Final report 2014/093: Tissue culture - managing impediments to adoption in Tully

Villaruz, J
Sugar Research Australia Limited

http://hdl.handle.net/11079/16343
Downloaded from Sugar Research Australia Ltd eLibrary
FINAL REPORT 2014/093

Tissue Culture – Managing Impediments to Adoption in Tully

Final report prepared by: Jordan Villaruz
Chief Investigator(s): Jordan Villaruz
Research organisation(s): Tully Cane Productivity Services Ltd (TCPSL)
Date: March 2017
Key Focus Area (KFA): KFA 7: Knowledge and technology transfer and adoption
© Sugar Research Australia Limited 2017

Copyright in this document is owned by Sugar Research Australia Limited (SRA) or by one or more other parties which have provided it to SRA, as indicated in the document. With the exception of any material protected by a trade mark, this document is licensed under a Creative Commons Attribution-NonCommercial 4.0 International licence (as described through this link). Any use of this publication, other than as authorised under this licence or copyright law, is prohibited.

http://creativecommons.org/licenses/by-nc/4.0/legalcode - This link takes you to the relevant licence conditions, including the full legal code.
In referencing this document, please use the citation identified in the document.

Disclaimer:
In this disclaimer a reference to “SRA” means Sugar Research Australia Ltd and its directors, officers, employees, contractors and agents.
This document has been prepared in good faith by the organisation or individual named in the document on the basis of information available to them at the date of publication without any independent verification. Although SRA does its best to present information that is correct and accurate, to the full extent permitted by law SRA makes no warranties, guarantees or representations about the suitability, reliability, currency or accuracy of the information in this document, for any purposes.

The information contained in this document (including tests, inspections and recommendations) is produced for general information only. It is not intended as professional advice on any particular matter. No person should act or fail to act on the basis of any information contained in this document without first conducting independent inquiries and obtaining specific and independent professional advice as appropriate.

To the full extent permitted by law, SRA expressly disclaims all and any liability to any persons in respect of anything done by any such person in reliance (whether in whole or in part) on any information contained in this document, including any loss, damage, cost or expense incurred by any such persons as a result of the use of, or reliance on, any information in this document.
The views expressed in this publication are not necessarily those of SRA.

Any copies made of this document or any part of it must incorporate this disclaimer.

SRA Research Project Final Report

SRA Project Code 2014/093

Project Title Tissue Culture – Managing Impediments to Adoption in Tully

Key Focus Area in SRA Strategic Plan Key Focus Area: 7. Knowledge and technology transfer and adoption.

Research Organization(s) Tully Cane Productivity Services Ltd (TCPRL)

Chief Investigator(s) Jordan Villaruz

- To increase the area in the Tully growing region which is commercially planted with material 2 years or less away from an approved seed source as recommended by SRA.
- To raise the numbers of growers from the current 4 to 25 annually using Tissue culture as a clean seed source.
- To increase the numbers of tissue culture seedlings ordered from Tully up to 10,000 plants each year and to increase the numbers of growers regularly ordering tissue culture.
- To develop the skills and knowledge of Productivity staff to advice on Tissue culture husbandry so they are confident to continue promoting tissue culture.
- To develop a local propagation pathway with the skills and commercial interest to reduce the time between order and delivery as well as reducing unit costs by increasing volume.
Executive Summary

The world is moving and changing every year, it is timely to investigate technologies available outside our current industry practice. When the opportunity arrives we need to take it and explore the possibilities as an industry. Therefore SRA, PEC unit and TCPSL staff developed a collaborative approach between the growers, industry, local tissue culture laboratory, local engineering shop and the TCPSL team to operate a Tissue Culture project called “Managing Impediments to Adoption in Tully”. The new skills learnt from day to day activities and grower demonstrations have been commercially adopted by the industry. This project helped TCPSL staff to improve their knowledge and skills, and allow grower and staff to share their experience and ideas in-order to achieve the goal. SRA and Tissue culture laboratory and nursery are heavily involved to support our needs in these trials. To start, an investigation took place as to the ability of local tissue culture laboratories to manage demand if other laboratories outside the district were unable to. There is a possibility that the Tully growers can access their materials through their local laboratory with SRA and TCPSL direction and control. Mission Beach Tissue Culture Laboratory was one such local business. This lab started to trial separation of plantlets from petri dish, to seedling development stage to nursery and on to the hardening stage.

