (Table 4.6a). Slow tillering cultivars such as N12 are associated with a high TSP. Cultivars that had low values of TSP in the plant crop were N31 (29%) at AK and N52 at PG (58%) and BH (45%). The TSP varied from 64 to 29% at AK, 79 to 58% at PG and 56 to 45% at BH. In the plant crop, the cultivar N52 ranked low across the three sites showing that tiller mortality is high in this cultivar, irrespective of growing conditions. A significant ($P<0.05$) moderate rank correlation was found between AK plant crop and PG plant crop ($r=0.66$), indicating similar rankings of some cultivars. Cultivars N19, N25, N35, N36, N40, N41, and N48 had similar ranking across all sites.

The TSP at AK varied from 59% to 34% while the range at PG was 51% to 36%. Cultivar N35 had low TSP values at AK and PG, while cultivars N41 and N19 had the high TSP values at AK and PG, respectively. Plant crops generally have a higher TSP, whereas ratoon crops have higher tiller mortality due to higher tillering (Vasantha et al., 2014). As expected, the ratoon TSP was lower than the plant crop TSP at PG, while the ratoon crop produced more tillers than the plant crop at AK. The correlation coefficient among PG and AK ratoon crops was moderate ($r=0.66$) and significant ($P<0.05$) (Table 4.6b). Cultivar ranking was consistent from site to site for the ratoon and plant crop, suggesting that TSP is a stable trait.

It was generally found that cultivar rankings were similar across ratoons only at AK. For example, cultivars N12, N25, N31, N41, N48, and N52 were ranked similarly between the plant and ratoon crops (Table 4.6a). This is further supported by the moderate and significant ($r=0.61$) correlation value at AK; showing that similar information on cultivar ranking for TSP was obtained from the ratoons. In contrast, the correlation value at PG was low and non-significant ($r=0.34$) across ratoons; indicating that the TSP cannot be adequately represented with values from only the plant crop. The reasons for the poor correlation between ratoons at PG compared with the good correlation at AK are unclear.

Table 4.6a) Means for tiller survival percentage (TSP) (%) for 12 cultivars at Amatikulu (AK), Pongola (PG), and Bruynshill (BH) in the plant and first ratoon crop cycles

	Plant crop			First ratoon	
Cultivar	**AK**	**PG**	**BH**	**AK**	**PG**
N12	64 e (1)	79 c (1)	56 b (1)	57 d (2)	47 ab (2)
N19	45 cd (6)	71 abc (5)	49 ab (7)	54 d (3)	51 b (1)
N25	47 cd (5)	68 abc (7)	49 ab (7)	53 d (4)	46 ab (4)
N31	29 a (12)	68 abc (7)	53 ab (4)	41 abc (9)	46 ab (4)
N35	44 cd (8)	63 ab (10)	46 a (10)	34 a (12)	36 a (12)
N36	38 bc (10)	58 a (11)	46 a (10)	50 cd (6)	46 ab (4)
N40	45 cd (6)	70 abc (6)	51 ab (5)	39 ab (10)	41 ab (10)
N41	53 d (2)	79 c (1)	51 ab (5)	59 d (1)	43 ab (8)
N48	51 d (4)	72 bc (4)	49 ab (7)	51 cd (5)	47 ab (2)
N51	40 bc (9)	74 bc (3)	55 ab (3)	50 cd (6)	45 ab (7)
N52	34 ab (11)	58 a (11)	45 a (12)	36 a (11)	42 ab (9)
NCo376	53 d (2)	68 abc (7)	56 b (1)	48 bcd (8)	39 ab (11)
Mean	**45**	**69**	**50**	**48**	**44**
§	**a**	**c**	**b**		
§§	**ab**	**c**		**b**	**a**

Means followed by the same letters do not differ significantly within a site and crop cycle. Numbers in brackets indicate ranking of cultivars (1 representing the cultivar with the highest TSP, while 12 represents the cultivar with the lowest TSP)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 4.6b) Cultivar rank correlation coefficients for TSP at Amatikulu (AK) and Pongola (PG) across ratoons and sites

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.34	1		
AK P	0.66*	0.07	1	
AK R	0.74*	0.66*	0.61*	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**P<0.01, * P<0.05

4.3.2 Stalk elongation rate (SER)

Tiller (stalk) heights were plotted against accumulated thermal time (Figures 4.4, 4.5, 4.6, and 4.7) and SER was calculated as the *x* coefficient (or slope) of the straight line. Stalk height was

generally lower at AK than at BH and PG (Figures 4.4 and 4.5) in the plant crop. Examples of the stalk elongation trends across the three sites in the plant crops are shown for cultivars N31 and NCo376 in Figures 4.4 and 4.5, respectively. In the ratoon crop, cultivars generally had a higher SER at PG than at AK (Figures 4.6 and 4.7). Final stalk height was generally lower in the ratoon crop than in the plant crop.

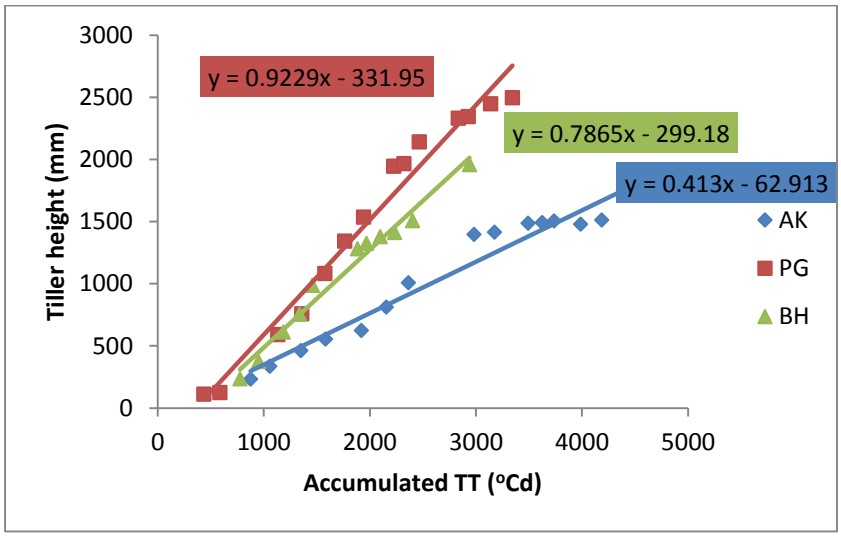

Figure 4.4 Stalk height of cultivar N31 in the plant crop at BH, PG, and AK as a function of accumulated thermal time using a base temperature of 10°C

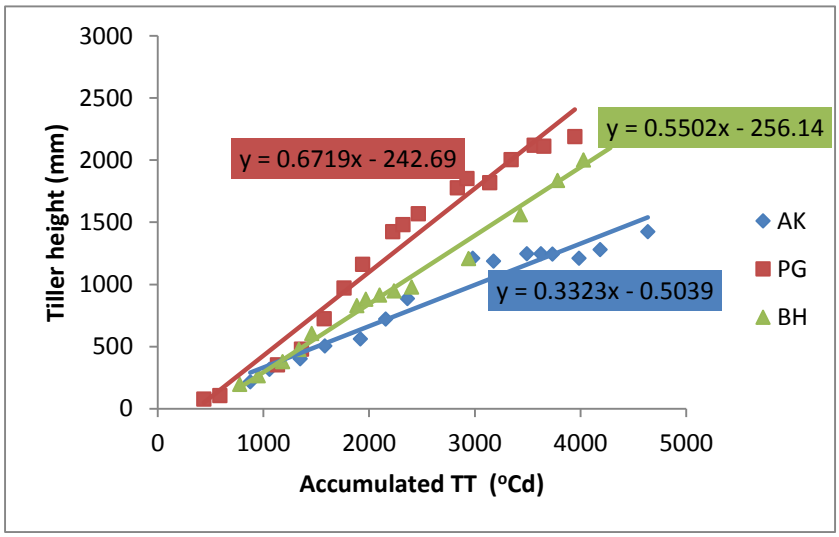

Figure 4.5 Stalk height of cultivar NCo376 in the plant crop at BH, PG, and AK as a function of accumulated thermal time using a base temperature of 10°C

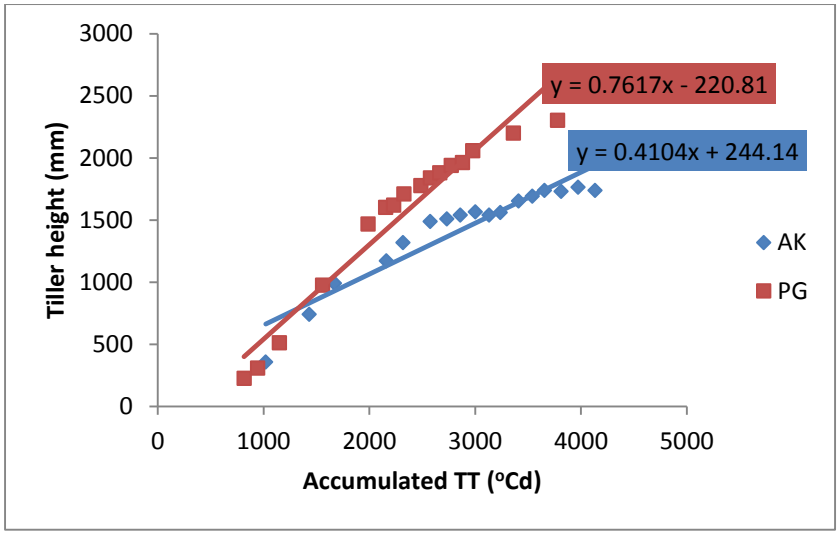

Figure 4.6 Stalk height of cultivar N31 in the first ratoon crop at PG and AK as a function of accumulated thermal time using a base temperature of 10°C

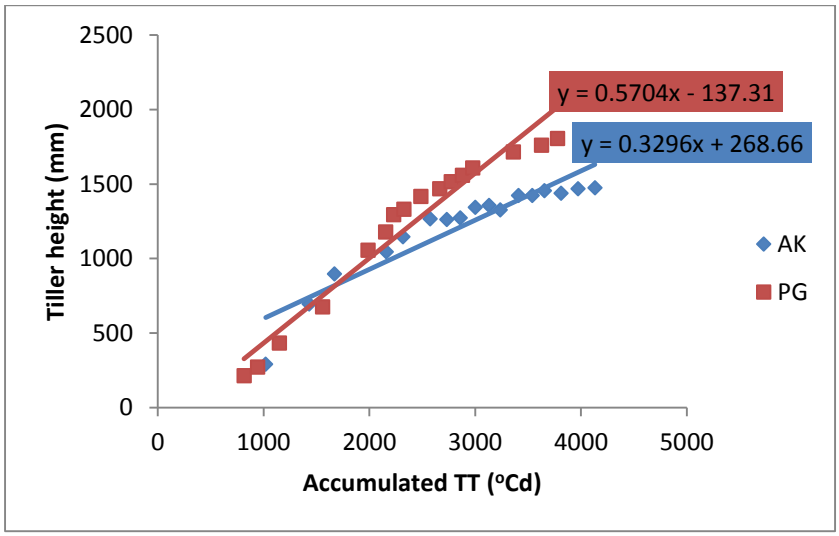

Figure 4.7 Stalk height of cultivar NCo376 for ratoon crop at PG and AK as a function of accumulated thermal time using a base temperature of 10°C

Table 4.7 shows the mean square values from the combined ANOVA for SER of the PG and AK trials for both the plant and first ratoon crops. The mean squares for the main effects of cultivar, crop cycle, and site were highly significant ($P<0.01$) for SER (Table 4.7). The C x S and R x S interaction mean squares were highly significant ($P<0.01$) (Table 4.7). The significant C x S interaction mean squares meant that SER may have to be determined for each cultivar at each site. The C x S x R interaction and the two way interaction of C x R were not

significant (Table 4.7). The significant interaction terms C x S and R x S imply changes in cultivar ranking for mean SER across sites and changes in overall SER across crop cycles, respectively. The mean square values for S were much higher than those of the other sources; indicating S accounted for more of the variation than other sources of varition.

Table 4.7 Mean square values from analysis of variance, combined across the plant and first ratoon crop cycles and sites (Pongola and Amatikulu) for stalk elongation rate (SER)

Source	d.f.	Mean square value
Cultivar (C)	11	0.066**
Crop cycle (R)	1	0.468**
Site (S)	1	5.119**
C x R	11	0.004
C x S	11	0.018**
R x S	1	0.108**
C x S x R	11	0.003

**, *; Significant at P<0.01 and P<0.05, respectively.

Table 4.8 shows the mean square values from ANOVA of the PG, AK, and BH plant crops only. The mean square values for main effects of C and S and the C x S interaction were highly significant for SER (Table 4.8). The significant interaction indicates that relative values of SER among cultivars differed by location. The order of importance of sources of variation was S > C > C x S.

Table 4.8 Mean square values from analysis of variance in the plant crop, combined across sites (Bruynshill, Amatikulu and Pongola) for stalk elongation rate (SER)

Source	d.f.	Mean square values
Cultivar (C)	11	0.067**
Site (S)	2	1.685**
C x S	22	0.016**

**, *; Significant at P<0.01 and P<0.05, respectively.

In the plant crop, the mean SER at AK (0.42) was lower than at BH (0.58), and PG (0.79) (Table 4.9a). Stalk elongation is highly sensitive to water stress; the low temperatures at BH and irrigation at PG may have reduced minor stress on the plants. Silva et al. (2008) reported that stalk height was the most affected yield component under non-irrigated conditions. In this study, the lower SER at AK was most likely due to moisture stress brought about by rainfed

conditions. The cultivars N31 and N52 had the highest SER at BH (0.79 and 0.78 respectively) and PG (0.92 and 0.89 respectively). Cultivar N51 was also the highest at PG. These SER values were significantly higher than values for N12 at both sites. Nine cultivars had high SER at AK. The consistent ranking of cultivars N12, N19, N25, N36, N40, N41, N48, N51, N52, and NCo376 and the highly significant and moderately strong (r=0.65) correlation coefficient across AK and PG suggest that SER is a stable trait in the plant crop. This medium correlation value across locations, indicates some cultivar interaction hence the significant C x S. It also suggests that such cultivars may be ideal candidates for use as group representatives in further studies.

In the ratoon crop, SER at PG (0.64) was higher than at AK (0.36). SER values ranged from 0.30 to 0.47 at AK and 0.49 to 0.76 at PG. Similar to the plant crop, cultivars with the highest SER had significantly different values from cultivar N12, which had values of 0.49 and 0.32 for PG and AK respectively. Differences in mean SER between the ratoon and plant crop were significant. These results are similar to a report by Singh and Singh (2000), where cultivars differed under different climatic conditions for SER. A highly significant and strong correlation value (r=0.89) was observed between AK and PG for the ratoon crop (Table 4.9b), showing that SER is stable across sites in the ratoon crop as well.

Within a site (AK), cultivars N12, N25, N40, N48, N51, N52, and NCo376 had similar rankings across crops. All cultivars at PG had similar ranking across crop cycles with the exception of cultivar N35. There was a highly significant ($P<0.01$) and strong correlation (r=0.94) between crop cycles at PG, while a moderate (r=0.66) and significant ($P<0.05$) correlation was observed at AK (Table 4.9b). This meant that the relationship between cultivar rankings was fairly strong thus single trait values can be determined from one site. Also, the moderate value indicates moderate to high interaction levels. Therefore, the genotypes across environments were not classified exactly equally, i.e. significant C x S or C x R interactions. Since SER is not sensitive to crop cycles, trait values determined in the plant crop may be appropriate for use in ratoon crops for the purposes of crop modelling. Additionally, stable cultivar rankings mean that some cultivars can be confidently chosen as group representatives (low, moderate, higher SER cultivars) in future physiological/phenology experiments.

Table 4.9a) Stalk elongation rate (SER) (mm $^{o}Cd^{-1}$) means in the plant and ratoon crops at Bruynshill (BH), Amatikulu (AK) and Pongola (PG)

	Plant crop			First ratoon	
Cultivar	**BH**	**AK**	**PG**	**AK**	**PG**
N12	0.50 a (9)	0.39 ab (10)	0.55 a (12)	0.32 ab (10)	0.49 a (12)
N19	0.56 b (6)	0.43 bc (4)	0.83 def (6)	0.34 abc (7)	0.62 bc (8)
N25	0.50 a (9)	0.39 ab (10)	0.77 c (9)	0.31 ab (11)	0.59 b (9)
N31	0.79 e (1)	0.41 abc (7)	0.92 g (1)	0.41 de (3)	0.76 e (1)
N35	0.57 b (5)	0.46 bc (2)	0.79 cd (8)	0.37 bcd (5)	0.66 bcde (5)
N36	0.62 c (4)	0.44 bc (3)	0.84 def (5)	0.36 abcd (6)	0.66 bcde (5)
N40	0.51 a (7)	0.40 abc (8)	0.68 b (10)	0.30 a (12)	0.56 ab (11)
N41	0.68 d (3)	0.42 bc (6)	0.85 def (4)	0.42 de (2)	0.71 cde (4)
N48	0.50 a (9)	0.40 abc (8)	0.82 cde (7)	0.33 abc (8)	0.63 bcd (7)
N51	0.51 a (7)	0.43 bc (4)	0.87 efg (3)	0.40 cd (4)	0.74 e (2)
N52	0.78 e (2)	0.49 c (1)	0.89 fg (2)	0.47 e (1)	0.73 de (3)
NCo376	0.49 a (12)	0.33 a (12)	0.67 b (11)	0.33 ab (8)	0.57 ab (10)
Mean	**0.58**	**0.42**	**0.79**	**0.36**	**0.64**
§	**b**	**a**	**c**		
§§		**b**	**d**	**a**	**c**

Means followed by the same letters do not differ significantly within a site and crop cycle. Numbers in brackets indicate ranking order of cultivars (1 representing the cultivar with the highest SER, while 12 represents the cultivar with lowest SER)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 4.9b) Cultivar rank correlation coefficients for SER at Amatikulu (AK) and Pongola (PG) across ratoons and sites

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.94**	1		
AK P	0.65*	0.66*	1	
AK R	0.85**	0.89**	0.66*	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**P<0.01, P<0.05

4.3.3 Heritability

Variance components and broad sense heritability for the five traits are shown in Table 4.10. The environmental variance (E) was higher than the genetic variance (C) for all traits studied. Similarly, Milligan et al. (1990) reported higher environmental variance than genetic variance for sucrose yield, cane yield, sucrose, stalk number, stalk length, stalk diameter, stalk weight, Brix, juice purity, and stalk density. In contrast genotypic variance was found to be higher than the environmental variance for cane yield, millable cane, single cane weight, stalk diameter, and stalk length (Chaudhary, 2001). The genetic (cultivar) variance was higher than that of the interaction variances for all traits.

