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WinRHIZO™ software for evaluating effects of farming systems on sugarcane root systems

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Abstract Sugarcane roots are notoriously difficult to study. The opaque nature of the soil matrix, large biomass, and ratooning habit of the crop make studying roots challenging. WinRHIZO™ is a specialist root analysis software that provides rapid and accurate measurement of a host of root characteristics in a fraction of the time that it would take by traditional methods. This paper demonstrates the use of WinRHIZO™ to evaluate root systems under different farming systems management. A paired site was identified in the Herbert district consisting of two commercial sugarcane blocks. One was conventionally managed while the other was under an improved farming system. Importantly, both blocks had been farmed this way for more than 14 years. WinRHIZO™ analysis found root systems in the fields managed under improved farming systems showed significant improvement in total root length, proportion of fine root hairs, root biomass, and many other root properties. This technology has wide ranging applicability as a tool for measuring the impact of farming decisions on root health.

Key words WinRHIZO™, roots, improved farming system, conventional farming system

INTRODUCTION

Sugarcane roots are notoriously difficult to study. The opaque nature of the soil matrix, large biomass, and ratooning habit of the crop make root study challenging. Most root biomass for sugarcane is found close to the surface and then declines approximately exponentially with depth. Typically, 50% of root biomass occurs in the top 20 cm of soil and 85% in the top 60 cm (Figure 1) (Blackburn 1984). Traditional methods of root digging and hand measurement in the laboratory are time consuming and laborious (Reghenzani 1993). WinRHIZO™ is a specialist root-analysis software that provides rapid and accurate measurement of a host of root characteristics in a fraction of the time taken using traditional methods.

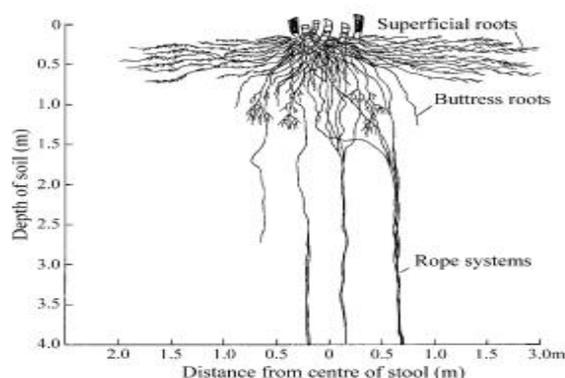


Figure 1. Root system of sugarcane (Blackburn 1984).

Here, we demonstrate the use of WinRHIZO™ and present an evaluation of root systems under different farming systems at a paired site that is part of a comparison of 10 paired sites. The ‘improved farming system’ is promoted as improving sugarcane productivity by improving the health of the soil. Practices such as controlled traffic, legume break cropping, green-cane harvesting and minimum tillage are encouraged as part of this system. These practices are thought to improve root health relative to conventional farming systems. There has been little work in the Australian sugar industry to utilise new technologies, such as WinRHIZO™, to evaluate the impacts of these farming system on sugarcane root systems.

METHODS AND MATERIALS

Design

We identified a paired site in the Herbert district with two commercial sugarcane blocks under different farming systems (Table 1). The sites were adjacent to each other across a headland and both were sandy loam soils. Neither site had any chemical constraints, such as low pH, sodicity or nutrient deficiencies. One was conventionally (CON) managed while the other was under an improved farming system (IFS). Importantly, both blocks had been farmed this way for more than 14 years.

Table 1. Farming systems at the two sites.