Planting tissue culture is similar to growing commercial cane, however it reduces the area required to plant new varieties, with less mechanical equipment required because the tissue culture seeds are guaranteed disease resistant and can be planted in high density patterns with trickle irrigation. Land preparation requires fine tilth, to maximise soil – root zone contact and an old style hand planter is used to prepare the row, mark the rows and drop the fertiliser beside before transplant. Tissue culture plants require great attention to soil moisture levels, especially in the three days after transplant to minimise the risk of “transplant shock”. Pre-emergent herbicide is applied 2 weeks after planting and when the seedlings are approximately 60cm high. Some cultivation takes place using a “cotton king” implement to finalise a row profile if required. Harvesting takes place with a commercial harvester adopted for seed cane harvesting.

Tissue culture helps to accelerate new varieties, absolutely clean seeds, and promote a better, healthier crop. Proof in the success of this project is the increasing volume of tissue culture plants local growers are ordering, this has increased in the 3 years of this project. There have been a few failures due to a combination or weather, materials left too long before they were planted in the ground, pest damage, weed problem and a general lack of information. All this is good experience from our point of view to help the industry learn, and develop solutions from this demonstration.

Over all this project has been important in our region, as it involves collaboration between local reseller, industry, growers, laboratory and researchers to find out the impediments of adopting tissue culture locally. Growers are keen to trial, experience and experiment with this new technology, to reduce costs and improve efficiency of seed cane production. In addition TCPSL can increase the volume of good seed cane available to growers. We believe as a Productivity Board we can manage and grow tissue culture in our region using the best information gathered from our demonstration.

We strongly recommend the following steps before deciding to plant tissue culture:

- Soil test before planting if ameliorants are required
- Order tissue culture plants early and if possible plant preferably in April. Less irrigation is needed.
• Or the next option is to plant July and August.
• Purchase and prepare all the materials needed before planting
  o Liquid fertilizer or growth enhancer and tub for soaking tissue culture plant before planting. Trickle type, lay flat, connectors and timer for watering system. Irrigation source (creek, town water, gravity feed water tank and/or bore water), tissue culture planter, long stick planter to fertilise the row. Chemicals for pre- emergence.
• Good land preparation (fine tilt) before planting
• Good weed management strategy in place
• Scratcher or cotton king for final hill up
• Monitor the plants every couple of days especially if newly planted and assess the situation until the crop is established and ready to use for planting or harvest

Tissue culture is good way to increase the cane populations of clean seed and mass propagate new varieties. This project is a good example of SRA funding and supporting local industry initiatives. This helps to improve the knowledge and skills of Productivity Board staff, builds a good collaboration to research and local industry, and creates beneficial networks with local agribusiness. We believe that the local Productivity Board are well engaged with local growers to allow any future funding of projects similar this to be beneficial.

**Background**

After cyclone Yasi, 2011 the majority of our new and major cane varieties were damaged, between 80 – 95% of all the Tully region. Some growers visited other regions to pick up clean seed cane and major varieties. TCPSL seed plots were severely damaged by the cyclone and it took one to two years to propagate and bring the plot back into its normal growing cycle and full production.

Another issue we encountered was when smut arrived in the sugar industry, 2006. We gathered a small group of growers to trial or plant any resistant or intermediate varieties from other regions like Burdekin, Ingham, Mossman, and Tableland using Q208A, KQ228A and Q199A and other resistant varieties. This required intensive labour and increased the cost of the propagation plot. Varieties Q208A and KQ228A then become our major varieties in Tully and are now the dominant cane in our region. The Tully region also offered some support to the Herbert and Burdekin regions who supplied large quantities of whole stalk and billet cane of Q200 and Q208 to Mackay and Bundaberg. This highlighted the need to be self-sufficient in having back up seed sources.

The Tissue culture project started in 2014, with the then SRA PEC research project (Dr Andrew Ward). Tissue culture hadn’t really been adopted in Far North before. Few growers and TCPSL staff had no idea how to grow and manage tissue culture plants. We saw that some work had taken place in the Herbert and Maryborough had virtually fully adopted tissue culture in their region.

TCPSL and the PEC unit were keen to learn and support each other and further demonstrate tissue culture plants. This project is a 3 year demonstration to our members and to TCPSL staff. We wanted to understand and broaden our knowledge of tissue culture practices, extend our knowledge in a new adoption of planting, growing, irrigating, fertilising and chemical application.
Key issues encountered in our industry:
1. Cyclone damage to our seed sources
2. The need to quickly increase the volume of disease resistant varieties
3. Increase the adoptions of new varieties
4. Adoption of technology associated with tissue culture propagation.