Broad sense heritability estimates (H^2) ranged from 0.49 to 0.97 for the traits across the two crops. H^2 was highest for FPOP (0.97) followed by SER (0.72), PTP (0.70), TSP (0.60), and TTPP (0.49), respectively. The high estimates of H^2 for FPOP, SER, and PTP were due to their high genetic variance values. This suggests that genetic variance had a high contribution to total variation for these traits. These results are similar to those reported by Jamoza et al. (2014) in Kenya who found high broad sense heritability for stalk number, weight, length, number of internodes, and stalk diameter, ranging from 82.6 to 92.4%. High heritability estimates for PTP, SER and FPOP suggest that simple selection for these traits would be effective.

A high broad sense heritability indicates high genetic potentials for these traits and a low effect of the environment. Moderate heritability estimates detected for TTTP (0.49) and TSP (0.60) indicate that heritabilities for these traits are lower than the other traits. These results suggest that PTP, FPOP, and SER are traits that can be selected for in a breeding programme. The stability of these traits also makes them good candidates for use in model-assisted breeding.

Table 4.10 Variance components and broad sense heritability for peak tiller population (PTP), thermal time to peak tiller population (TTPP), final population (FPOP), tiller survival percentage (TSP), and stalk elongation rate (SER)

Source	PTP (x 10^8)	TTPP	FPOP (x10^7)	TSP (x 10^{-2})	SER (x 10^{-2})
C	7.23	15877	20.47	0.21	0.30
C x S	2.69	15141	0	0.08	0.19
C x R	0	0	0	0.13	0.01
C x S x R	4.82	14979	7.33	0.02	0
E	16.43	101946	27.83	0.51	0.39
H^2	**0.70**	**0.49**	**0.97**	**0.60**	**0.72**

4.4 Conclusions

For the trait PTP, rank correlations between sites in the ratoon crop were stronger than in the plant crop, suggesting that this trait is more stable in the ratoons. At PG the rank correlation between the plant and ratoon crop was strong, but weak at AK. Some cultivars showed consistent rankings across sites and across ratoons within sites, suggesting that they can be used as indicator cultivars for this trait in future studies involving cultivar characterisation. The trait TTPP generally showed poor rank correlations between sites and ratoons within sites, suggesting that it is more influenced by environmental factors. Its use as a trait for model-assisted breeding will, therefore, be limited. Of note was the consistent ranking of cultivar N12 as a high TTPP (slow growth rate) cultivar across sites and ratoons.

The trait FPOP showed high rank correlations between sites in the plant crop, but poor correlations between sites in the ratoon. Within a site, however, the correlations between the plant and ratoon crops were strong. This suggests that information on FPOP measured in the plant crop can adequately represent relative values obtained in the ratoon and that cultivars may be characterised for this trait by single-site and single-ratoon experiments in future. Cultivars such as N12 and NCo376 (high FPOP), and N36 and N48 (low FPOP) were identified as ideal indicator cultivars in future studies. The trait TSP also showed consistent rankings across sites in the plant and ratoon crop showing that it was relatively stable. At PG, however, the correlations between the plant and ratoon crops were weak.

The trait SER was the most stable trait of all, showing strong rank correlations between sites and across ratoons within sites. Information on SER in ratoon crops can be obtained from data gathered in the plant crop only, and cultivars may therefore be characterised using single-site,

single-ratoon studies in future. Cultivars NCo376 and N12 were identified as ideal indicator cultivars for low SER, while cultivars N31 and N52 were identified as high SER cultivars.

The tiller population dynamics were more affected by E, indicating the major role of environmental factors in influencing these traits. SER however, was more affected by G as evidenced by the larger value of genetic variance. The traits SER, PTP and FPOP had a high heritability (ranging from 0.70 to 0.97) as they were less influenced by the interaction components, these traits could thus be used as additional selection tools for sugarcane yield improvement. The moderate heritability for TTPP and TSP, meant that cultivar rankings for these traits were less consistent between ratoons and from site to site. Follow-up studies should involve the statistical grouping of cultivars into categories for each trait, and the determination of mean values for each category. These mean values should then be incorporated and validated using the Canegro crop model.

4.5 References

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CHAPTER 5

Quantifying sugarcane cultivar differences and genotype x environment interactions for leaf growth and development traits

Abstract

Some of the cultivar coefficients used to express overall leaf growth and development in the Canegro model are LAR, LAI, LFmax, LAmax and TTLAmax. Estimates of these coefficients for a wider range of cultivars are not available. The primary objective of this study was to quantify the cultivar coefficient values for some leaf growth traits for a diverse range of sugarcane cultivars. An additional objective was to determine the stability and heritability of these traits across environments and crop stages to determine their potential contribution to future model-assisted breeding.

Three cultivar trials were established at three separate sites on SASRI research farms; AK, PG and BH. The same set of 12 cultivars was tested at the three sites. The trials were planted in randomised complete block designs with four replications and harvested for two crops. The cultivar traits LAR, LAmax, LFmax, TTLAmax and LAI were determined from measurements of leaf length, leaf width and leaf height along the stalk at two-week intervals together with relevant temperature data from automatic weather stations. Cultivar rankings across sites and across crops within sites were evaluated for stability. The data was analysed using GENSTAT to estimate the variance components and broad-sense heritability for each trait.

For the traits LAmax and LAR, strong rank correlations were observed across sites and ratoons within sites, suggesting that these traits were stable. Cultivars N36 and N52 showed consistently high LAmax values compared with N41, which showed consistently low values. For LAR and LFmax, cultivar N35 consistently showed the highest values in all environments compared with N12, suggesting that these are ideal indicator cultivars for these traits. The LAI was more stable across crops within a site compared with across sites for each crop.

All traits produced high broad sense heritability estimates, ranging from 0.84 for LFmax to 0.99 for LAmax with the exception of TTLAmax. Therefore, LAI, LAR, LFmax and LAmax are highly heritable and can be selected for in a breeding programme. The relatively stable rankings of cultivars across sites and crops suggest that some traits can be quantified per

cultivar by single-site and single-crop experiments in future. The cultivar coefficient values determined here will help refine the crop model's ability to simulate cultivar growth differences across environments. The range of values for these traits will also contribute to model-wise exploration of G x E interactions and future model-assisted breeding efforts for sugarcane.

5.1 Introduction

Leaves are 'photosynthetic engines' of the plant, producing sugar that is stored in the stalks. One of the most important characteristics of leaves with regard to their function is their total area. The leaves become both longer and wider as the plant develops, until a stable leaf size is established. Leaf area depends on the number of leaves and average surface area per leaf (van Dillewijn, 1952) and gives an idea of the plants' photosynthetic capacity (Patil et al., 2009). The area of the individual leaf blades is smallest at the base of the plant and gradually increases toward the top until a maximum is reached. As new leaves appear and individual leaves expand, total leaf area increases.

LAI is the total one-sided area of leaf tissue per unit ground surface area (Watson, 1947). The LAI is very important in the simulation of sugarcane (*Saccharum* spp.) yield in crop models as LAI influences the PAR intercepted by the canopy. The amount of solar radiation intercepted is used during photosynthesis and is directly linked to yield (Sandhu et al., 2012). The fraction of PAR that is intercepted by the crop is a function of LAI; meaning that an increase in leaf area can improve interception of solar radiation (Shoko et al., 2009). The Canegro model uses LAI to simulate FiPAR; which is calculated as a function of the total number of leaves per tiller.

LAI differs greatly between cultivars. Generally, cultivars having high population density possess higher LAI (Iqbal et al., 2011). Rafiq et al. (2007) showed that early maturing cultivars like CPF-237 attain maximum LAI earlier than late or medium late maturing cultivars like SPF-213. In contrast, Robertson et al. (1996) investigated early growth between plant crops of two cultivars and found no difference between leaf area per stalk. Zhou (2003) plotted LAI against days after planting and accumulated thermal time. There was a marked difference among the cultivars in LAI towards the peak LAI. Cultivar N14 had the highest LAI while ZN7 had the lowest, with values that ranged from 0.082-1.833 and 0.062-1.229 respectively.

Leaf development and growth traits that are important in determining LAI include leaf appearance, elongation of the newly-emerged leaves, increase in leaf number, senescence of old leaves and an increase in leaf area (Nehbandani et al., 2013). Leaf appearance is the number of leaves that become visible on a stem per unit time (Streck et al., 2003). Leaves will emerge based on a phyllochron interval. Phyllochron is the time interval between the appearance of successive leaves. This interval can be expressed on a calendar basis or in thermal time (TT), measured in units of degree-days (°C day).

Leaf size and area is driven by leaf elongation rate (LER, cm d^{-1}) which is dependent on air-temperature as well as water availability. Leaf area expansion is calculated from increases in length and leaf width. Leaves stop expanding once a maximum allowable blade area is reached. The maximum allowable blade area increases with successive leaves (Inman-Bamber and Kiker, 1997) until a specific number of leaves LF_{MAX} has formed. The maximum allowable blade area remains at a constant value LA_{MAX}.

It has been shown that these leaf development traits are influenced by genotype. Sinclair et al. (2004) studied leaf appearance of four cultivars and showed values for leaf appearance rate ranging from 0.0085 to 0.0115 leaf TU^{-1}, or a phyllochron interval of 118-87 TU. Values of leaf appearance rates reported by Inman-Bamber (1994) for cultivars NCo376 and N12 were 0.0092 and 0.0085 leaf TU^{-1}, respectively (109 and 118 TU phyllochron interval, respectively). Marin (2011) reported maximum leaf size to range between 733-796 cm^2 for two Brazilian cultivars. Both cultivars reached maximum leaf size at leaf number 25, which is similar to results of Sinclair et al. (2004) for cultivar CP72-2086 in Florida. Zhou (2003) evaluated four cultivars in one environment. The cultivars had different leaf sizes. Values ranged from 355.3 cm^2 (NCo376) to 457.8 cm^2 (N14). In sugarcane, it has been shown that leaf size and leaf elongation rate show consistent genotypic variation (Bonnet, 1998; Robertson et al., 1998). However, little genetic and phenotypic information on these traits are available for a wide range of genotypes in different locations.

The primary objective of this study was to quantify the cultivar coefficient values for some leaf growth traits for a diverse range of sugarcane cultivars. An additional objective was to determine the stability and heritability of these traits across environments and crop stages to determine their potential contribution to future model-assisted breeding.

5.2 Material and methods

The details of all trials, including site characteristics, cultivars, experimental designs, plot dimensions and crop characteristics were described in detail in Chapter 3. For brevity, only the details of the data collection and analysis relevant to this chapter are briefly mentioned below.

Leaves of the tagged plants were numbered chronologically from the bottom up. Leaf development measurements (leaf length, and width) were done every 2 to 3 weeks on the numbered leaves. The LAR was determined from fitting a straight line-equation when thermal time was plotted against leaf number. The LAR was the coefficient of x. The LFmax is the maximum leaf number obtained from the graph of leaf number against thermal time. The LAmax was determined as the maximum LA produced when LA was plotted against thermal time. The TTLAmax is the thermal time required to reach LAmax.

The data for the parameters was analysed using Genstat v14 (VSN International Ltd, 2011). A combined ANOVA was performed for AK and PG where main and interaction effects were tested. The main effects were cultivar, site and ratoon. The trial at BH was harvested on a 24-month cutting cycle, and as a result, was only harvested for the plant crop. Therefore, a separate ANOVA was also conducted for BH, AK and PG to test the main effects of site and cultivar for the plant crop only. Treatment mean comparisons were made using Duncan's LSD test at P=0.05.

The variance components were calculated using the mean squares from the ANOVA. Estimates of broad sense heritability for the different traits were calculated using the variance components method. Broad sense heritability was determined from the equation: $h^2 = \sigma^2 g / \sigma^2 p$, as described in Chapter 3.

5.3 Results and discussion

5.3.1 Leaf area (LA)

LA was determined at each measurement date and plotted against thermal time. LA usually increases slowly, reaching a peak and then drops as leaves start to senesce. Examples of this are shown for the plant crop for cultivars N12 (Figure 5.1), N25 (Figure 5.2) and NCo376 (Figure 5.3). LAmax was consistently highest at BH, followed by PG and AK respectively. In BH leaf area increased, then remained constant before continuing to increase, reaching a peak and then dropped as leaves started to senesce. The first phase of rapid increase could be associated with the increase in leaf area of successive leaves on the stalks, rapid leaf emergence and tillering during summer. The leaf area then remained stabilised due to the cold winter conditions experienced in the first winter (the BH crops grew through a 24-month cycle). Further increases in leaf area may be caused by the return of the warmer conditions (second summer) and leaf expansion.

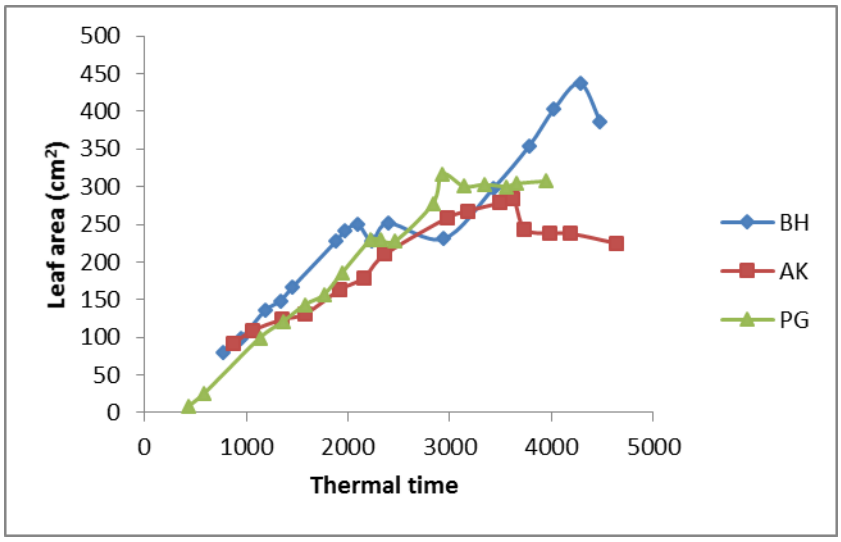

Figure 5.1 Leaf area for cultivar N12 at PG, AK and BH as a function of accumulated thermal time (ºCd)

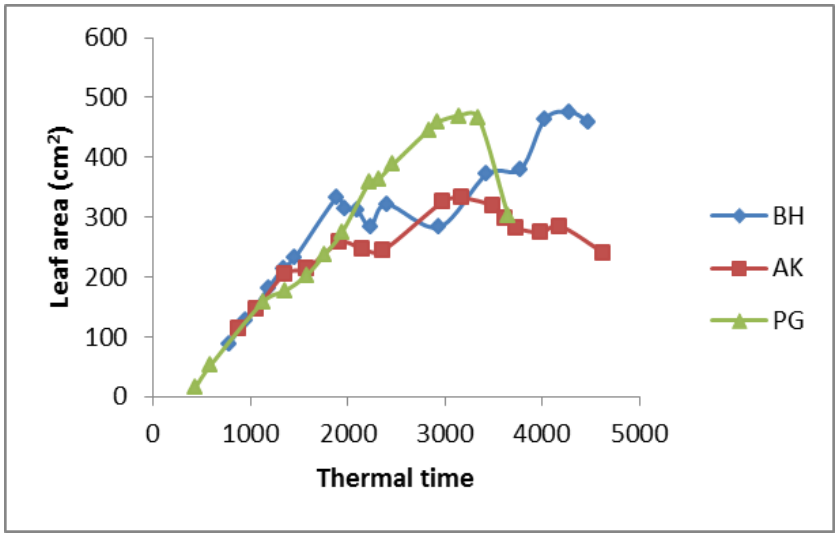

Figure 5.2 Leaf area for cultivar N36 at PG, AK and BH as a function of accumulated thermal time (ºCd)

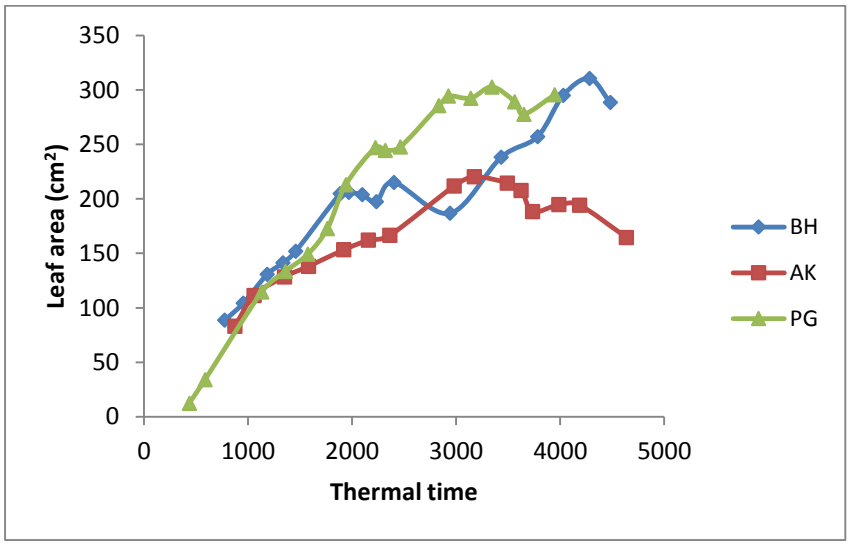

Figure 5.3 Leaf area for cultivar NCo376 at PG, AK and BH as a function of accumulated thermal time (ºCd)

Table 5.1 shows the mean square values from the combined ANOVA of the PG and AK trials for both the plant and first ratoon crops for LAmax, and TTLAmax. The main effects of cultivar (C), site (S) and crop cycle (R) were generally highly significant ($P<0.01$) to significant ($P<0.05$) for LAmax. For TTLAmax, only the C main effect was significant (Table 5.1).The cultivar x site (C x S) interaction was significant for TTLAmax ($P<0.05$) meaning that the relative cultivar rankings across sites varied for this trait. The crop cycle x site (R x S) interaction was significant for LAmax. The non-significant three-way C x S x R interaction for

both LAmax and TTLAmax showed that the relative cultivar values for these traits did not vary from one site to the next, and from the plant to the ratoon crop. The mean square value for S was higher than the mean square values for both other traits. The main effect of S therefore contributed most to total variance for the two traits.