Parameter	Farming system	
	Conventional	Improved
Block size (ha)	2.27	2.39
2016 plant date	August	August
2017 harvest date (estimate)	18 August	11 September
Cultivar	Q240 [®]	Q240 [®]
Crop class	Plant	Plant
Row spacing (m)	1.67	1.83
Single or dual row	Single	Single wide mouth
Controlled traffic	No	Yes
Permanent beds	No	Yes
2017 yield (t/ha)	135.31	88.66
2017 CCS	14.08	14.96
Fallow management	Bare	Mixed legume
Number tillage passes (harvest to plant)	6	4
off-set disc	4	2
ripper	1	0
rotary hoe	1	1
zonal tillage	0	0
Renovator	0	1
wavy disk zonal	0	0
other	0	0
Planting method	V-furrow	Mound
Fertiliser placement	Stool split	Stool split
Irrigation	Nil	Nil
Green-cane harvest	Yes	Yes
Last mill mud application	Never	Never
Last lime application	2016 2.5 t/ha	2016 2.5 t/ha
Last gypsum application	Never	Never
Last grub insecticide	Never	Never
Last laser level	Never	Never
Years block farmed this way	30+	14
Summary	Full working multiple passes, conventional planting into furrow, stool split fertiliser	Permanent beds, min till zonal, mound (Mizzi) planting, stool split fertiliser

Sites were both Q240[®] plant cane planted in a single-row configuration. Cane was wide-mouth planted on mounded permanent beds at IFS, compared with a traditional v-shaped furrow at CON. IFS is on a 1.83 m rows,

controlled-traffic system with a multi-species fallow, whereas CON site had a bare fallow, no controlled traffic, and narrower row spacing 1.67 m. The number of tillage passes is significantly lower at IFS. Yield was significantly lower at IFS than at CON in 2017, with the grower suggesting this was due to a phototoxic effect of a new herbicide; yields on this farm are typically well above that.

Sampling was undertaken at both sites after a fallow and then replanting. At this time the sugarcane crop had been established for 28 weeks. Both blocks were planted in the middle of August 2016.

Sugarcane biomass

At each block, we marked out 10 transects, each transect a 5-m section of sugarcane row. Transects were located across the width of the block at points at least 20 m from the headland. At each replicate above-ground biomass was measured by collecting 30 stalks of cane. Fresh weight and stalk height measurements were taken. Five measurements of effective rooting depth were taken at each transect using an analogue penetrometer to determine the depth at which 2000 kPa pressure was reached.

Root collection

At each of the biomass sites an area around the base of 10 stools was cleared of debris, and a coring cylinder (200 mm long by 100 mm diameter) was placed 5 cm away from each stool and driven into the ground using a jackhammer (Figure 2). The 10 cores were then dug out and combined to form one sample. Soil was removed from the cores and placed into a sodium tri-polyphosphate solution (50 g/10 L water) to help the soil disperse from the roots (Figure 2).



Figure 2. Field collection of root samples: Top left - placement of cores; Top right - inserting cores using jackhammer; Bottom - emptying out the cores.

Root washing and processing

The roots were hand washed through a 5 mm then a 2 mm sieve, separating the roots from the trash and all other organic and inorganic matter, and stored in a 50% ethanol to preserve them until scanning and analysis (Figure 3).

Roots were drained, patted dried and laid out onto clear A3 Perspex sheets in a single layer, and covered by a blue backboard. Roots were then scanned with a flat-bed scanner (Microtek ScanMaker 9800XL) at 400 dpi. The blue backboard was used to provide a contrast for root detection (Figure 4). Roots were dried at 60°C for 5 days and wet and dry weights recorded.



Figure 3. Root washing: Left - root washing; Right - washing fine particles out of roots.