Outputs and Achievement of Project Objectives

To increase the area in the Tully growing region which is commercially planted with material 2 years or less away from an approved seed source as recommended by SRA

Over the last 3 years this project was widely demonstrated around the Tully region. Many growers ordered the new varieties as Tissue culture. The plantlets were distributed and planted into:
- Different plant spacing
- Different types of irrigation
- Different planting times
- Different fertiliser applications
- Hill up vs Conventional
- Difference climate conditions
- Pot sizes 50:40mm vs 105:25mm
- Different planters

Table 1: Seedling Numbers per Year for Tully

<table>
<thead>
<tr>
<th>Year</th>
<th>Number of tissue culture orders</th>
</tr>
</thead>
<tbody>
<tr>
<td>2014</td>
<td>1,600 seedlings</td>
</tr>
<tr>
<td>2015</td>
<td>5,700 seedlings</td>
</tr>
<tr>
<td>2016</td>
<td>17,000 seedlings</td>
</tr>
<tr>
<td>2017</td>
<td>7,300 seedlings</td>
</tr>
</tbody>
</table>

There was an increased demand for tissue culture seedlings since the projects inception in 2014 in the Tully region as the table above reflects.

To increase the numbers of tissue culture seedlings ordered from Tully up to 10,000 plants each year and to increase the numbers of growers regularly ordering tissue culture.

Since the commencement of the tissue culture demonstrations in 2014 the order of seedlings has increased. Growers who have participated in our demonstration are from different growing areas: Kennedy, Bilyana, Murray, Warrami, Euramo, Riversdale, Lower Tully, Syndicate, Feluga and El Arish.

Table 2: Growers Involved

<table>
<thead>
<tr>
<th>Year</th>
<th># Growers</th>
<th>District</th>
<th>DEMO / Order</th>
</tr>
</thead>
<tbody>
<tr>
<td>2014</td>
<td>1</td>
<td>Warrami</td>
<td>A. Nucifora</td>
</tr>
<tr>
<td>Year</td>
<td>Variety</td>
<td>Description</td>
<td></td>
</tr>
<tr>
<td>------</td>
<td>---------</td>
<td>------------------------------------------------------------------------------</td>
<td></td>
</tr>
<tr>
<td>2014</td>
<td>Euramo</td>
<td>D. Marsilio &amp; S. Silvestro</td>
<td></td>
</tr>
<tr>
<td></td>
<td>TCPSL Demo</td>
<td>Mission Beach Seed Plot</td>
<td></td>
</tr>
<tr>
<td>2015</td>
<td>Warrami</td>
<td>Mackay and L. Blenners, Henry and Nucifora</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Bilyana</td>
<td>Bosworth and Machan</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Murray</td>
<td>Dore</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Euramo</td>
<td>Silvestro</td>
<td></td>
</tr>
<tr>
<td></td>
<td>TCPSL Demo</td>
<td>Mission Beach Seed Plot</td>
<td></td>
</tr>
<tr>
<td>2016</td>
<td>Warrami</td>
<td>C. Condon, G. Henry, A. Nucifora, L. Blenner &amp; P. Vecchio</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Murray</td>
<td>Upper</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Sweet cane, G. Dore &amp; Dore &amp; Co</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Kennedy</td>
<td>D. Singh</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>Euramo</td>
<td>D. Marsilio, S. Silvestro, L. Cabin, F. Vecchio,</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Syndicate</td>
<td>A. Apap</td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>El Arish</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Lower Tully</td>
<td>L. Collins</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Bilyana</td>
<td>R. Bosworth</td>
<td></td>
</tr>
</tbody>
</table>

**Table 3: Total number of Varieties Ordered**

<table>
<thead>
<tr>
<th>Varieties</th>
<th>Number of varieties orders</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>2014</strong></td>
<td></td>
</tr>
<tr>
<td>Q231A</td>
<td>200</td>
</tr>
<tr>
<td>Q232A</td>
<td>200</td>
</tr>
<tr>
<td>Q242A</td>
<td>100</td>
</tr>
<tr>
<td>Q252A</td>
<td>600</td>
</tr>
<tr>
<td>Q253A</td>
<td>500</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>1,600 seedling</strong></td>
</tr>
<tr>
<td><strong>2015</strong></td>
<td></td>
</tr>
<tr>
<td>Q253A</td>
<td>1400</td>
</tr>
<tr>
<td>Q208A</td>
<td>600</td>
</tr>
<tr>
<td>Q252A</td>
<td>1700</td>
</tr>
<tr>
<td>Q242A</td>
<td>500</td>
</tr>
<tr>
<td>Q250A</td>
<td>500</td>
</tr>
<tr>
<td>Q231A</td>
<td>100</td>
</tr>
<tr>
<td>Q240A</td>
<td>600</td>
</tr>
<tr>
<td>Q247A</td>
<td>100</td>
</tr>
<tr>
<td>Q254A</td>
<td>100</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>5,700 seedlings</strong></td>
</tr>
<tr>
<td><strong>2016</strong></td>
<td></td>
</tr>
<tr>
<td>SRA1A</td>
<td>6,900</td>
</tr>
<tr>
<td>SRA2A</td>
<td>5,250</td>
</tr>
</tbody>
</table>


This table indicates that there is an increase of grower orders and growers continual participation.