Table 5.1 Mean square values from analysis of variance, combined across ratoons (plant and first ratoon) and sites (Pongola and Amatikulu) for maximum leaf area (LAmax) and thermal time to reach maximum leaf area (TTLAmax)

Source	d.f.	Mean square values	
		TTLAmax	LAmax
Cultivar (C)	11	286459*	32056**
Site (S)	1	517739	334365**
Crop cycle (R)	1	20989	14709*
C x S	11	273084*	1857
C x R	11	117569	2400
R x S	1	194040	29356*
C x S x R	11	98773	3896

**, *; Significant at P<0.01 and P<0.05, respectively.

Table 5.2 shows the mean square values from ANOVA of the PG, AK and BH plant crop only. The main effects of C and S were highly significant for all traits, with the exception of the significant effect of C on TTLAmax (Table 5.2). The C x S interaction was highly significant for LAmax (P<0.01) and significant for TTLAmax (P<0.05). Therefore the cultivar values for the traits differed by site for the plant crop. The main effect of S was higher than the C x S interaction and C mean square value for both traits.

Table 5.2 Mean square values from analysis of variance for plant crop, combined across sites (Bruynshill, Pongola and Amatikulu) for maximum leaf area (LAmax) and thermal time to reach maximum leaf area (TTLAmax)

Source of variation	d.f.	Mean square values	
		TTLAmax	LAmax
Cultivar (C)	11	581045*	27142**
Site (S)	2	4059574**	152754**
C x S	22	528907*	6715**

**, *; Significant at P<0.01 and P<0.05, respectively.

5.3.1.1 Maximum leaf area (LAmax)

In the plant crop the mean LAmax was higher at PG followed by BH and AK respectively (Table 5.3a). Drought stress experienced at AK is known to limit plant growth through increased leaf rolling. This could be one of the reasons for the lower LAmax experienced at AK. The cultivar N52 had the highest LAmax at PG and AK; while cultivar N48 had the highest value at BH (Table 5.3a).The cultivar N36 had the second highest LAmax at PG, AK and BH showing that this cultivar is able to express this trait consistently across environments. Average LAmax ranged from 306.30 to 524.50 cm^2, 228.80 to 363.90 cm^2 and 265.40 to 444.30 cm^2 at PG, AK and BH respectively. Cultivar N41 had the lowest LAmax at AK and BH and second lowest at PG showing that it too expresses this trait consistently across environments. The correlation of ranks of cultivars was high and significant ($P<0.05$) among the two sites in the plant crop ($r=0.81$) (Table 5.3b). Cultivars N12, N48 and N52 had similar rankings at PG and AK; these consistent rankings suggest that this trait may be stable across sites in the plant crop.

Similar to the plant crop, PG had a higher LAmax than AK for the ratoon crop. The LAmax was higher for AK ratoon crop than the AK plant crop, while LAmax was lower for PG ratoon crop than the plant crop at PG. Cultivar N52 and N36 had the highest LAmax at PG and AK, respectively. This was in keeping with these cultivar's high rankings in the plant crop. The LAmax varied from 237 to 406.8 cm^2 at AK and 277.7 to 425.9 cm^2 at PG. In the ratoon crop all cultivars except N19, N25 and N48 had similar rankings across sites. The correlation values between the ratoon crop at AK and PG were high ($r=0.78$) and significant ($P<0.05$), suggesting that this trait is also consistent across sites in the ratoon crop (Table 5.3b).

Within a site cultivar rankings were similar across ratoons. Cultivars N12, N19, N25, N35, N36, N41, N48, N51, N52 and NCo376 ranked similarly across crops at AK while cultivars N12, N19, N31, N36, N41, N48, N51, N52 and NCo376 had similar rankings across crops at PG. The similar rankings of cultivars across ratoons means that the LAmax can be adequately represented with values from only the plant crop and is not significantly influenced by the environment. The correlation coefficient for LAmax was significant and high within sites across ratoons; with values of 0.79 and 0.64 for PG and AK respectively (Table 5.3b). This means that cultivar characterisation for LAmax in ratoons may be possible with information from the plant crop only.

Table 5.3 a) Maximum leaf area (LAmax) (cm^2) of 12 cultivars at Amatikulu (AK), Pongola (PG) and Bruynshill (BH) for the plant crop and first ratoon

	Plant crop			First ratoon	
Cultivar	**PG**	**AK**	**BH**	**AK**	**PG**
N12	338.50$^{a\ (10)}$	287.10$^{cd\ (8)}$	439.20$^{d\ (3)}$	277.00$^{ab\ (9)}$	326.00$^{ab\ (10)}$
N19	385.70$^{b\ (7)}$	281.40$^{bc\ (9)}$)	386.60$^{bcd\ (7)}$	262.80$^{ab\ (10)}$	374.60$^{ab\ (5)}$
N25	415.30$^{bc\ (6)}$	299.60$^{cd\ (7)}$	409.20$^{cd\ (6)}$	297.70$^{ab\ (6)}$	343.50$^{ab\ (9)}$
N31	427.40$^{c\ (5)}$	244.80$^{ab\ (10)}$	268.30$^{a\ (11)}$	370.30$^{bc\ (2)}$	376.20$^{ab\ (4)}$
N35	382.30$^{b\ (9)}$	299.80$^{cd\ (6)}$	338.00$^{bc\ (9)}$	304.70$^{abc\ (4)}$	366.50$^{ab\ (6)}$
N36	477.10$^{d\ (2)}$	330.30$^{de\ (2)}$	443.70$^{d\ (2)}$	337.70$^{abc\ (3)}$	425.90$^{b\ (1)}$
N40	435.40$^{c\ (3)}$	319.30$^{cd\ (3)}$	421.80$^{d\ (4)}$	280.80$^{ab\ (8)}$	350.30$^{ab\ (8)}$
N41	311.40$^{a\ (11)}$	228.80$^{a\ (12)}$	265.40$^{a\ (12)}$	246.20$^{a\ (11)}$	277.70$^{a\ (12)}$
N48	433.60$^{c\ (4)}$	318.00$^{cd\ (4)}$	444.30$^{d\ (1)}$	298.60$^{ab\ (5)}$	406.00$^{b\ (2)}$
N51	385.40$^{b\ (8)}$	314.70$^{cd\ (5)}$	420.80$^{d\ (5)}$	291.40$^{ab\ (7)}$	357.00$^{ab\ (7)}$
N52	524.50$^{e\ (1)}$	363.90$^{e\ (1)}$	346.30$^{bc\ (8)}$	406.80$^{c\ (1)}$	402.20$^{b\ (3)}$
NCo376	306.30$^{a\ (12)}$	236.70$^{a\ (11)}$	322.50$^{ab\ (10)}$	2370$^{a\ (12)}$	310.00$^{ab\ (11)}$
Mean	**401.90**	**293.70**	**375.50**	**300.90**	**359.70**
§	**c**	**a**	**b**		
§§	**c**	**a**		**a**	**b**

Means followed by the same letters do not differ significantly within site and crop cycle. Numbers in brackets indicate ranking order of cultivars (1 representing the cultivar with the highest LAmax while 12 represents the cultivar with the lowest LAmax)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 5.3b) Cultivar rank correlation coefficients for LAmax at Amatikulu (AK) and Pongola (PG) across ratoons and sites

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.79*	1		
AK P	0.81*	0.66*	1	
AK R	0.75*	0.78*	0.64*	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, *; Significant at P<0.01 and P<0.05, respectively.

5.3.1.2 Thermal time to reach maximum leaf area (TTLAmax)

The TTLAmax for the plant crop was higher at BH, followed by AK and PG (Table 5.4a). Plants took longer to reach LAmax at BH due to cooler temperatures, which may have caused a delay in leaf area development. The warmer and irrigated conditions at PG accelerated the rate at which leaf area accumulated. Cultivars NCo376, N51 and N35 had the highest TTLAmax at PG, AK and BH respectively, although they took longer to reach their LAmax than other cultivars. The TTLAmax varied from 3084 to 3444 at PG, 3127 to 3715 at AK and 2592 to 4381 at BH. A weak and non-significant (r=0.32) correlation between the plant crop at AK and PG meant that cultivar rankings differed among the sites (Table 5.4b). This suggests that TTLAmax may not be stable across sites in the plant crop.

In the ratoon crop the mean TTLAmax at PG was lower than the plant crop mean; while it was higher than the plant crop mean at AK. Similarly to the plant crop, higher TTLAmax mean was obtained at AK than at PG. In the ratoon crop the TTLAmax ranged from 2960 to 3752 and 2761 to 3703 at AK and PG respectively. Cultivar N51 had the highest TTLAmax at PG and AK, while cultivars N25 and N35 had the lowest TTLAmax at AK and PG respectively. The correlation coefficient was weak and non-significant (r=0.15) between AK and PG, showing that this trait was not stable across sites in the ratoon crop and therefore cultivar characterisation from a single site experiment is not possible.

Ranking for most cultivars was similar across ratoons within AK, for example cultivars N12, N25, N31, N35, N36, N40, N41, N48, N51, N52 and NCo376 had similar rankings between the plant and ratoon crop. The high and significant correlation coefficient (r=0.85) across ratoons at AK indicated a strong relationship between cultivar rankings. In contrast, a weak correlation existed (r=0.04) between ratoons at PG, indicating that the TTLAmax cannot adequately be represented by values from the plant crop only. Reasons for the strong correlation between ratoons at AK compared with the weak correlation between ratoons at PG are unclear.

Table 5.4a) Thermal time to reach maximum leaf area (TTLAmax) (ºCd) of 12 cultivars at Amatikulu (AK), Pongola (PG) and Bruynshill (BH) for the plant crop and first ratoon

	Plant crop			First ratoon	
Cultivar	**AK**	**PG**	**BH**	**AK**	**PG**
N12	3558$^{bcd\ (3)}$	3370$^{a\ (3)}$	4281$^{b\ (3)}$	3637$^{a\ (2)}$	3217$^{ab\ (6)}$)
N19	3175$^{abc\ (7)}$	3343$^{a\ (5)}$	4218$^{b\ (5)}$	3035$^{a\ (11)}$	3006$^{ab\ (11)}$
N25	3141$^{ab\ (11)}$	3239$^{a\ (8)}$	4268$^{b\ (4)}$	2960$^{a\ (12)}$	3403$^{ab\ (3)}$
N31	3582$^{cd\ (2)}$	3084$^{a\ (11)}$	2592$^{a\ (12)}$	3589$^{a\ (4)}$	3085$^{ab\ (9)}$
N35	3175$^{abc\ (7)}$	3396$^{a\ (2)}$	4381$^{b\ (1)}$	3194$^{a\ (9)}$	2761$^{a\ (12)}$
N36	3175$^{abc\ (7)}$	3241$^{a\ (7)}$	3682$^{ab\ (7)}$	3564$^{a\ (5)}$	3374$^{ab\ (4)}$
N40	3127$^{a\ (12)}$	3347$^{a\ (4)}$	4381$^{b\ (1)}$	3108$^{a\ (10)}$	3532$^{b\ (2)}$
N41	3175$^{abc\ (7)}$	3265$^{a\ (6)}$	3174$^{ab\ (10)}$	3238$^{a\ (8)}$	3136$^{ab\ (7)}$
N48	3367$^{abcd\ (4)}$	3191$^{a\ (10)}$	3682$^{ab\ (7)}$	3563$^{a\ (6)}$	3040$^{ab\ (10)}$
N51	3715$^{d\ (1)}$	3239$^{a\ (8)}$	4156$^{b\ (6)}$	3752$^{a\ (1)}$	3703$^{b\ (1)}$
N52	3329$^{abcd\ (5)}$	3084$^{a\ (11)}$	3032$^{ab\ (11)}$	3632$^{a\ (3)}$	3116$^{ab\ (8)}$
NCo376	3206$^{abc\ (6)}$	3444$^{a\ (1)}$	3668$^{ab\ (9)}$	3469$^{a\ (7)}$	3358$^{ab\ (5)}$
Mean	**3310**	**3270**	**3793**	**3395**	**3228**
§	**a**	**a**	**b**		
§§	**ab**	**ab**		**b**	**a**

Means followed by the same letters do not differ significantly within site and ratoon. Numbers in brackets indicate ranking order of cultivars (1 representing the cultivar with the highest TTLAmax while 12 represents the cultivar with the lowest TTLAmax)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 5.4b) Cultivar rank correlation coefficients for TTLAmax at Amatikulu (AK) and Pongola (PG) across ratoons and sites

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.04	1		
AK P	-0.32	-0.20	1	
AK R	-0.32	0.15	0.85**	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, Significant at P<0.01

5.3.2 Leaf and leaf area development

The leaf number was determined at each measurement date and plotted against thermal time to calculate LAR. A linear fit was found to represent the data for all cultivars in all three sites. Examples of this are shown for the plant crop for cultivars N12 (Figure 5.4), N25 (Figure 5.5) and NCo376 (Figure 5.6). Generally similar rates were found at PG and BH, while consistently lower rates were obtained at AK. The results were slightly lower than leaf appearance rates reported by Inman-Bamber (1994) for cultivars NCo376 and N12 of 0.0092 and 0.0085 leaf $^{o}Cd^{-1}$. The lower leaf appearance rate at AK was thought to be associated with the moisture stress found in the coastal rainfed environment. This highlights the sensitivity of leaf appearance rate to moisture stress. BH consistently showed the highest LFmax while AK showed the lowest. A linear fit was also found to represent the data for the ratoon crop. Examples of this are shown for cultivars N12 (Figure 5.7) and NCo376 (Figure 5.8). Once again, the LAR at PG was higher than that observed at AK.

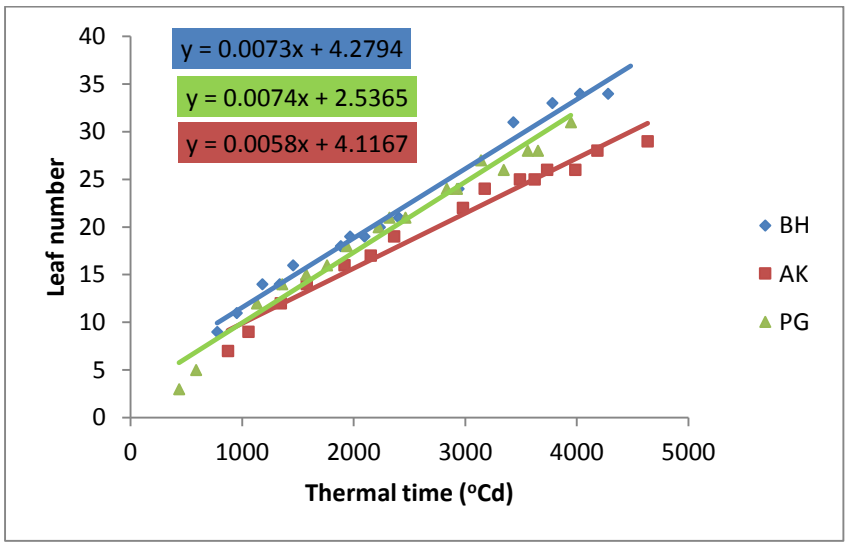

Figure 5.4 Maximum leaf number for cultivar N12 as a function of cumulative thermal time (base temperature 10°C) in the plant crops at BH, AK and PG

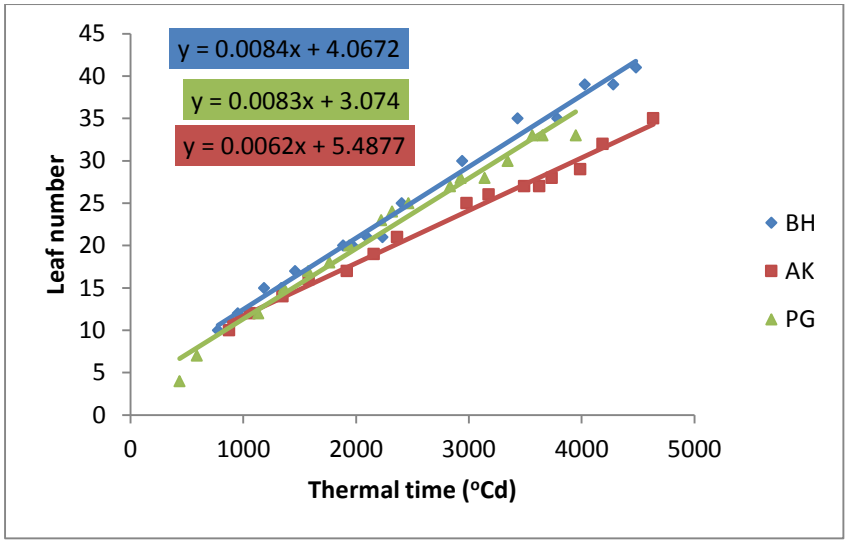

Figure 5.5 Maximum leaf number for cultivar N25 as a function of cumulative thermal time (base temperature 10°C) at BH, AK and PG

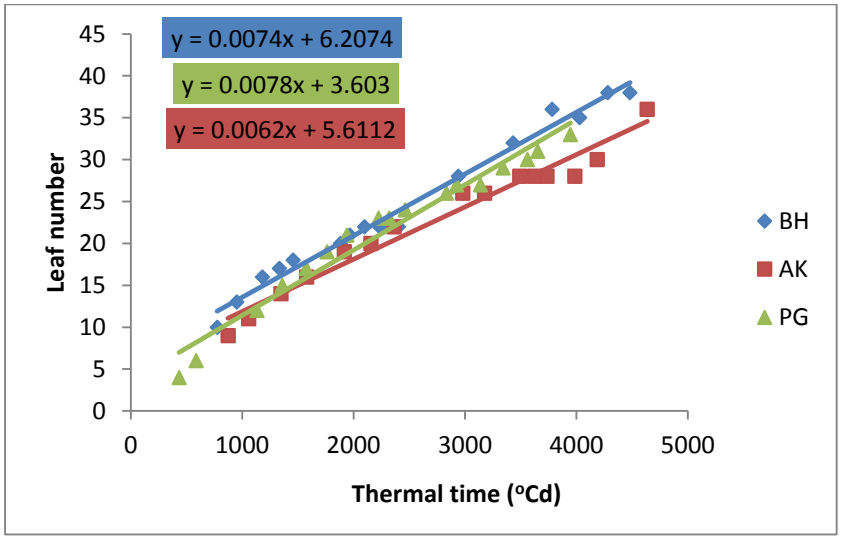

Figure 5.6 Maximum leaf number for cultivar NCo376 as a function of cumulative thermal time (base temperature 10°C) at BH, AK and PG