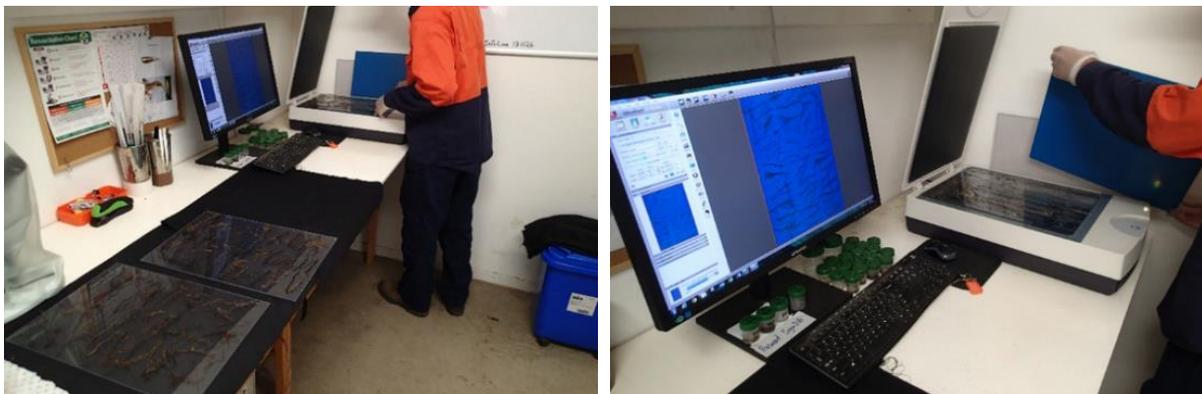


Figure 4. Preparing to scan roots: Left - scanning roots; Right - scanning roots using WinRHIZO™.

Root analysis

Scanned images of the washed root cores were then processed using WinRHIZO™ software (WinRHIZO 2017, Regent Instruments, Canada). A colour calibration chart was made to identify the different colours and sizes that the roots presented (Figure 5). Root classes were set as: primary roots $1 < x < 5$ mm, secondary roots $0.5 < x \leq 1.0$ mm, and tertiary (fine) roots $0 < x < 0.5$ mm.

Measurements were compared between fields by a two-sample t-test in Statistix 10 at a 95% confidence level. Data was first checked for normality by the Shapiro-Wilk test and transformations made where required.



Figure 5. Scanned roots: Left - scanned roots using Microtek ScanMaker 9800XL; Right - after WinRHIZO™ analysis showing the software's measurement of roots by size class (primary – green, secondary – yellow, and tertiary – red).

RESULTS

Root system health was significantly better at IFS than at CON, although both sites were the same cultivar, ratoon and sampled at the same age (Figure 6). Higher stalk populations at IFS were associated with the reduced average stalk weight.

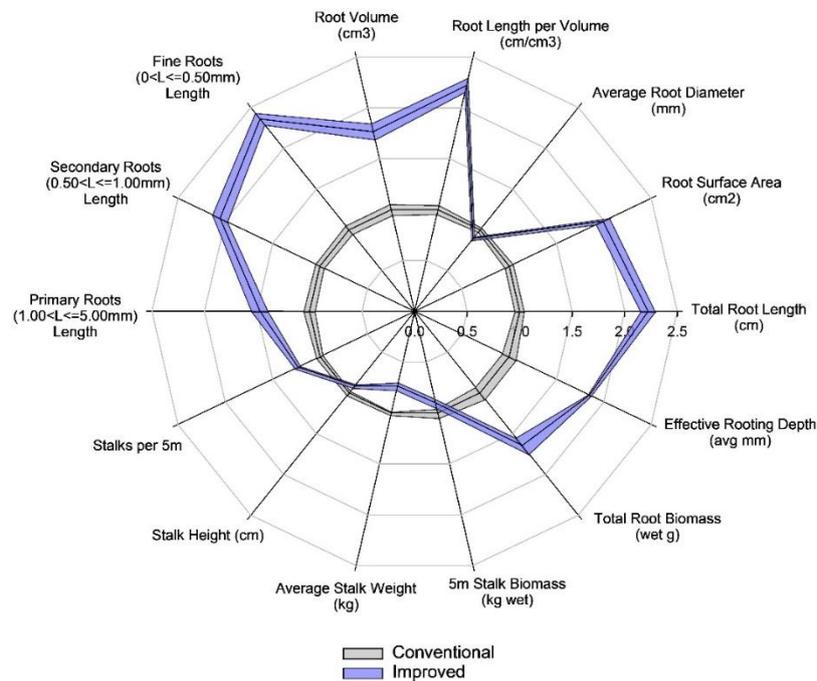


Figure 6. Comparison of 14 root-system and above-ground measurements at CON and IFS. The values for each variable have been normalised to 1 for CON and IFS values are shown as a proportion of CON. Width of the bands indicates 1 standard error.

IFS sugarcane had significantly increased rooting depth, total root length, total root biomass and increases in the lengths of all root diameter classes (Figures 7-10, 12) ($P < 0.05$). There was no significant difference in average root diameter (Figure 11).