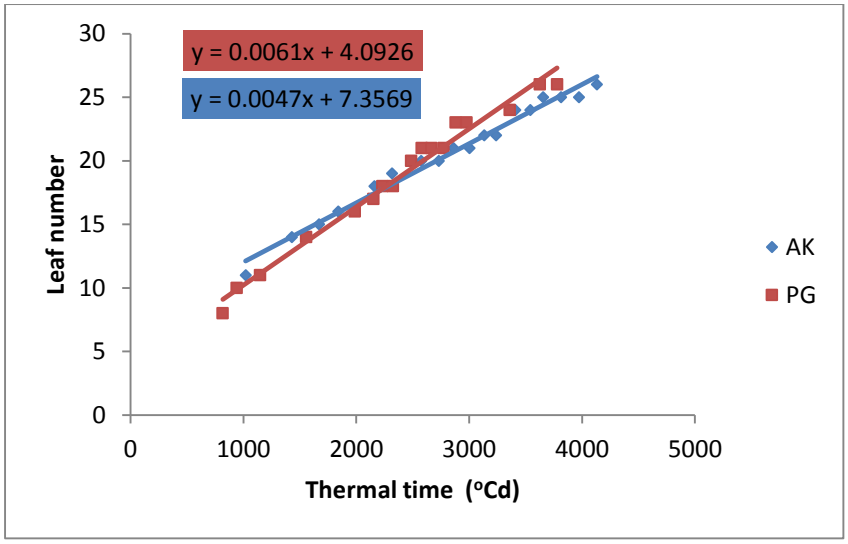

Figure 5.7 Maximum leaf number for first ratoon crop for cultivar N12 as a function of cumulative thermal time (base temperature 10 °C) at AK and PG

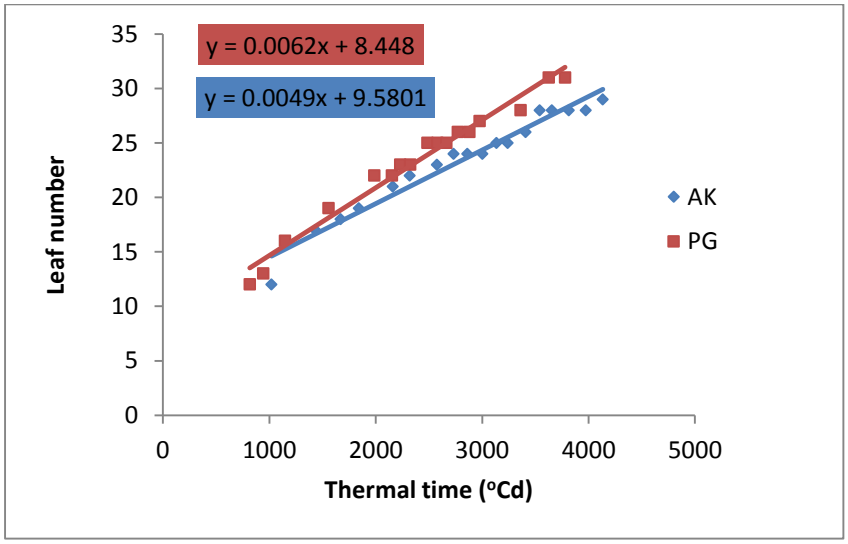

Figure 5.8 Maximum leaf number for first ratoon crop for cultivar NCo376 as a function of cumulative thermal time (base temperature 10°C) at AK and PG

Table 5.5 shows the mean square values from the combined ANOVA of the PG and AK trials for both the plant and first ratoon crops for LAR and LFmax. The main effects of cultivar (C), site (S) and crop cycle (R) were generally highly significant ($P<0.01$) for LAR and LFmax. The cultivar x site (C x S) interaction was significant for LFmax ($P<0.05$) (Table 5.5). The crop cycle x site (R x S) interaction was highly significant for LAR and LFmax. The cultivar x crop cycle (C x R) interaction was only significant ($P<0.05$) for LFmax. The three way C x S x R

interaction was highly significant (P<0.01) for LFmax, showing that the relative cultivar values for LFmax varied from one site to the next and from the plant to the ratoon crop. The mean square value for S was higher than the other mean square values for LAR; while R mean square was the highest for LFmax. The main effect of S and R therefore contributed most to total variance for the two traits.

Table 5.5 Mean square values from analysis of variance, combined across ratoons (plant and first ratoon) and sites (Pongola and Amatikulu) for leaf appearance rate (LAR) and maximum leaf number (LFmax)

Source	d.f.	Mean square values	
		LAR (x 10^{-8})	LFmax
		$^{o}Cd^{-1}$	-
Cultivar (C)	11	486**	62.26**
Site (S)	1	10270**	125.13**
Crop cycle (R)	1	3554**	371.30**
C x S	11	53	8.52*
C x R	11	41	9.00*
R x S	1	432**	81.38**
C x S x R	11	34	7.81**

**, *; Significant at P<0.01 and P<0.05, respectively.

Table 5.6 shows the mean square values from ANOVA of the PG, AK and BH plant crop only. The main effects of C and S were highly significant for all traits (Table 5.6). The C x S interaction was highly significant for LFmax (P<0.01) and significant for LAR (P<0.05), showing that the cultivar values for all traits differed by site for the plant crop. The main effects (C and S) mean squares were higher than the C x S interaction mean square value for most traits. The main effect of S contributed most to total variance for the two traits.

Table 5.6 Mean square values from analysis of variance for plant crop, combined across sites (Bruynshill, Pongola and Amatikulu) for leaf appearance rate (LAR) and maximum leaf number (LFmax)

Source	d.f.	Mean square values	
		LAR (x 10^{-8})	LFmax
		$^{o}Cd^{-1}$	-
Cultivar (C)	11	37.78**	81.78**
Site (S)	2	491.20**	399.65**
C x S	22	3.85*	19.90**

**, *; Significant at P<0.01 and P<0.05, respectively.

5.3.2.1 Leaf appearance rate (LAR)

In the plant crop the highest mean LAR occurred at PG (0.0080 leaf $^{o}Cd^{-1}$) and was followed by BH (0.0079 leaf $^{o}Cd^{-1}$) and AK (0.0057 leaf $^{o}Cd^{-1}$) respectively (Table 5.7a). Water stress has a negative effect on internode elongation, thus reducing the rate at which new leaves appear. The cultivar N35 had the highest LAR across all three sites; while cultivar N12 and NCo376 had relatively low leaf appearance rates across the three sites. This suggests that these cultivars may be ideal indicator cultivars for this trait in future studies. The rates of leaf appearance were in the range of 0.0068 - 0.0090, 0.0056 - 0.0069 and 0.0062 - 0.0090 leaf $^{o}Cd^{-1}$ for PG, AK and BH respectively. The correlation between AK and PG plant crop was highly significant (P<0.01) and high (r=0.84), showing that LAR is stable across sites in the plant crop.

The ratoon crop at PG (0.0068 leaf $^{o}Cd^{-1}$) had a higher mean LAR than that at AK (0.0056 leaf $^{o}Cd^{-1}$), which was consistent with the response observed in the plant crop. The mean LAR was lower for the ratoon crop than for the plant crop at AK and PG, varying from 0.0045 - 0.0068 $^{o}Cd^{-1}$ in AK and 0.0052 - 0.0079 $^{o}Cd^{-1}$ at PG. Similarly to the plant crop; cultivar N35 had the highest LAR for AK and PG respectively; while cultivars NCo376 and N12 had the lowest mean LAR at both sites. The cultivar rank correlation between PG and AK for the ratoon crop was strong and significant (r=0.71) indicating good stability of the trait across sites in the ratoon crop as well.

It was generally found that cultivar rankings were similar across ratoons within a site. All cultivars were ranked similarly between the plant and ratoon crop at AK and PG. The correlation value was significant and high within sites across ratoons, with values of 0.75 and

0.84 for PG and AK respectively (Table 5.7b). Cultivar characterisation for LAR in ratoons can therefore be adequately represented from information gathered in the plant crop in future experiments.

Table 5.7a) Leaf appearance rates (LAR) (leaf $^{o}Cd^{-1}$) of 12 cultivars at Amatikulu (AK), Pongola (PG) and Bruynshill (BH) for the plant crop and first ratoon

	Plant crop			First ratoon	
Cultivars	**PG**	**AK**	**BH**	**PG**	**AK**
N12	$0.0068^{a(12)}$	$0.0057^{ab(11)}$	$0.0062^{a(12)}$	$0.0059^{abc(10)}$	$0.0045^{a(12)}$
N19	$0.0078^{bcd(8)}$	$0.0060^{ab(8)}$	$0.0082^{cd(5)}$	$0.0069^{cdef(6)}$	$0.0056^{bcd(6)}$
N25	$0.0080^{cde(5)}$	$0.0059^{ab(10)}$	$0.0080^{bcd(6)}$	$0.0073^{def(4)}$	$0.0055^{bcd(7)}$
N31	$0.0078^{bcd(8)}$	$0.0060^{ab(8)}$	$0.0078^{bc(7)}$	$0.0065^{bcd(9)}$	$0.0054^{bcd(8)}$
N35	$0.0090^{f(1)}$	$0.0069^{c(1)}$	$0.0090^{e(1)}$	$0.0079^{f(1)}$	$0.0068^{e(1)}$
N36	$0.0083^{de(4)}$	$0.0065^{bc(3)}$	$0.0084^{cde(4)}$	$0.0069^{cdef(6)}$	$0.0062^{de(2)}$
N40	$0.0075^{bc(10)}$	$0.0061^{abc(7)}$	$0.0073^{b(11)}$	$0.0058^{ab(11)}$	$0.0053^{bc(10)}$
N41	$0.0085^{ef(2)}$	$0.0069^{c(1)}$	$0.0085^{de(3)}$	$0.0068^{bcde(8)}$	$0.0060^{cd(4)}$
N48	$0.0080^{cde(5)}$	$0.0062^{abc(5)}$	$0.0077^{bc(9)}$	$0.0077^{ef(2)}$	$0.0054^{bcd(8)}$
N51	$0.0085^{ef(2)}$	$0.0062^{abc(5)}$	$0.0078^{bc(7)}$	$0.0070^{def(5)}$	$0.0057^{bcd(5)}$
N52	$0.0080^{cde(5)}$	$0.0064^{abc(4)}$	$0.0089^{e(2)}$	$0.0074^{def(3)}$	$0.0062^{de(2)}$
Nco376	$0.0073^{ab(11)}$	$0.0056^{a(12)}$	$0.0074^{b(10)}$	$0.0052^{a(12)}$	$0.0051^{ab(11)}$
Mean	**0.0080**	**0.0057**	**0.0079**	**0.0068**	**0.0056**
§	**b**	**a**	**b**		
§§	**d**	**b**		**c**	**a**

Means followed by the same letters do not differ significantly within site and crop cycle. Numbers in brackets indicate ranking order of cultivars (1 representing the cultivar with the highest LAR while 12 represents the cultivar with the lowest LAR)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 5.7b) Cultivar rank correlation coefficients for LAR at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.75*	1		
AK P	0.84**	0.57	1	
AK R	0.86**	0.71*	0.84**	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, *; Significant at P<0.01 and P<0.05, respectively.

5.3.2.2 Maximum leaf number (LFmax)

The highest LFmax for the plant crop was obtained at BH followed by that at AK and PG respectively (Table 5.8a). The LFmax ranged from 27.0 to 37.0, 28.3 to 35.8 and 27.3 to 43.0 at PG, AK and BH respectively. The higher LFmax at BH was due to the longer growing season. Cultivar N35 had the highest LFmax in PG and BH, and the second highest at AK, showing that this trait is expressed consistently with this cultivar. Despite the significant C x S interaction relative ranking for cultivars N12, N35, N48 and NCo376 was similar across sites for the plant crop. This is enhanced by the moderately strong (r=0.62) and significant rank correlation at PG and AK for the plant crop, suggesting that this trait is fairly stable across sites for the plant crop and that grouping cultivars into low, moderate, and high range values for LFmax may be possible.

PG had a higher LFmax in the ratoon crop than observed at AK, which was in contrast to the response observed in the plant crop. Similarly to the plant crop the LFmax of the ratoon crop was highest for cultivar N35 at both PG and AK (Table 5.8a), confirming that this cultivar may be a good indicator cultivar for this trait in future experiments. The LFmax for cultivar N35 was similar across sites and ratoons. Cultivar N12 produced the lowest LFmax at AK and PG. The LFmax values for the ratoon crop ranged from 25.0 to 31.5 at PG and 23.0 to 33.5 at AK. The overall mean LFmax at both sites was lower for the ratoon crop than the plant crop. The correlation coefficient between the ratoon crops at AK and PG was weak and non-significant (r=0.47). Cultivar characterisation for LFmax in the ratoon is therefore unlikely to be accurate from a single site experiment.

There was generally a weak and non-significant correlation (r=0.37) between ratoons at PG (Table 5.8b). At AK, most cultivars (i.e. N12, N19, N25, N31, N35, N36, N48, and N51) had similar rankings in the plant and ratoon crop (Table 5.8a). The relationship between these cultivar rankings was moderately strong (r=0.71) and significant (Table 5.8b). The weak and non-significant correlation between ratoons at PG (r=0.37) meant that cultivars cannot be characterised from information gathered from the plant crop only. However, the strong correlation at AK meant that accurate data can be collected from one crop only. The reason for the strong correlation between crops at AK compared with the weak correlation between crops at PG is unclear.

Table 5.8a) Maximum leaf number (LFmax) of 12 cultivars at Amatikulu (AK), Pongola (PG) and Bruynshill (BH) for the plant crop and first ratoon

	Plant crop			First ratoon	
	PG	**AK**	**BH**	**PG**	**AK**
N12	28.0$^{ab(9)}$	28.3$^{a(12)}$	31.5$^{abc(10)}$	25.0$^{a(12)}$	23.0$^{a(12)}$
N19	27.5$^{ab(11)}$	32.8$^{b(7)}$	39.3$^{de(2)}$	28.5$^{bcde(7)}$	28.8$^{bc(6)}$
N25	31.0$^{cd(4)}$	33.0$^{b(6)}$	39.0$^{de(3)}$	30.3$^{def(3)}$	28.3$^{bc(8)}$
N31	27.0$^{a(12)}$	32.8$^{b(7)}$	27.3$^{a(12)}$	27.8$^{bcd(9)}$	28.5$^{bc(7)}$
N35	37.0$^{f(1)}$	35.5$^{cd(2)}$	43.0$^{e(1)}$	31.5$^{f(1)}$	33.5$^{d(1)}$
N36	28.8$^{abc(7)}$	33.8$^{bcd(3)}$	36.8$^{cd(6)}$	28.5$^{bcde(7)}$	30.5$^{c(2)}$
N40	30.0$^{bcd(5)}$	32.0$^{b(11)}$	35.8$^{bcd(7)}$	26.0$^{ab(11)}$	28.3$^{bc(8)}$
N41	33.8$^{e(2)}$	35.8$^{d(1)}$	35.3$^{bcd(9)}$	29.5$^{cdef(4)}$	30.0$^{bc(4)}$
N48	28.3$^{ab(8)}$	32.3$^{b(10)}$	35.5$^{bcd(8)}$	30.8$^{ef(2)}$	27.5$^{b(11)}$
N51	29.8$^{bcd(6)}$	33.8$^{bcd(3)}$	38.8$^{de(4)}$	29.0$^{cdef(5)}$	30.0$^{bc(4)}$
N52	28.0$^{ab(9)}$	32.8$^{b(7)}$	31.0$^{ab(11)}$	29.0$^{cdef(5)}$	30.5$^{c(2)}$
Nco376	31.8$^{de(3)}$	33.3$^{bc(5)}$	37.0$^{cd(5)}$	27.3$^{abc(10)}$	28.0$^{bc(10)}$
Mean	**30**	**33**	**36**	**29**	**27**
§	**a**	**b**	**c**		
§§	**b**	**c**		**a**	**a**

Means followed by the same letters do not differ significantly within site and crop cycle. Numbers in brackets indicate ranking order of cultivars (1 representing the cultivar with the highest LFmax while 12 represents the cultivar with the lowest LFmax)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 5.8b) Cultivar rank correlation coefficients for LFmax at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**
PG R	0.37		
AK P	0.62*	0.55	
AK R	0.22	0.47	0.71*

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

* Significant at P<0.05

5.3.3 Maximum leaf area index (LAI)

Table 5.9 shows mean square values for maximum LAI for the plant and ratoon crops combined across AK and PG. The mean square values for the main effects of cultivar (C) and site (S) were highly significant (P<0.01) while the effect of crop cycle (R) was found to be significant (P<0.05) (Table 5.9). The R x S interaction was also highly significant (P<0.01).The C x S interaction and the three-way C x S x R interaction were found not to be significant. The mean squares for the C x S were larger than the other cultivar interaction mean squares; meaning that site had a greater effect on the LAI of cultivars than ratoons. The mean square value for S was much higher than the other sources, once again indicating S as the most important source of variation.

Table 5.9 Mean squares for leaf are index (LAI) of different sugarcane cultivars under two environments (Pongola and Amatikulu) and seasons (plant and ratoon crop)

Source of variation	d.f.	LAI
Cultivar (C)	11	0.0046**
Site (S)	1	0.0208**
Crop cycle (R)	1	0.0038*
C x S	11	0.0008
C x R	11	0.0007
R xS	1	0.0190**
C x S x R	11	0.0007

**, *; Significant at P<0.01 and P<0.05, respectively.

Table 5.10 shows the mean square values from ANOVA of the PG, AK and BH plant crops only. The mean square values for main effects of C and S were highly significant for LAI (Table 5.10). The C x S interaction was not significant, showing that the LAI values for cultivars did not differ by location. The S was the most important source of variation.

Table 5.10 Mean square values from analysis of variance for plant crop, combined across sites (Bruynshill, Pongola and Amatikulu) for LAI

Source	d.f.	LAI
Cultivar (C)	11	0.0072**
Site (S)	2	0.0814**
C x S	22	0.0015

**; Significant at P<0.01.

The mean LAI of the plant crop at AK (0.11) was lower than that at BH (0.19) and PG (0.15) (Table 5.11a). Leaf area is highly sensitive to water stress, and a reduction in leaf area results in reduced LAI. The low temperature conditions in BH and irrigated conditions at PG may have reduced stress on the plants. In this study, the lower LAI obtained at AK in this study was probably due to moisture stress brought about by poor precipitation in the season. The cultivar N36 had the highest LAI across all sites while cultivars NCo376, N41 and N31 had the lowest LAI at PG, AK and BH, respectively. The cultivars N12, N19, N25, N35, N40, N52 and NCo376 had similar rankings at both PG and AK. The relationship between these cultivars was moderately strong and significant (r=0.69) (Table 5.11b), showing that LAI may be considered as a fairly stable trait across sites in the plant crop.

In the ratoon crop mean LAI was similar at PG and at AK. LAI values ranged from 0.09 to 0.16 at AK and at PG. Cultivars had similar rankings for LAI within each site. Mean LAI was generally lower than the plant crop at PG, while LAI was higher for the ratoon crop than the plant crop at AK. The moderate correlation (r=0.56) between cultivars among AK and PG for the ratoon crop (Table 5.11b) meant that cultivar characterisation may be represented adequately with information from one site only.

Within a site, it was generally found that cultivar rankings were similar across crops at AK but different at PG. For example cultivars N12, N19, N25, N35, N36, N41, N48, N51, and NCo376 showed consistent rankings across ratoons at AK (Table 5.11a). This is further supported by the high and significant (r=0.76) correlation value at AK. Despite non-significant C x R interactions, cultivar rankings at PG were different across ratoons. The correlation coefficient was weak and non-significant among cultivars at PG (r=0.48) suggesting that LAI cannot be adequately represented with values from the plant crop only. The change in ranks across ratoons at PG means that LAI was highly influenced by the environment.

Table 5.11a) Maximum leaf area index (LAI) for 12 cultivars at Amatikulu (AK), Pongola (PG) and Bruynshill (BH) for plant and ratoon crop

	Plant crop			First ratoon	
Cultivar	**PG**	**AK**	**BH**	**PG**	**AK**
N12	0.12$^{ab\ (11)}$	0.10$^{ab\ (9)}$	0.18$^{abc\ (8)}$	0.09$^{a\ (12)}$	0.11$^{ab\ (8)}$
N19	0.15$^{bcd\ (7)}$	0.10$^{abc\ (9)}$	0.20$^{abc\ (5)}$	0.14$^{bcd\ (3)}$	0.10$^{ab\ (10)}$
N25	0.16$^{cde\ (4)}$	0.12$^{abc\ (4)}$	0.25$^{c\ (1)}$	0.12$^{abc\ (5)}$	0.13$^{ab\ (4)}$
N31	0.13$^{abc\ (10)}$	0.11$^{abc\ (7)}$	0.13$^{a\ (12)}$	0.12$^{abcd\ (5)}$	0.13$^{ab\ (4)}$
N35	0.18$^{de\ (2)}$	0.12$^{abc\ (4)}$	0.23$^{bc\ (3)}$	0.16$^{d\ (1)}$	0.14$^{ab\ (3)}$
N36	0.19$^{e\ (1)}$	0.14$^{c\ (1)}$	0.25$^{c\ (1)}$	0.13$^{abcd\ (4)}$	0.16$^{b\ (1)}$
N40	0.18$^{de\ (2)}$	0.13$^{bc\ (2)}$	0.23$^{bc\ (3)}$	0.12$^{abc\ (5)}$	0.11$^{ab\ (8)}$
N41	0.15$^{bcd\ (7}$)	0.08$^{a\ (11)}$	0.14$^{a\ (11)}$	0.11$^{abc\ (9)}$	0.09$^{a\ (12)}$
N48	0.14$^{bc\ (9)}$	0.13$^{bc\ (2)}$	0.20$^{abc\ (5)}$	0.15$^{cd\ (2)}$	0.13$^{ab\ (4)}$
N51	0.16$^{cde\ (4)}$	0.11$^{abc\ (7)}$	0.20$^{abc\ (5)}$	0.11$^{abc\ (9)}$	0.12$^{ab\ (7)}$
N52	0.16$^{cde\ (4)}$	0.12$^{abc\ (4)}$	0.16$^{ab\ (9)}$	0.12$^{abcd\ (5)}$	0.16$^{b\ (1)}$
NCo376	0.11$^{a\ (12)}$	0.08$^{a\ (11)}$	0.16$^{ab\ (9)}$	0.11$^{ab\ (9)}$	0.10$^{ab\ (10)}$
Mean	**0.15**	**0.11**	**0.19**	**0.12**	**0.12**
§	**b**	**a**	**c**		
§§	**b**	**a**		**a**	**a**

Means followed by the same letters do not differ significantly within site and crop cycle. Numbers in brackets indicate ranking order of cultivars (1 representing the cultivar with the highest LAI while 12 represents the cultivar with the lowest LAI)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 5.11b) Cultivar rank correlation coefficients for maximum LAI at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**
PG R	0.48		
AK P	0.69*	0.66*	
AK R	0.55	0.56	0.76*

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

* Significant at P<0.05.

5.3.4 Heritability

Estimates of variance components and broad sense heritability of the leaf development traits are shown in Table 5.12. The genetic (C) variance was the highest source of variation for LFmax and LAI, while the environmental (E) variance was the highest for LAR, LAmax and TTLAmax. The C variance was higher than the C x S, C x R and C x S x R variance components for most of the traits. This suggests that these traits are, to a large extent, genetically controlled. The E variance was greater than the C variance for most of the traits. This indicates that the environment had an important role in the expression of these traits therefore the diverse cultivars can provide sufficient information for use in breeding programmes.

Broad sense heritability estimates were high for most traits (84-99%) with the exception of TTLAmax. These were higher than estimates reported by Olaoye et al. (2012) of 63-78% on number of leaves, leaf length and leaf area. The high heritability estimates suggest that a large portion of total variance is heritable. These characters can be used for selection in sugarcane breeding programmes. They may also be good candidates for model-wise exploration of G x E interactions in sugarcane.

Table 5.12 Variance components and broad sense heritability for maximum leaf number (LFmax), leaf appearance rate (LAR), maximum leaf area (LAmax), thermal time to reach max leaf area (TTLAmax) and leaf area index (LAI)

Source of variation	LAR (x 10^{-8})	LFmax	LAmax	TTLAmax	LAI
C	26.65	3.29	1980.94	0	0.0015
C x R	0.81	0.15	0	23450	0
C x S	2.32	0.09	0	21789	0
C x S x R	1.15	1.19	228.25	0	0.0007
E	29.45	3.07	2003.50	139393	0.0012
H^2	**0.88**	**0.84**	**0.99**	**0**	**0.98**

5.4 Conclusions

The cultivar trait coefficients used in Canegro are based exclusively on experimental work done on the cultivar NCo376. Values of trait coefficients for other contrasting sugarcane cultivars are not available, and as a result, the Canegro model is unable to simulate performance of the current range of available commercial cultivars. The ability to model cultivar differences could assist with breeding and cultivar selection. The correlation values for LAmax within sites in the plant and ratoon crops at AK and PG were high and significant, suggesting that this trait is consistent (stable) across sites. This means that cultivar characterisation for LAmax in ratoons may be possible with information from the plant crop only at one location. The trait TTLAmax showed poor rank correlations in the plant and ratoon crop at AK and PG, indicating that the trait is not stable. A strong relationship between cultivar rankings across ratoons at AK existed, in contrast a weak correlation existed between ratoons at PG, indicating that the TTLAmax cannot be adequately represented with values from the plant crop only.

The rank correlations for LAR between sites and across ratoons within sites were highly significant and strong; showing that LAR is the most stable of all traits. All cultivars were ranked similarly within sites across crops. Cultivar characterisation for LAR can thus be adequately represented from information gathered from one crop cycle at a single-site. The LFmax showed consistent rankings across sites in the plant crop; but poor correlations between sites in the ratoon crop. This suggests that this trait is fairly stable across sites in the plant crop and that grouping of cultivars into low, moderate, and high range values for LFmax may be a possibility. LFmax showed a strong rank correlation across crops at AK and a low rank correlation at PG, meaning that accurate data can be collected from one crop only at AK.

Due to the moderately strong rank correlation across sites, the LAI is considered as a fairly stable trait across sites in the plant and ratoon crop. This meant that cultivar characterisation may be represented adequately with information from one crop cycle at one site only. At AK, the rank correlation was strong but weak at PG. Some cultivars such as N12 and NCo376 showed consistent rankings across ratoons at AK, suggesting that they can be used as group representatives for LAI in future studies.

Most traits generally had a high heritability (ranging from 0.84-0.99). This suggests that leaf development traits could be selected for in sugarcane yield improvement programmes. If these

traits are found to be associated with cane and sugar yields, it may be possible to select for them or investigate ideal trait combinations for different environments i.e. ideotype development. Follow-up studies should involve the statistical grouping of cultivars into categories for each trait, and the determination of mean values for each category. These mean values should then be incorporated and validated using the Canegro crop model.

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CHAPTER 6

Quantifying sugarcane cultivar differences and genotype x environment interactions for biomass and yield traits

Abstract

Sugarcane models, like the Canegro model, can estimate yield and water use (and many other traits that affect these) given certain weather and soil information. The Canegro model has the ability to predict growth and development for separate cultivars using a genetic (cultivar) coefficient for each cultivar. However, currently the only cultivar coefficient in the Canegro model is for NCo376. The objective of this chapter was to investigate the G x E interactions, and to determine the stability and heritability of cane and sucrose yields as well as some of the final harvestable biomass components for 12 cultivars that represent a wide range of sugarcane phenotypes. Cultivar coefficients for stable and heritable traits (cane, sucrose yield, and biomass components) would be useful for model simulations.

Three cultivar trials were established in 2011 at three sites on SASRI research farms and commercial fields; AK, PG, and BH. Data collected included population counts on selected lines and other yield-related information obtained by sampling of 12-16 stalks per plot. ERC%, ERC yield, cane yield, total biomass, and brown and green trash biomass were measured. Broad sense heritability (H^2) was determined for these traits from the variance components obtained from the analyses of variance.

The mean squares for C x S for cane yield, ERC% and ERC yield greatly exceeded those for the C x S x R and C x R interactions. The ERC% was relatively stable across sites and crop cycles, suggesting that most cultivars in the study would be acceptable indicator cultivars for future studies. In general, cane and ERC yields were stable across crop cycles at PG (irrigated), but not at AK (rainfed). For total biomass, higher rank correlations were found between crops at PG than at AK, suggesting that total biomass may be stable across crop cycles under irrigated conditions only. The amounts of green and brown trash generally showed poor rank correlations between sites and crop cycles within sites, suggesting that they are strongly influenced by the environment.

Heritability (H^2) estimates ranged from 0 for green trash to 0.93 for total biomass. The C x R, C x S, and C x S x R interaction components had less influence on ERC% and total biomass than the genotypic variance, while C x S had a higher influence than the genotypic variance on cane yield, ERC yield, brown trash, and green trash. Environmental variance was lower than the genotypic variance for total biomass. The high H^2 estimates confirmed that selection for cane yield, ERC%, and total biomass would be effective for identifying new cultivars with high values for these traits. Currently, it is the practice of many seletion programs to make selections on these traits.

6.1 Introduction

Sugarcane yield is expressed as tonnes per hectare of fresh cane (cane yield) and/or sucrose (Donaldson, 2008). Cane yield is dependent on the total number of stalks and the weight of individual stalks. More than 50% of the biomass produced at harvest is partitioned into the stalk, of which 30% is dry matter, consisting of 60% sucrose and 40% fibre (Moore and Maretzki, 1996). This partitioning of biomass into sucrose helps determine the estimated recoverable crystal (ERC) yield which is the final yield of sucrose in the millable stalk (Robertson et al., 1996). The ERC yield in sugarcane is obtained by multiplying cane yield and estimated recoverable crystal content (ERC %).

Biomass yields and composition and ultimately sugar yields vary depending on various factors such as cultivar (genotype), year, age, crop cycle, harvest period, and location (Kim et al., 2011). There are numerous examples of cultivar differences in sugarcane yield, yield components and biomass partitioning. Khan et al. (2004) reported significant differences for cane yield and its yield components studied. Highest cane yield was produced by AEC86-347 (174.40 ton ha^{-1}) followed by BL4 (136.13 ton ha^{-1}) and CP 67-417 (129.74 ton ha^{-1}). Redshaw and Nuss (2001) compared yields of South African cultivars across sites and harvesting cycles and revealed significant cultivar differences across sites. The highest ERC yields were generally observed at Pongola, Komatidraai in Mpumalanga, and Ubombo in Swaziland. Most of these cultivars had the highest ERC yields when harvested early to mid-season. Arain et al. (2011) and Gomathi et al. (2013) also observed significant differences in mean values of different cultivars in cane yield and cane components. These factors result in yield differences that make selection in breeding programmes a challenge. Breeders, therefore, need to expose a range of cultivars to a wide range of conditions (crop cycle, year, site etc.) and select for

cultivars that are most productive, stable, and have high heritability for yield traits across sites (Zhou, 2005). Knowledge of stability and heritability of cane yield and biomass components allows for genetic improvement and will help improve model simulations.

Crop simulation models can be useful in plant breeding programmes as they may help identify ideal cultivars for a range of environments (Yin et al., 2003). The Canegro model is able to identify traits that contribute to increases in potential yield by using daily weather data, cultivar, soil properties, and management input data to simulate sugarcane crop growth to predict cane yield, sucrose yield, crop biomass, nitrogen and water use (Lisson et al., 2005). Briefly, the Canegro model calculates daily increments in total biomass using PAR. A constant fraction of aboveground biomass is partitioned to stalk growth, regardless of environmental conditions (Singels and Bezuidenhout, 2002). The fraction of stalk growth allocated to structural growth is determined by the ratio of mature to immature stalks as well as water status and temperature conditions. Sucrose accumulation in the model is calculated on an internode basis. The partitioning of stalk dry mass between stalk structure and sucrose is regulated by sink capacity, the thermal age of the internode, and characteristics of the cultivar. Sucrose partitioning occurs after the partitioning of leaves, roots, and stalk fibre.

The previous two chapters focused on the quantification of lower-level sugarcane cultivar traits for 12 cultivars and assessed their potential use for model-assisted breeding. Those lower-level traits integrate with each other to eventually determine the partitioning of biomass into cane and sucrose yields. It follows, therefore, that an assessment of the lower-level traits would be incomplete without a link to the final harvestable cane or sucrose yield. Therefore, the objectives of this chapter were to quantify and investigate the G x E interactions and to determine the stability and heritability of cane and sucrose yield as well as some of the final harvestable biomass components for 12 diverse sugarcane cultivars.

6.2 Material and methods

The details of all trials, including site characteristics, cultivars, experimental designs, plot dimensions, and crop characteristics were described in detail in Chapter 3. For brevity, only the details of the data collection and analysis relevant to this chapter are briefly mentioned below.

Three to four rows from each plot were harvested manually. The total stalk weight from these three-four rows of each harvested plot was used to calculate cane yield (ton ha^{-1}). A random sample of 12 to 16 stalks was taken from the selected plot rows in each plot at harvest. The samples were partitioned into brown leaves (defined as the dead leaf material), green leaves, and millable stalk biomass (including meristems- biomass above the stalks' natural breaking points). Stalk population were counted on these rows. Total biomass (ton ha^{-1}) was determined as the sum of the biomasses from the sample rows and the 12 to 16 stalks of sugarcane used to estimate brown and green leaf biomasses. The mass of each individual plant part was also calculated and expressed in tonnes per hectare (ton ha^{-1}). The 12 to 16 stalks sampled were sent to SASRI's Mount Edgecombe mill room where the sucrose, fibre, and non-sucrose components of each sample were determined. The ERC% is calculated from the sucrose, non-sucrose, and fibre contents, and represents the cane payment system used in South Africa. ERC yield was the product of cane yield and ERC%.

The data for the parameters was analysed using Genstat version 14 (VSN International Ltd, 2011). A combined ANOVA was performed for AK and PG where main and interaction effects were tested. The main effects were cultivar, site, and ratoon (fixed). The trial at BH was harvested on a 24-month cutting cycle, and as a result, was only harvested for the plant crop. A separate ANOVA was therefore also conducted for BH, AK, and PG to test the main effects of site and cultivar for the plant crop only. Treatment mean comparisons were made using Duncan's LSD test at P=0.05. The variance components were calculated using the mean squares from the ANOVA. Estimates of broad sense heritability for the different traits were calculated using the variance components method. Broad sense heritability was determined from the equation: $h^2 = \sigma^2 g / \sigma^2 p$ as shown in Chapter 3.

6.3 Results and discussion

6.3.1 Yield and yield components

The mean ERC yield (ton ha^{-1}) for the plant crop across all three sites for each of the 12 cultivars is given in Figure 6.1. There was large variation in ERC yield among the three sites. Bruynshill generally had higher yields, followed by Pongola and Amatikulu. This could be due to the 24-month harvest cycle at Bruynshill, while the lower ERC yields at Amatikulu were attributed to the lower rainfall conditions. The means were significantly higher for most of the cultivars at Bruynshill than at Pongola and Amatikulu with the exception of cultivars N35 and N41. Cultivar means were significantly higher at Pongola than at Amatikulu for all cultivars.