DISCUSSION AND CONCLUSIONS

The WinRHIZO™ root analysis software, together with the sampling and processing methodology that we describe, have vastly improved our capacity to study and analyse root systems of sugarcane and to compare the impact of different farming systems on root health. WinRHIZO™ can measure colour differences among roots, which opens up future research possibilities such as measurement of pathogenic lesions.

Through investigating sugarcane root systems, we have been able to get a better understanding of the impacts of farming systems on plant health. Our study showed that improved practices, which promote greater rooting depth and carbon inputs, may result in significant improvements to the root systems during the important crop establishment phase of sugarcane growth. Although more fields need to be studied in order to obtain conclusive evidence of the impact of farming systems on root systems, the WinRHIZO™ system will be a useful tool in that analysis.

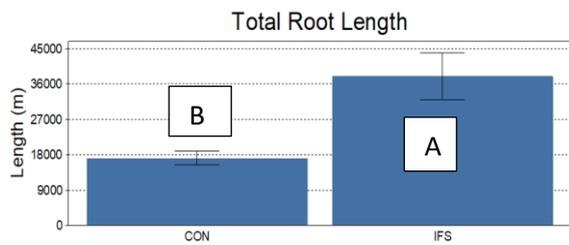


Figure 7. Total root length.

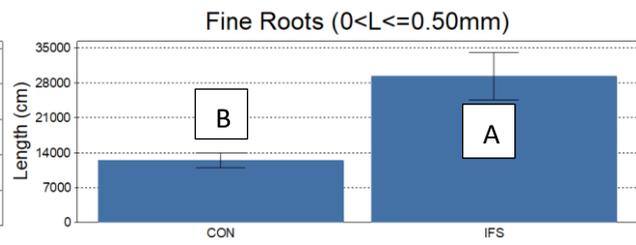


Figure 8. Total length of fine roots.

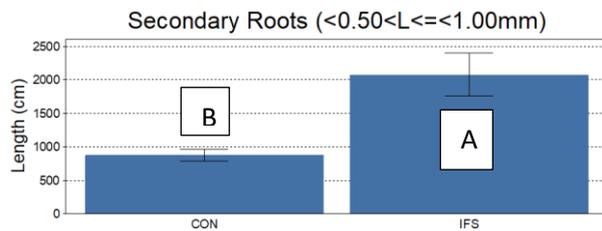


Figure 9. Total length of secondary roots.

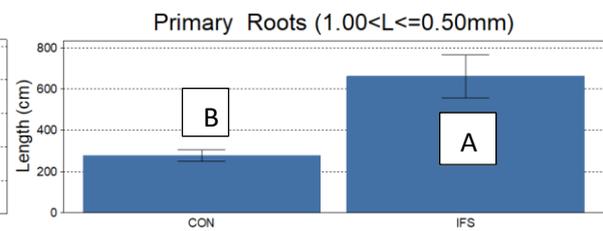


Figure 10. Total length of primary roots.

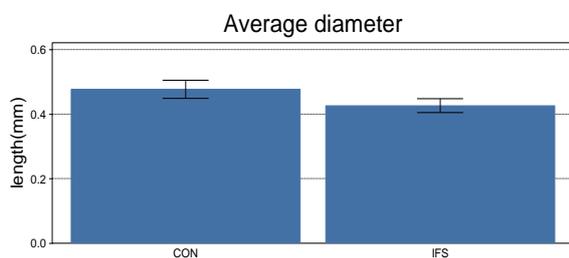


Figure 11. Total average root diameter.

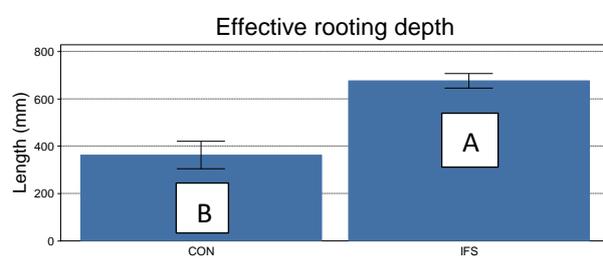


Figure 12. Effective rooting depth. Depth to 2000 kPa soil pressure.

On each of Figures 7-12, A indicates a statistically significant difference (of 95% C.I.) to B.

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