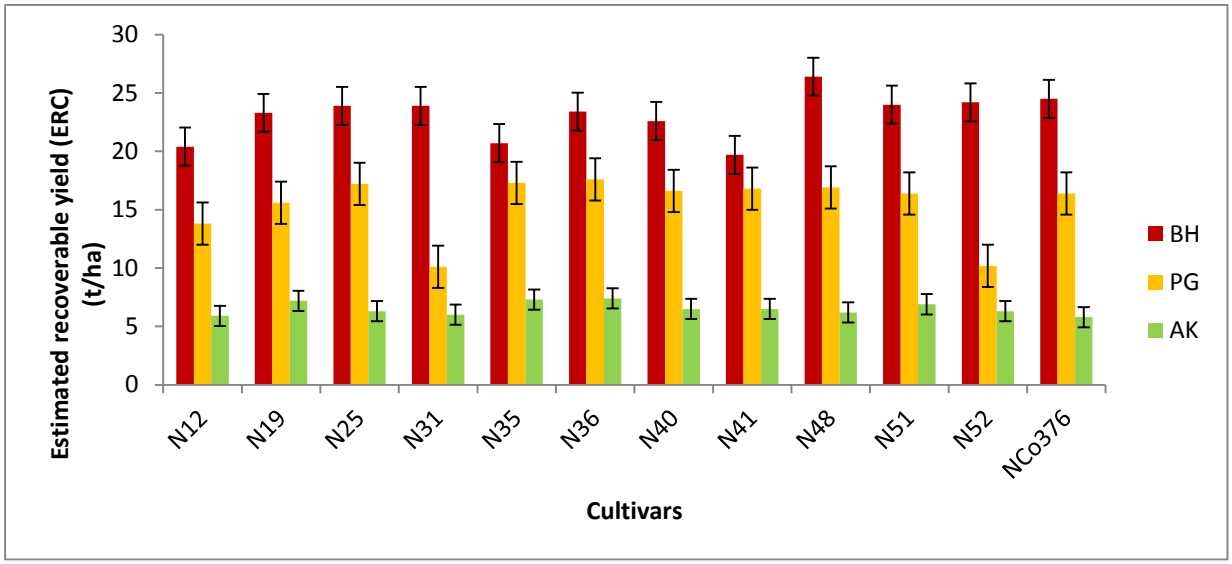

Figure 6.1 Estimated recoverable crystal yield (ERC) for different cultivars across sites (AK, PG and BH)

Table 6.1 shows mean square values for cane yield, ERC%, and ERC yield from the combined ANOVA of the PG and AK trials for both the plant and first ratoon crops. The mean square values were highly significant for the main effects of site (S), ratoon (R), and cultivar (C) for all traits (Table 6.1). The mean square value for S was higher than that for the R and C for both cane and ERC yield. Therefore, S made the largest contribution to variation for cane and ERC yield. In contrast, the ERC% mean square value for R was larger than the other components suggesting that R had a larger effect on variation in ERC%.

The cultivar x site (C x S) interaction was highly significant for ERC% and ERC yield (P<0.01) (Table 6.1). This indicated the varying response of cultivars to site differences for those traits. The ratoon x site (R x S) interaction was highly significant for cane and ERC yields. The variance due to cultivar x ratoon (C x R) was not significant for all traits, showing that the relative cultivar values for these traits did not vary across ratoons. The three way C x S x R interaction was significant (P<0.05) for ERC%, showing that the relative cultivar values for this trait varied from one site to the next, and from the plant to the ratoon crop. The mean square for the effects of C x S greatly exceeded the C x S x R and C x R interaction mean squares for all three traits.

Table 6.1 Mean square values from analysis of variance, combined across the plant and first ratoon crop cycles and sites (Pongola and Amatikulu) for estimated recoverable crystal (ERC) yield, estimated recoverable crystal content (ERC%) and cane yield

	d.f.	Cane Yield	ERC%	ERC Yield
		ton ha^{-1}	%	ton ha^{-1}
Cultivar (C)	11	561.70**	22.44**	21.21**
Site (S)	1	159230.10**	16.74**	2087.77**
Crop cycle (R)	1	32746.10**	302.76**	74.25**
C x S	11	236.00	6.61**	16.70**
C x R	11	98.20	1.38	5.34
R x S	1	26064.60**	3.88	290.66**
C x S x R	11	30.70	2.09*	3.32

**, *; Significant at P<0.01 and P<0.05, respectively.

Table 6.2 shows the mean square values from ANOVA of the PG, AK, and BH plant crop only. The main effects (C and S) mean squares were highly significant (P<0.01) for all traits with the exception of the effect of C on ERC yield which was significant at P = 0.05. The mean square values were highly significant for the C x S interaction for ERC % and ERC yield (P<0.01) and significant for cane yield (P<0.05). Therefore the relative cultivar values for all traits differed by site for the plant crop. The main effect (C and S) mean square values exceeded the C x S interaction mean square for cane yield and ERC %, indicating that the main effects of C and S accounted for more variation than the C x S interaction for these two traits.

Table 6.2 Mean square values from analysis of variance of the plant crop, combined across sites (Bruynshill, Pongola and Amatikulu) for estimated recoverable crystal (ERC) yield, ERC %, and cane yield

	d.f.	Cane yield	ERC %	ERC yield
		ton ha^{-1}	%	ton ha^{-1}
Cultivar (C)	11	830.30**	12.30**	13.54*
Site (S)	2	179534.30**	44.41**	3328.13**
C x S	22	348.30*	5.13**	15.02**

**, *; Significant at P<0.01 and P<0.05, respectively.

6.3.1.1 Estimated recoverable crystal (ERC) yield

In the plant crop, mean ERC yield was highest at BH, followed by PG and AK, respectively (Table 6.3a). The longer growth period of 24 months of the crop at BH compared with the crops at AK and PG (12 months) could be the reason for the higher ERC yield. Various cultivars (such as N48, N36, N35, N25, N41 and N40) had a high ERC yield at PG. While cultivars N48, N52 and NCo376 had the highest ERC yield at BH. There were no differences among cultivars at AK. Mean ERC yield varied from 20.26 to 25.09 ton ha^{-1} at BH, 10.21 to 18.17 ton ha^{-1} at PG, and non-ignificant means varied from 5.83 to 7.48 ton ha^{-1} at AK. The differences in ERC yield across cultivars in all three sites for the plant crop was highly significant (P<0.01) (Table 6.2). Only cultivar N12 had similar rankings across all three sites (Table 6.3a). Although the correlation between PG and AK was low and non-significant (r=0.37), rankings of cultivars N31, N35, N36, N40, and N41 were similar at PG and AK (Table 6.3b).

In the ratoon crop, PG had a higher mean ERC yield than at AK, which was similar to the response observed in the plant crop (Table 6.3a). There were no differences in ERC yield among cultivars at AK. In the ratoon crop ERC yield ranged from 9.10 to 14.36 ton ha^{-1} at PG and the non-significant mean ERC yields at AK ranged from 6.73 to 9.25 ton ha^{-1}. Some cultivars (i.e. N25, N31, and N51) had particularly substantial contrasting rankings across the two sites. The correlation coefficient was low and non-significant (r=0.05) across sites for the ratoon crop, indicating a weak relationship between cultivar rankings for ERC yield. The ERC yield for the ratoon crop at PG was lower than the plant crop while the ratoon crop at AK had a higher ERC yield than the plant crop. This shows that ERC yield between a plant and ratoon crop varied between the irrigated (PG) and rainfed (AK) conditions, and this may be attributed to different seasonal variations at the two sites.

Within a site, a weak and non-significant correlation value of 0.27 was observed at AK across crop cycles (Table 6.3b). Cultivars N12, N31, N35, N40, N41, N51, N52, and NCo376 were ranked similarly in the plant and ratoon crops at PG. In addition, there was a moderately significant correlation coefficient of 0.57 at PG across crop cycles. This shows that in general, relative ERC yields of the cultivars tested were similar across ratoons at PG but not at AK. In addition a non-significant C x S interaction was found at PG (appendix A1). This may be due to the stabilizing effect of irrigation at PG, compared with variable cultivar responses to the rainfed seasonal conditions at AK.

Table 6.3a) Estimated recoverable crystal (ERC) yield of 12 cultivars across Bruynshill (BH), Pongola (PG), and Amatikulu (AK) for the plant and ratoon crop

	Plant crop			First ratoon	
	BH	**PG**	**AK**	**PG**	**AK**
N12	21.23^{ab} (11)	13.06^{ab} (10)	5.96^{a} (11)	9.12^{a} (11)	6.73^{a} (12)
N19	24.28^{ab} (5)	15.84^{bc} (9)	7.22^{a} (3)	12.83^{bc} (4)	7.37^{a} (9)
N25	23.17^{ab} (8)	17.52^{c} (4)	6.32^{a} (8)	14.36^{c} (1)	7.54^{a} (7)
N31	24.31^{ab} (4)	10.21^{a} (12)	6.07^{a} (10)	10.67^{ab} (10)	9.25^{a} (1)
N35	21.51^{ab} (10)	17.58^{c} (3)	7.38^{a} (2)	13.62^{bc} (2)	7.59^{a} (6)
N36	24.01^{ab} (6)	17.86^{c} (2)	7.48^{a} (1)	12.12^{abc} (7)	8.52^{a} (3)
N40	21.80^{ab} (9)	16.84^{c} (6)	6.52^{a} (6)	12.30^{bc} (6)	6.81^{a} (11)
N41	20.26^{a} (12)	16.97^{c} (5)	6.59^{a} (5)	13.47^{bc} (3)	8.23^{a} (4)
N48	25.09^{b} (1)	18.17^{c} (1)	6.25^{a} (9)	11.58^{abc} (8)	7.87^{a} (5)
N51	23.18^{ab} (7)	16.56^{c} (8)	6.99^{a} (4)	11.50^{abc} (9)	9.04^{a} (2)
N52	24.85^{b} (2)	10.41^{a} (11)	6.34^{a} (7)	9.10^{a} (12)	7.13^{a} (10)
NCo376	24.82^{b} (3)	16.58^{bc} (7)	5.83^{a} (12)	12.48^{bc} (5)	7.45^{a} (8)
Mean	**23.21**	**15.63**	**6.58**	**11.93**	**7.79**
§	**c**	**b**	**a**		
§§		**d**	**a**	**c**	**b**

Means followed by the same letters do not differ significantly within a site and crop cycle. Numbers in brackets indicate ranking of cultivars (1 representing the cultivar with the highest ERC yield, while 12 represents a cultivar with the lowest ERC yield)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 6.3b) Cultivar rank correlation coefficients for estimated recoverable crystal (ERC) yield at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	PG R	AK P
PG R	0.57*		
AK P	0.37	0.34	
AK R	0.22	0.05	0.27

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, *; Significant at P<0.01 and P<0.05, respectively.

6.3.1.2 Estimated recoverable crystal content (ERC%)

In general, the ERC% at BH was highest followed by AK and PG respectively (Table 6.4a). This may be due to the longer growing season (24 months) at BH. The higher ERC% at AK than at PG in the plant crop may be attributed to water stress, which is known to increase sucrose content. The ERC% ranged from 12.2 to 14.10% at BH, 7.33 to 13.43% at PG, and 9.85 to 13.95% at AK. The cultivars N19, N25, N35, N36, N40, N41, N48, and N51 showed similarly high ERC% at PG, while cultivar N40 showed the highest ERC % at AK. Cultivar N52, NCo376 and N25 showed the lowest ERC% at AK and cultivar N52 and N31 had the lowest ERC % at PG. There was a moderately high and significant cultivar rank correlation (r=0.76) in the plant crop between AK and PG (Table 6.4b) suggesting that cultivar rankings were fairly stable across sites for this trait. The consistent rankings of cultivars and the strong correlation suggests that ERC% may be stable across sites.

The ratoon crop at AK had a higher mean ERC% than that at PG, which was similar to the response observed in the plant crop. Mean ERC% varied from 11.07 to 15.80% at PG and 12.47 to 15.38 % at AK. Cultivar N52 ranked lowest at AK and was amongst the low ranking cultivars at PG. This shows that the low ERC% in cultivar N52 is consistent across sites in the first ratoon, suggesting that N52 may be a good candidate for use as a low sucrose cultivar in future studies. Although correlation for cultivar ranking between AK and PG was weak (Table 6.4b), cultivars N12, N36, N41, N51, and NCo376 also had similar rankings across both sites. Overall ERC% mean obtained in the ratoon crops was higher than that of the plant crop at both sites. The low and non-significant (r=0.34) rank correlation between means at PG and AK in the ratoon crop showed that ERC % was generally not stable across sites in the ratoon.

Cultivar rankings for ERC% were similar across ratoons within sites (Table 6.4a). The ERC% rankings were similar for most cultivars in the two crop cycles at AK with the exception of cultivars N31, N35, N40, and N48. Similarly, cultivars N12, N25, N31, N36, N40, N41, N51, N52, and NCo376 had similar rankings between the plant and ratoon crop at PG (Table 6.4a). These preliminary results show that the correlation coefficient was moderate and significant (r=0.69) between crop cycles at AK and strong and highly significant (r=0.84) at PG (Table 6.4b). This shows that ERC% is a stable trait from plant to ratoon crops within a site, meaning that cultivar characterisation for this trait may be possible from single crop experiments. The fairly consistent rankings of cultivars across sites and ratoons also means that cultivar grouping into low, moderate, and high range values are feasible. These are preliminary results

Table 6.4a) Estimated recoverable crystal content (ERC%) of 12 cultivars at Amatikulu (AK), Bruynshill (BH) and Pongola (PG) in the plant and two locations in the ratoon crop

	Plant crop			**First ratoon**	
	BH	**PG**	**AK**	**PG**	**AK**
N12	12.55$^{ab\ (11)}$	10.90$^{b\ (10)}$	11.18$^{bcd\ (9)}$	13.22$^{bc\ (8)}$	14.25$^{bc\ (9)}$
N19	12.60$^{ab\ (10)}$	11.83$^{bc\ (6)}$	12.28$^{de\ (4)}$	15.03$^{def\ (3)}$	14.68$^{bc\ (6)}$
N25	13.88$^{bc\ (2)}$	11.68$^{bc\ (7)}$	10.65$^{abc\ (10)}$	13.95$^{bcde\ (6)}$	14.22$^{bc\ (10)}$
N31	13.38$^{abc\ (4)}$	7.70$^{a\ (11)}$	11.85$^{cde\ (6)}$	11.50$^{a\ (11)}$	15.38$^{c\ (1)}$
N35	13.10$^{abc\ (8)}$	12.45$^{bc\ (5)}$	12.55$^{e\ (2)}$	15.38$^{ef\ (2)}$	14.65$^{bc\ (7)}$
N36	13.85$^{bc\ (3)}$	13.38$^{c\ (2)}$	12.40$^{de\ (3)}$	14.60$^{cdef\ (4)}$	14.95$^{bc\ (3)}$
N40	13.38$^{abc\ (4)}$	13.43$^{c\ (1)}$	13.95$^{f\ (1)}$	15.80$^{f\ (1)}$	14.88$^{bc\ (4)}$
N41	14.10$^{c\ (1)}$	12.95$^{bc\ (4)}$	12.13$^{de\ (5)}$	14.47$^{cdef\ (5)}$	14.70$^{bc\ (5)}$
N48	13.32$^{abc\ (6)}$	13.00$^{bc\ (3)}$	11.68$^{bcde\ (7)}$	13.35$^{bcd\ (7)}$	15.18$^{bc\ (2)}$
N51	12.65$^{abc\ (9)}$	11.18$^{bc\ (8)}$	11.40$^{bcde\ (8)}$	12.70$^{ab\ (9)}$	14.55$^{bc\ (8)}$
N52	13.30$^{abc\ (7)}$	7.33$^{a\ (12)}$	9.85$^{a\ (12)}$	11.07$^{a\ (12)}$	12.47$^{a\ (12)}$
NCo376	12.22$^{a\ (12)}$	10.95$^{b\ (9)}$	10.53$^{ab\ (11)}$	12.40$^{ab\ (10)}$	14.08$^{b\ (11)}$
Mean	**13.19**	**11.40**	**11.70**	**13.62**	**14.50**
§	**b**	**a**	**a**		
§§		**a**	**a**	**b**	**c**

Means followed by the same letters do not differ significantly within a site and crop cycle. Numbers in brackets indicate ranking of cultivars (1 representing the cultivar with the highest ERC %, while 12 represents the cultivar with the lowest ERC %)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 6.4b) Cultivar rank correlation coefficients for estimated recoverable crystal content (ERC %) at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	PG R	AK P	AK R
PG P	1			
PG R	0.84**	1		
AK P	0.76*	0.84**	1	
AK R	0.55	0.34	0.69*	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, *; Significant at P<0.01 and P<0.05, respectively.

6.3.1.3 Cane yield

The highest mean cane yield in the plant crop was obtained at BH, followed by PG and AK respectively (Table 6.5a). The higher cane yield at PG may have been due to the ample water supply compared to the dryland crop at AK-where there was moisture stress due to unreliable rainfall. The cane yield ranges for AK, PG, and BH were 47.04 to 64.64, 119.90 to 151.30, and 143.70 to 203.90 ton ha^{-1} respectively. Cultivar NCo376 was ranked high at BH and PG, while all cultivars had similar cane yields. The cane yields of all cultivars at AK were similar (Table 6.5a). A significant and moderate rank correlation was found between AK plant crop and PG plant crop for cane yield (r=0.60), indicating similar rankings of some cultivars (Table 6.5b).

In the ratoon crop, mean cane yield in PG was significantly higher than that at AK (Table 6.5a). The cane yield ranged from 69.34 to 102.33 ton ha^{-1} in PG and 45.21 to 62.44 ton ha^{-1} at AK. In the ratoon crop, cultivars N25, NCo376, and N31 had high cane yields at PG, all cultivars had similar yields at AK. The rank correlation coefficient between AK and PG for the ratoon crop was 0.44, indicating a weak relationship between the cultivars rankings (Table 6.5b). This shows that cane yield may not be stable across sites in the ratoon.

The low and non-significant (r=0.47) correlation value at AK meant that a single trait value per cultivar may not be adequate to represent cane yield across ratoons. Cultivars N12, N19, N25, N35, N36, N40, N48, N51 and NCo376 had similar rankings across ratoons at PG (Table 6.5b). This is enhanced by the moderate and significant (r=0.62) correlation across ratoons, indicating a moderately strong relationship between cultivar ranking across ratoons at PG (Table 6.5b). This shows that cane yield rankings were generally stable across ratoons at PG, but not at AK.

Once again, this might be related to the stabilising effect of irrigation at PG, compared with the strong seasonal influences of rainfed conditions at AK.

Table 6.5a) Cane yield of 12 cultivars at Amatikulu (AK), Pongola (PG) and Bruynshill (BH) for the plant and ratoon crop

	Plant crop			First ratoon	
	BH	**PG**	**AK**	**PG**	**AK**
N12	169.80$^{abc\ (8)}$	119.90$^{a\ (12)}$	53.37$^{a\ (9)}$	69.34$^{a\ (12)}$	47.33$^{a\ (11)}$
N19	192.30$^{cd\ (2)}$	133.80$^{abcde\ (7)}$	59.06$^{a\ (5)}$	85.31$^{abc\ (8)}$	50.32$^{a\ (10)}$
N25	167.20$^{abc\ (9)}$	150.00$^{de\ (2)}$	59.84$^{a\ (4)}$	102.33$^{c\ (1)}$	52.67$^{a\ (7)}$
N31	182.00$^{bcd\ (6)}$	132.50$^{abcd\ (9)}$	51.18$^{a\ (11)}$	93.90$^{bc\ (3)}$	60.14$^{a\ (2)}$
N35	164.10$^{abc\ (10)}$	141.00$^{bcde\ (5)}$	58.72$^{a\ (6)}$	90.25$^{abc\ (6)}$	51.79$^{a\ (9)}$
N36	173.80$^{bc\ (7)}$	133.50$^{abcde\ (8)}$	60.01$^{a\ (3)}$	83.04$^{abc\ (10)}$	57.13$^{a\ (3)}$
N40	162.60$^{ab\ (11)}$	125.50$^{ab\ (11)}$	47.04$^{a\ (12)}$	77.64$^{ab\ (11)}$	45.21$^{a\ (12)}$
N41	143.70$^{a\ (12)}$	131.40$^{abc\ (10)}$	54.38$^{a\ (8)}$	93.15$^{bc\ (4)}$	55.97$^{a\ (5)}$
N48	187.60$^{bcd\ (3)}$	139.90$^{bcde\ (6)}$	53.36$^{a\ (10)}$	86.61$^{abc\ (7)}$	51.97$^{a\ (8)}$
N51	183.60$^{bcd\ (5)}$	148.10$^{cde\ (3)}$	61.60$^{a\ (2)}$	90.53$^{abc\ (5)}$	62.44$^{a\ (1)}$
N52	187.00$^{bcd\ (4)}$	142.80$^{bcde\ (4)}$	64.64$^{a\ (1)}$	83.65$^{abc\ (9)}$	57.08$^{a\ (4)}$
NCo376	203.90$^{d\ (1)}$	151.30$^{e\ (1)}$	55.56$^{a\ (7)}$	100.74$^{c\ (2)}$	52.91$^{a\ (6)}$
Mean	**176.47**	**137.48**	**56.56**	**88.04**	**53.75**
§	**c**	**b**	**a**		
§§		**c**	**a**	**b**	**a**

Means followed by the same letters are not significantly different within a site and crop cycle. Numbers in brackets indicate ranking of cultivars (1 represents the cultivar with the highest cane yield, while 12 represents the cultivar with the lowest cane yield)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 6.5 b) Cultivar rank correlation coefficients for cane yield at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.62*	1		
AK P	0.60*	0.08	1	
AK R	0.36	0.44	0.47	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, *; Significant at P<0.01 and P<0.05, respectively

6.3.2 Biomass yield and components

Table 6.6 shows the mean square values from the combined ANOVA of the PG and AK trials for both the plant and first ratoon crops for total biomass, as well as green and brown trash. The stalk component of biomass is represented by cane yield, discussed above. The main effects of cultivar (C), site (S), and crop cycle (R) were significant ($P<0.05$) to highly significant ($P<0.01$) for all traits, except for the non-significant effect of C on green trash (Table 6.6). The cultivar x site (C x S) interaction was significant for both the green and brown trash, indicating the varying response of cultivars to site occurred for these two traits. The C x R and the three way (C x S x R) interaction were not significant for any of the traits. This shows that the relative cultivar values for these traits did not vary from one site to the next, and from the plant to the ratoon crop. The mean square value for S was higher than the other mean square values for these traits. The main effect of S therefore contributed most to total variance for the traits. The mean square for the interaction effects of C x S was higher than that of the C x S x R and C x R for all three traits.

Table 6.6 Mean square values from analysis of variance, combined across crop cycles (plant and first ratoon crops) and sites (Pongola and Amatikulu) for total biomass, green trash and brown trash

Source	d.f.	Total biomass	Green trash	Brown trash
		ton ha^{-1}	ton ha^{-1}	ton ha^{-1}
Cultivar (C)	11	1866.90*	87.85	31.57**
Site (S)	1	323498.50**	12976.16**	4731.88**
Ratoon (R)	1	6045.50**	409.41**	677.47**
C x S	11	922.60	84.87*	22.85*
C x R	11	321.70	31.60	3.74
R x S	1	830.50	0.78	507.66**
C x S x R	11	193.90	22.16	9.54

**, *; Significant at $P<0.01$ and $P<0.05$, respectively.

Table 6.7 shows the mean square values from ANOVA of the PG, AK and BH plant crop only. The main effect of C was significant ($P<0.05$) for all traits (Table 6.7), while the effect of S was highly significant ($P<0.01$) for all traits. The C x S interaction was significant for green and brown trash ($P<0.05$) but not significant for total biomass. The relative differences among cultivars for green and brown trash therefore differed by site in the plant crop. The mean square of each main effect (C and S) exceeded the C x S interaction mean square; indicating that the main effect of C and S accounted for more variation than the C x S interaction.

Table 6.7 Mean square values from analysis of variance in the plant crop, combined across sites (Bruynshill, Pongola and Amatikulu) for total biomass, green trash and brown trash

Source	d.f.	Total biomass	Green trash	Brown trash
Cultivar (C)	11	1892.80*	72.11*	13.66*
Site (S)	2	168249.10**	3425.03**	662.00**
C x S	22	1371.10	62.34*	10.79*

**, *; Significant at P<0.01 and P<0.05, respectively.

6.3.2.1 Total biomass

The highest mean total biomass for the plant crop was produced at BH, followed by PG and AK (Table 6.8a). AK produced the lowest amount of biomass because of the rainfed conditions. The total biomass ranges for BH, AK, and PG were 133.10 to 243.80, 51.41 to 90.74, and 128.80 to 183 ton ha^{-1}, respectively. In the plant crop, most cultivars did not differ significantly in total biomass at PG, BH, and AK. Although AK and PG had a low and non-significant correlation (r=0.47) in the plant crop (Table 6.8b), the relative rankings of some cultivars (i.e. N12, N19, N31, N40, N41 and N51) were consistent across AK and PG for the plant crop. The poor rank correlation shows that total biomass may not be stable across sites.

In the ratoon crop PG produced a higher total biomass than that produced at AK (Table 6.8a). The total biomass in the ratoon crop varied from 112.90 to 172.20 ton ha^{-1} at PG and 48.06 to 70.19 ton ha^{-1} at AK. Similar to the plant crop, nine cultivars did not differ in total biomass in the ratoon crop at PG; while all 12 cultivars had similar biomass yields at AK. A low and non-significant correlation value (r=0.28) was observed across sites (Table 6.8b), showing that site had a large effect on cultivar rankings for total biomass in the ratoon crops. This shows that total biomass may not be stable in the ratoon crop as well.

Cultivars ranked similarly across crop cycles at PG only (Table 6.8a), with the exception of cultivars N31 and N48. A strong (r=0.86) and highly significant correlation was found at PG across crop cycles (Table 6.8b). In contrast, the correlation coefficient was low and non-significant between crop cycles at AK, indicating a weak relationship between the cultivar rankings. This meant that cultivar characterization for total biomass in ratoons cannot be adequately represented from information gathered from a single crop only under rainfed

conditions. However, under irrigated conditions, there may be opportunity to characterise cultivars from experiments conducted in a single crop cycle.

Table 6.8a) Mean total above ground biomass (ton ha^{-1}) of 12 cultivars at Bruynshill (BH), Pongola (PG), and Amatikulu (AK) for the plant and ratoon crop

	Plant crop			First ratoon	
	BH	**PG**	**AK**	**PG**	**AK**
N12	194.00$^{abc\ (4)}$	128.90$^{a\ (11)}$	51.41$^{a\ (12)}$	112.90$^{a\ (12)}$	60.56$^{a\ (7)}$
N19	180.70$^{ab\ (7)}$	158.20$^{ab\ (6)}$	75.60$^{ab\ (4)}$	138.90$^{abc\ (7)}$	60.43$^{a\ (8)}$
N25	181.90$^{ab\ (6)}$	183.00$^{b\ (1)}$	75.54$^{ab\ (5)}$	172.20$^{c\ (1)}$	69.18$^{a\ (3)}$
N31	161.70$^{ab\ (10)}$	128.80$^{a\ (12)}$	51.90$^{a\ (11)}$	121.00$^{ab\ (9)}$	69.83$^{a\ (2)}$
N35	133.10$^{a\ (12)}$	168.70$^{ab\ (2)}$	73.33$^{ab\ (6)}$	162.70$^{bc\ (3)}$	58.19$^{a\ (9)}$
N36	174.40$^{ab\ (8)}$	152.70$^{ab\ (8)}$	76.18$^{ab\ (3)}$	140.10$^{abc\ (6)}$	61.78$^{a\ (6)}$
N40	170.00$^{ab\ (9)}$	149.80$^{ab\ (10)}$	56.45$^{a\ (10)}$	117.60$^{ab\ (11)}$	48.06$^{a\ (12)}$
N41	152.10$^{ab\ (11)}$	157.60$^{ab\ (7)}$	65.75$^{ab\ (8)}$	140.70$^{abc\ (5)}$	65.44$^{a\ (5)}$
N48	243.80$^{c\ (1)}$	159.80$^{ab\ (4)}$	65.80$^{ab\ (7)}$	129.90$^{abc\ (8)}$	56.18$^{a\ (10)}$
N51	188.00$^{abc\ (5)}$	162.20$^{ab\ (3)}$	76.71$^{ab\ (2)}$	164.10$^{bc\ (2)}$	70.19$^{a\ (1)}$
N52	205.40$^{bc\ (2)}$	151.70$^{ab\ (9)}$	90.74$^{b\ (1)}$	119.20$^{ab\ (10)}$	68.85$^{a\ (4)}$
NCo376	200.10$^{bc\ (3)}$	158.30$^{ab\ (5)}$	65.27$^{ab\ (9)}$	155.90$^{abc\ (4)}$	51.64$^{a\ (11)}$
Mean	**182.10**	**154.98**	**68.72**	**139.60**	**61.69**
§	**c**	**b**	**a**		
§§		**c**	**a**	**b**	**a**

Means followed by the same letters are not significantly different within a site and crop cycle. Numbers in brackets indicate ranking of cultivars (1 representing the cultivar with the highest total biomass, while 12 represents the cultivar with the lowest total biomass)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 6.8b) Cultivar rank correlation coefficients for total biomass at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.86**	1		
AK P	0.46	0.43	1	
AK R	0	0.28	0.37	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**, *; Significant at P<0.01 and P<0.05, respectively

6.3.2.2 Green trash

The green trash mass of the plant crop was highest at PG, followed by BH and AK respectively (Table 6.9a). The higher mean for green trash at PG may be attributed to the irrigated conditions and warmer environment, which sustained green trash growth until harvest. The ranges of green trash produced were from 13.60 to 26.68 ton ha^{-1} at BH, 10.32 to 17.98 ton ha^{-1} at AK, and 22.83 to 43.62 ton ha^{-1} at PG. The cultivar x site interaction was significant in the plant crop showing that relative differences among cultivars for green trash biomass varied by site in the plant crop. This finding was further supported by the non-significant and low (r = 0.04) cultivar rank correlation between AK and PG in the plant crop (Table 6.9b)

Similar to the plant crop, PG had a higher mean for green trash in the ratoon crop compared to that at AK. There was no separation among cultivar means at AK and little separation at the other two sites. AK had a low (r=0.12) and non-significant correlation with PG for the ratoon crop (Table 6.9b); indicating a weak relationship between the cultivar rankings. This suggests that green trash is not stable across sites in the ratoon crop as well. In the ratoon crop the mean for green trash varied from 15.36 to 7.29 ton ha^{-1} at AK and 36.82 to 22.38 ton ha^{-1} at PG. The mean for green trash was lower than that of the plant crop for both AK and PG.

Within a site, green trash for cultivars N19, N36, N40, N41, N52, and NCo376 ranked similarly between the plant and ratoon crop at AK (Table 6.9a). Most cultivars ranked similarly in the plant and ratoon crops at PG, with the exception of cultivars N40, N41, N51 and N52. Despite this, low and non-significant correlation coefficients were observed at PG (r=0.26) and AK (r=0.33) across ratoons (Table 6.9b). This means that a single trait value per cultivar may not be adequate to represent green trash across ratoons. This may be due to the response of certain cultivars specifically. For example, cultivars N12 at AK and N40 at PG may have influenced the overall correlation values. Other measures of stability of the traits (aside from basic correlations of the ranks) across sites and ratoons may reveal different trends.

Table 6.9a) Mean green trash (ton ha^{-1}) of 12 cultivars at Amatikulu (AK), Pongola (PG) and Bruynshill (BH) for the plant and ratoon crop

	Plant crop			First ratoon	
	BH	**AK**	**PG**	**AK**	**PG**
N12	26.68$^{c\ (1)}$	11.38$^{a\ (10)}$	28.37$^{ab\ (8)}$	15.36$^{c\ (1)}$	30.85$^{ab\ (3)}$
N19	16.70$^{ab\ (9)}$	15.09$^{a\ (6)}$	29.91$^{ab\ (6)}$	11.17$^{abc\ (8)}$	25.23$^{ab\ (8)}$
N25	21.87$^{abc\ (4)}$	15.23$^{a\ (3)}$	43.62$^{c\ (1)}$	11.75$^{bc\ (6)}$	36.82$^{b\ (1)}$
N31	18.84$^{abc\ (7)}$	10.38$^{a\ (11)}$	22.83$^{a\ (12)}$	11.89$^{bc\ (4)}$	24.90$^{a\ (10)}$
N35	13.60$^{a\ (12)}$	15.10$^{a\ (5)}$	31.40$^{ab\ (5)}$	9.15$^{ab\ (10)}$	27.86$^{ab\ (6)}$
N36	16.09$^{ab\ (10)}$	13.04$^{a\ (8)}$	32.31$^{ab\ (4)}$	11.21$^{abc\ (7)}$	25.40$^{ab\ (7)}$
N40	20.17$^{abc\ (6)}$	10.32$^{a\ (12)}$	35.15$^{bc\ (2)}$	7.29$^{a\ (12)}$	23.55$^{a\ (11)}$
N41	15.41$^{ab\ (11)}$	15.13$^{a\ (4)}$	25.24$^{ab\ (11)}$	11.97$^{bc\ (3)}$	28.19$^{ab\ (5)}$
N48	23.64$^{bc\ (2)}$	13.72$^{a\ (7)}$	27.67$^{ab\ (10)}$	8.81$^{ab\ (11)}$	24.95$^{a\ (9)}$
N51	22.28$^{abc\ (3)}$	16.08$^{a\ (2)}$	27.97$^{ab\ (9)}$	11.81$^{bc\ (5)}$	30.44$^{ab\ (4)}$
N52	17.07$^{ab\ (8)}$	17.98$^{a\ (1)}$	29.81$^{ab\ (7)}$	15.36$^{c\ (1)}$	22.38$^{a\ (12)}$
NCo376	21.82$^{abc\ (5)}$	13.04$^{a\ (8)}$	34.68$^{bc\ (3)}$	9.95$^{ab\ (9)}$	31.82$^{ab\ (2)}$
Mean	**19.51**	**13.87**	**30.75**	**11.31**	**27.70**
§	**b**	**a**	**c**		
§§		**b**	**d**	**a**	**c**

Means followed by the same letters are not significantly different within a site and ratoon. Numbers in brackets indicate ranking of cultivars (1 representing the cultivar with the highest green trash, while 12 represents the cultivar with the lowest green trash)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 6.9b) Cultivar rank correlation coefficients for green trash at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**
PG R	0.26		
AK P	-0.04	0.20	
AK R	-0.45	0.12	0.33

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**P<0.01, * P<0.05

6.3.2.3 Brown trash

The brown trash yield from the plant crop at AK was lower than that at BH and PG (Table 6.10a). The cultivars N48, N35, N51 and N19 had the highest yield of brown trash at BH. Cultivars had similar rankings at AK, while cultivars N35, N25, and N31 ranked top at PG. The brown trash yield varied from 7.43 to 15.94 ton ha^{-1} at BH, 3.18 to 5.75 ton ha^{-1} at AK and 8.54 to 14.30 ton ha^{-1} at PG. The rankings of some cultivars were fairly consistent across all

three sites, e.g. N35, N36 and NCo376. However, the low and non-significant cultivar rank correlation (r=0.44) for AK and PG plant crop for brown trash indicates a weak relationship between the cultivar rankings (Table 6.10b).

PG had a higher brown trash yield in the ratoon crop than AK, similar to the response observed in the plant crop. Cultivar N35 produced the highest brown trash yield for PG. Cultivars NCo376 and N25 had the highest brown trash yield at AK for the ratoon crop. The brown trash yield varied from 3.39 to 8.22 and 13.18 to 26.71 ton ha^{-1} at AK and PG respectively. The rank correlation between means at PG and AK in the ratoon was low and non-significant (r=-0.13) showing that there may be a C x S interaction for dead leaf material at harvest.

At PG cultivars N12, N19, N25, N35, N40, N51, N52 and NCo376 had similar ranking across ratoons at PG. Generally, the brown trash rankings of the cultivars were similar across ratoons at PG, meaning that cultivar trait values for brown trash in ratoons can adequately be represented from information gathered in the plant crop. There was a significant and moderate correlation (r=0.60) between ratoons at PG for cultivar ranking, while a low (r=0.24) and non-significant cultivar correlation was observed at AK (Table 6.10a). The weak relationship between rankings across ratoons at AK may be due to a greater sensitivity to seasonal variation under rainfed conditions, whereby a drier season could lead to higher leaf senescence.

Table 6.10a) Brown trash (ton ha^{-1}) of 12 cultivars at Bruynshill (BH), Pongola (PG), and Amatikulu (AK) for the plant and ratoon crop

	Plant crop			First ratoon	
	BH	**AK**	**PG**	**AK**	**PG**
N12	11.07$^{a\ (5)}$	3.55$^{a\ (10)}$	8.54$^{a\ (12)}$	5.02$^{ab\ (3)}$	13.18$^{a\ (12)}$
N19	11.28$^{ab\ (4)}$	4.54$^{a\ (4)}$	11.11$^{abcd\ (7)}$	4.41$^{ab\ (7)}$	18.47$^{a\ (5)}$
N25	7.43$^{a\ (12)}$	5.12$^{a\ (3)}$	13.39$^{de\ (2)}$	6.87$^{bc\ (2)}$	19.41$^{a\ (4)}$
N31	8.48$^{a\ (10)}$	3.56$^{a\ (9)}$	11.99$^{cde\ (3)}$	4.03$^{a\ (9)}$	15.71$^{a\ (11)}$
N35	12.07$^{ab\ (2)}$	5.24$^{a\ (2)}$	14.30$^{e\ (1)}$	4.16$^{ab\ (8)}$	26.71$^{b\ (1)}$
N36	9.57$^{a\ (8)}$	3.72$^{a\ (8)}$	11.23$^{abcd\ (6)}$	3.39$^{a\ (12)}$	19.42$^{a\ (3)}$
N40	9.55$^{a\ (9)}$	3.18$^{a\ (12)}$	10.00$^{abc\ (9)}$	4.66$^{ab\ (6)}$	16.16$^{a\ (9)}$
N41	8.15$^{a\ (11)}$	4.31$^{a\ (5)}$	9.09$^{ab\ (10)}$	3.80$^{a\ (10)}$	17.57$^{a\ (6)}$
N48	15.94$^{b\ (1)}$	3.23$^{a\ (11)}$	11.38$^{ab\ (5)}$	3.41$^{a\ (11)}$	16.25$^{a\ (8)}$
N51	11.60$^{ab\ (3)}$	5.75$^{a\ (1)}$	11.46$^{bcd\ (4)}$	4.90$^{ab\ (4)}$	19.55$^{a\ (2)}$
N52	9.96$^{a\ (7)}$	4.21$^{a\ (6)}$	9.04$^{ab\ (11)}$	4.81$^{ab\ (5)}$	16.07$^{a\ (10)}$
NCo376	10.46$^{a\ (6)}$	4.03$^{a\ (7)}$	10.29$^{abc\ (8)}$	8.22$^{c\ (1)}$	17.43$^{a\ (7)}$
Mean	**10.46**	**4.20**	**10.99**	**4.81**	**17.99**
§	**b**	**a**	**b**		
§§		**a**	**b**	**a**	**c**

Means followed by the same letters are not significantly different within a site and crop cycle. Numbers in brackets indicate ranking of cultivars (1 representing the cultivar with the highest brown trash, while 12 represents the cultivar with the lowest brown trash)

§ mean separations across sites

§§ mean separations across sites and crop cycle

Table 6.10b) Cultivar rank correlation coefficients for brown trash at Amatikulu (AK) and Pongola (PG) across ratoons and site

	PG P	**PG R**	**AK P**	**AK R**
PG P	1			
PG R	0.60*	1		
AK P	0.44	0.75*	1	
AK R	-0.16	-0.13	0.24	1

AK P- Amatikulu plant crop, **AK R** – Amatikulu ratoon crop, **PG P** – Pongola plant crop, **PG R** – Pongola ratoon crop

**P<0.01, * P<0.05

6.3.3 Heritability

Broad sense heritability and variance components for all traits are shown in Table 6.11. The genetic variance (C) was the highest for total biomass. This shows that variation in this trait is mostly cultivar related, while the environmental variance (E) was the highest for cane yield, ERC%, ERC yield, green trash, and brown trash. In contrast, Chaudhary (2001) reported that

genetic variance was higher than the environmental variance for cane yield, millable cane, single cane weight, stalk diameter, and stalk length. The C and E values were similar for ERC% and total biomass. The C x S interaction variation was the highest of all cultivar interaction variance components for each trait.

Cane yield (0.72), ERC% (0.74) and total biomass (0.93) had high heritability values. Similarly high heritability estimates results were recorded by Tadesse et al. (2014) for sugar yield (86.09%), cane yield (75.02%), and millable cane (81.51%). Chaudhary (2001) reported a high heritability estimate for cane weight (84%), Nair et al. (1980) and Singh et al. (1994) reported similar results for millable cane. The high estimate of heritability for these traits is due to the high C values. This indicates that simple selection for these traits would be effective for sugarcane cultivar breeding programmes. While this is well-known for the conventional yield and yield components, information on heritability of biomass components is limited. Such information may be valuable to breeding programmes targeted to breeding for cogeneration and biomass.

Table 6.11 Variance components and broad sense heritabilities of cane yield, ERC yield, ERC%, total biomass, green trash, and brown trash

Source	Cane yield	ERC yield	ERC%	Total biomass	Green trash	Brown trash
C	16.14	0.16	1.03	816.50	0	0.91
C x R	8.44	0.25	0	15.98	1.18	0
C x S	25.66	1.67	0.57	91.09	7.84	1.66
C x S x R	0	0	0.24	0	0	0.38
E	183.70	4.21	1.12	701.70	35.55	8.00
H^2	**0.72**	**0.12**	**0.74**	**0.93**	**0**	**0.46**

6.4 Conclusions

The cultivar x site (C x S) interaction was highly significant for ERC% and ERC yield. The results showed that some cultivars had similar rankings across sites for ERC%, showing that this trait is relatively stable, particularly in the plant crop. However, weak correlation coefficient values were found between cultivar rankings across sites for the ratoon crop for ERC%. Within a site, ERC% was also stable across ratoons, showing that cultivars could be characterised by single ratoon experiments in future studies. Some cultivars such as N40 consistently showed high ERC% across sites and ratoons suggesting that it may be a good indicator cultivar for future studies. Similarly, other cultivars like N52 showed consistently low ERC%. The low cultivar correlations between AK and PG for the plant and ratoon crop for cane yield, ERC yield, green trash yield, brown trash and total biomass meant that these traits were less stable than ERC%.

The ERC yield generally showed poor rank correlations across sites in the plant and ratoon crops, which may have been caused by seasonal differences at AK and PG. At PG, the rank correlation between the plant and the ratoon crop was moderate, but weak at AK. This meant that cultivar characterisation from experiments with only one crop cycle is a possibility under irrigated conditions. The poor rank correlations across sites in the plant and ratoon crops for total biomass suggest that cultivars may not be characterised well for this trait by single-site experiments. A strong rank correlation was found at PG across crop cycles, but the rank correlation coefficient was weak at AK, indicating a weak relationship between the cultivar rankings. This meant that cultivar characterisation for total biomass in ratoons cannot be adequately represented from information gathered from one crop only under rainfed conditions

Cultivar rankings for green trash showed poor rank correlations between sites and crop cycles within sites. Cultivars, therefore, cannot be characterised for this trait using single-site and single-ratoon experiments in future, hence its use as a trait for model-assisted breeding will be limited. The poor rank correlations for brown trash across sites in the plant and ratoon crop indicate that it was not stable across sites.

Total biomass had a high genetic variance estimate. Heritability estimates for six traits ranged from 0 to 0.93. High heritabilities were recorded for cane yield, ERC%, and total biomass suggesting that these traits can effectively be selected for in breeding programmes and can be

used in crop models. Follow-up studies should involve the statistical grouping of cultivars into categories for each trait, and the determination of mean values for each category. These mean values should then be incorporated and validated using the Canegro crop model.

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CHAPTER 7

7.1 General discussion and conclusions

Crop models such as Canegro are able to identify traits that contribute to increases in potential yield; thus have the potential to assist with breeding and cultivar selection (Zhou et al., 2003). Currently the cultivar traits in the Canegro model are based exclusively on experimental work done on the cultivar NCo376 and estimates for a wider range of cultivars are not available for key growth parameters. The primary objective of this study was to quantify the cultivar coefficient values for some lower-level cultivar traits and cane and sucrose yield, and some of the final harvestable biomass components for a diverse range of sugarcane cultivars. Additionally the G x E interactions were investigated, and stability and heritability of these traits were determined to assess their potential use for model-assisted breeding. It was envisaged that the cultivar coefficient values determined here could help refine the crop model's ability to simulate cultivar growth differences across environments.

Data from this study was used to analyse and identify various cultivar traits that are stable across sites and crops. SER was found to be one of the most stable cultivar traits that could be used for model-assisted breeding. The SER showed strong rank correlations between sites and within sites across ratoons. Cultivars NCo376 (values ranging from 0.33-0.67) and N12 (values ranging from 0.32-0.55) were identified as ideal indicator cultivars for low SER, while cultivars N31 (values ranging from 0.41-0.92) and N52 (values ranging from 0.47-0.89) were identified as high SER cultivars. Some of these values were similar to the maximum stalk elongation rate of up to 48 mm d^{-1} recorded for Q127, 44 mm d^{-1} for Q96, and 38 mm d^{-1} for Ql17 by Shannon and Holden (1996).

With regard to the other tiller and stalk traits, it was found that the cultivar x site (C x S) interaction was highly significant for PTP and SER and significant for TTPP and TSP. This indicated the varying relative responses of cultivars to site for these traits. This study represents one of the first indications of significant C x S interactions for these lower-level traits, which could be possible drivers of G x E interactions in sugarcane. Additionally, the three way C x S x R interaction was significant for PTP and FPOP, showing that the relative cultivar values for these traits varied from one site to the next, and from the plant to the ratoon crop. Once again, these findings could help to explain the nature of G x E interactions in sugarcane. Other studies have focused on G x E interactions of individual yield components only, such as stalk height,

stalk weight and cane yield (Jamoza et al., 2014). Despite the significant C x S interaction PTP was relatively stable across sites for the plant and ratoon crop. For example cultivars N36 and N48 had consistent rankings across sites in the plant and ratoon crop. In general, values for PTP ranged from 238 839 to 153 571 stalks ha^{-1} at PG and 335 417 to 177 083 stalks ha^{-1} for AK for the plant crop and 289 068 to 153 646 stalks ha^{-1} in AK and 441 964 to 247 321 stalks ha^{-1} in PG for the ratoon crop. Donaldson et al. (2011) reported lower maximum tiller population values of 111 964 ha^{-1} and 55 000 ha^{-1} for the December ratoon and 68 214 ha^{-1} and 111 964 ha^{-1} in the May ratoon for NCo376 and N26 respectively. While Zhou (2003) reported peak tiller population among the cultivars ZN6, ZN7, N14 and NCo376 to range from 150 700 to 265 300 ha^{-1}.

The weak rank correlations between sites and ratoons within sites for TTPP suggest that this trait is more influenced by environmental factors. Its use as a trait for model-assisted breeding will therefore be limited. The trait FPOP ranged from 67708 to 162946 stalks ha^{-1} and from 73958 to 166964 stalks ha^{-1} in the plant and ratoon crop respectively. Within sites across ratoons FPOP showed consistent cultivar rankings. This suggests that information on FPOP measured in the plant crop can adequately represent relative values obtained in the ratoon and that cultivars may be characterised for this trait by single-ratoon experiments in future. Cultivars such as N12 and NCo376 (high FPOP), and N36 (low FPOP) were identified as ideal indicator cultivars in future studies. Rafiq and Sattar (2013) reported similar range values of 44 350 to 147 650 stalks ha^{-1} for millable canes of different genotypes.

Environmental factors had a major role on the tiller population dynamics. The SER however, was more affected by G as evidenced by the larger value of genetic variance. The traits SER, PTP and FPOP had a high heritability (ranging from 0.70 to 0.97) while TTPP and TSP had moderate heritability estimates; meaning that the cultivar rankings for these traits were less consistent from ratoon to ratoon and from site to site. Heritability estimates for conventional yield and yield components have been reported. For example, high heritability estimates were reported by Jamoza et al. (2014) for stalk diameter, number of millable canes, single stalk weight and number of internodes. This study, on the other hand, represents the first report of heritability estimates for lower-level traits used in crop modelling. This information will help guide future decisions and experiments linking crop modelling with sugarcane improvement efforts. For example, only traits showing high heritability should ideally be used for model-wise exploration of factors influencing G x E interactions.

With regard to the leaf phenology traits, the LAmax showed strong rank correlations across sites and ratoons. This means that cultivar characterisation for LAmax may be possible with information from single-site and single ratoon experiments in future. The C x S interaction was significant for TTLAmax and the poor rank correlations in the plant and ratoon crop at AK and PG indicated that this trait is not stable across sites. The moderately strong rank correlation for LAI meant that this trait is fairly stable trait across sites in the plant and ratoon crop. This meant that cultivar characterisation may be represented adequately with information from one site only. For LAI some cultivars such as N12 and NCo376 showed consistent rankings across ratoons at AK, suggesting that they can be used as indicator cultivars for LAI in future studies.

Of all leaf phenology traits, LAR was the most stable, showing strong rank correlation between sites and across ratoons. All cultivars had similar rankings within sites across crops. Cultivar characterization for LAR can thus be adequately represented from information gathered from single-site and single-ratoon experiments in future. A cultivar such as N35 consistently showed high LAR, suggesting that it will be a good indicator cultivar in future experiments. The LFmax showed consistent rankings across sites in the plant crop only suggesting that this trait is fairly stable across sites in the plant crop only. The reasons for lower stability of the trait in the ratoon crop are unclear.

Most leaf phenology traits generally had a high heritability (ranging from 0.84 to 0.99). This suggests that leaf development traits could be selected for in sugarcane yield improvement programmes. If these traits are therefore found to be associated with cane and sugar yields, it may be possible to select for them or investigate ideal trait combinations for different environments, i.e. ideotype development. Follow-up studies should involve the statistical grouping of cultivars into categories for each trait, and the determination of mean values for each category. These mean values should then be incorporated and validated using the Canegro crop model.

Regarding the sugar yield components, the ERC% was identified as the most stable trait. Some cultivars such as N40 (high ERC%) and N52 (low ERC%) showed consistent rankings across sites and ratoons suggesting that they may be good indicator cultivars for future studies. Cane yield also showed moderate rank correlations between sites in the plant crop, but poor correlations in the ratoon crop. This may be linked to the harsher season experienced in the ratoon crop, where some cultivars may have struggled under rainfed conditions at AK.

Similarly, the ERC yield generally showed poor rank correlations across sites in the plant and ratoon crop. Total biomass showed poor rank correlations across sites in the plant and ratoon crop suggesting that cultivars may not be characterised for this trait by single-site experiments in future. A strong rank correlation was found at PG across ratoons but weak correlations were found across ratoons at AK. This meant that cultivar characterisation for total biomass in ratoons cannot be adequately represented from information gathered from one crop only under rainfed conditions, while this may be possible under irrigated conditions.

The poor rank correlations for green trash between sites and ratoons within sites, suggest that this trait is strongly influenced by environmental factors. Cultivars can therefore not be characterised for this trait by single-site and single-ratoon experiments in future; hence its use as a trait for model-assisted breeding will be limited. The poor rank correlations for brown trash across sites in the plant and ratoon crop indicate that it was not stable across sites. Total biomass had a high genetic variance estimate. High heritability estimates were recorded for cane yield, ERC% and total biomass. A large proportion of the total variance is heritable for these traits, thus these traits can be easily selected for in breeding programmes (as they currently are) and can be used in crop models.

If there is a significant G x E interaction variance, one or more of the various methods for measuring the stability of genotypes can be used to identify the stable genotype(s). In our study the rank correlation coefficient was used to determine stability. Other methodologies can also be used to assess the stability of genotypes. The most important are based on analysis of variance, linear regression, multivariate analysis and non-parametric statistics (Bastos et al., 2007). The methods of Wricke (1965), Eberhart and Russell (1966), additive main effect and multiplicative interactions (AMMI) (Zobel et al., 1988), and Lin and Binns (1988) are examples of each of these classes respectively. Stability measures based on ranks require no statistical assumptions about the distribution of the data set (Huehn, 1990). They are easy to use and interpret and, compared with parametric measures, are less sensitive to errors of measurement. For selection in breeding the rank orders of the cultivars provide the most essential information; a cultivar is considered stable if its ranking is relatively constant across environments (Flores et al., 1998). The other (parametric) methods rely on statistical assumptions. The results of these methods can therefore be misleading if the statistical assumptions are not satisfied. Given the wide range of stability measures available, future work

should involve quantifying the stability of the traits using different statistical measures for confirmation.

Data from this study can be used to calibrate or validate the Canegro model. This will enhance the capability of the model to predict cultivar effects on yields of sugarcane crops and will lead to further insights into the physiology of sugarcane. The range values for the different traits determined here will be used in exploratory studies whereby the traits are varied hypothetically in the model and the effects on yield observed. The stability and heritability estimates determined in this study will help identify traits worthy of model-assisted breeding in future studies.

Future work needs to focus on measuring the traits defined here in more environments and for more ratoon crops. An attempt could also be made to categorise cultivars into groups with sets of similar traits and range values. Categorisation of cultivars has been attempted by Inman-Bamber (1994). Once grouped according to leaf, tillering, or biomass categories, further physiological and crop modelling work can be done on the group representatives (indicator cultivars) alone, without having to study a full set of cultivars. In this respect, this study has served as an appropriate first step toward integrating crop modelling and plant breeding efforts for sugarcane improvement.

7.2 References

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Appendix 1A

Analysis of variance

Variate: ERC_yield

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Block stratum	3	61.810	20.603	3.83	
Block.*Units* stratum					
Cultivar	11	390.671	35.516	6.60	<.001
crop	1	329.365	329.365	61.23	<.001
Cultivar.crop	11	78.368	7.124	1.32	0.230
Residual	69	371.174	5.379		
Total	95	1231.387			

Message: the following units have large residuals.

Block 2 *units* 2 5.20 s.e. 1.97

Tables of means

Variate: ERC_yield

Grand mean 13.78

Cultivar	N12	N19	N25	N31	N35	N36	N40
	11.09	14.34	15.94	10.44	15.60	14.99	14.57

Cultivar	N41	N48	N51	N52	NCo376
	15.22	14.87	14.03	9.76	14.53

crop	P	R
	15.63	11.93

Cultivar	crop	P	R
N12		13.06	9.12
N19		15.84	12.83
N25		17.52	14.36
N31		10.21	10.67
N35		17.58	13.62
N36		17.86	12.12
N40		16.84	12.30
N41		16.97	13.47
N48		18.17	11.58
N51		16.56	11.50
N52		10.41	9.10
NCo376		16.58	12.48

Standard errors of means

Table	Cultivar	crop	Cultivar crop
rep.	8	48	4
d.f.	69	69	69
e.s.e.	0.820	0.335	1.160

Standard errors of differences of means

Table	Cultivar	crop	Cultivar crop
rep.	8	48	4
d.f.	69	69	69
s.e.d.	1.160	0.473	1.640

Least significant differences of means (5% level)

Table	Cultivar	crop	Cultivar crop
rep.	8	48	4
d.f.	69	69	69
l.s.d.	2.313	0.944	3.272

Stratum standard errors and coefficients of variation

Variate: ERC_yield

Stratum	d.f.	s.e.	cv%
Block	3	0.927	6.7
Block.*Units*	69	2.319	16.8