To develop the skills and knowledge of Productivity staff to advise on Tissue culture husbandry so they are confident to continue promoting tissue culture.

TCPSL staff organised 10 different chemical mix trials to control grass and weed issues.

All trails were sprayed on 22/10/16. Start time 8:30 am, sprayed by Quadbike @ 218 l/ ha, Wind speed 10km/ha, Temp 26.2, Humidity 65.1 and Delta T = 6.2 (See table below)

**Table 4: Herbicide Treatments**

<table>
<thead>
<tr>
<th>Trials</th>
<th>Products</th>
<th>Rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1</td>
<td>Stomp Xtra</td>
<td>3.2L/ha</td>
</tr>
<tr>
<td>T2</td>
<td>Stomp + 2-4 D amine</td>
<td>3.2 + 4L</td>
</tr>
<tr>
<td>T3</td>
<td>Stomp + 2-4D amine + Atrazine</td>
<td>3.2 + 2 + 3 kg</td>
</tr>
<tr>
<td>T4</td>
<td>Stomp + Soccer</td>
<td>3.2 + 2.0</td>
</tr>
<tr>
<td>T5</td>
<td>Stomp + Soccer + 2-4D amine</td>
<td>3.2 + 2.0 +4L</td>
</tr>
<tr>
<td>T6</td>
<td>Dual Gold</td>
<td>2L</td>
</tr>
<tr>
<td>T7</td>
<td>Dual gold + 2-4 D</td>
<td>2L + 4L</td>
</tr>
<tr>
<td>T8</td>
<td>Dual Gold + atrazine + 2-4D amine</td>
<td>2L + 3L +4L</td>
</tr>
<tr>
<td>T9</td>
<td>Dual gold + Soccer + 2-4Damine</td>
<td>2+ 2+ 4Lt</td>
</tr>
<tr>
<td>------------</td>
<td>-------------------------------</td>
<td>------------</td>
</tr>
<tr>
<td>T10</td>
<td>Krismat</td>
<td>2kg</td>
</tr>
</tbody>
</table>

Overall it was found that Trial Number 8 achieved the best results and Trial Number 10 had the worst phyto-toxicity effect. More in depth trials will be conducted in following years. As you can see in the above photo we had a severe nut-grass problem in our tissue culture plot which was present in all treatment sites. We overcame this with 2 treatments of the generic version of Sempra. This was applied 2-3 weeks after the pre-emergent trial, because conventional pre-emergent brews do not generally control nut-grass. We plan to keep spraying the nut grass throughout the year in our tissue culture and where we plan to plant our next batch of plantlets. When conducting further herbicide trials the nut grass should not be an issue.

- TCPSL staff trialled three different spacing distances 30cm, 60cm and 80cm at grower demonstration sites. The best outcome of this trial was a planting of 60cm apart. Spacing’s of 30cm were too close and experienced overcrowding, resulting in small stalks. The planting of 80cm was acceptable except if you encounter problems such as plant shock, pest damage, planting, chemicals and water management, this lead to gappy rows if plants died. (see photo in appendix 1, 2 and 3)

- TCPSL staff have done a stick count of tissue culture (planted 29/9/16), Mother plot (planted 13/9/16) and commercial release (planted 13/9/16). The graph below shows the average number of sticks per 10 meter plot in five different varieties Q231^a, Q232^a, Q242^a Q252^a, and Q253^a.
We changed the pot size from Elle size 105:25mm to 50:40mm size pot. This increased the plants survival and helped to develop a stronger root system. (see photo in appendix 4 & 5)

Staff demonstrated different products of liquid starter at time of planting. Three products were trialled: Calpro, SeaSol and Supa stand Phos. There was no obvious difference in germination, cane height or cane stalk size. There was a difference in product purchase prices, Seasol was more expensive. So, all products could be recommended. (See photo appendix 6)

Growers that have already used seedlings for other crops such as watermelon and pumpkins were able to assist by using their planter and setting up trickle tape irrigation.

Irrigation setups using different types of water sources such as bore, town water, gravity feed using 3-5 000 litre tanks, natural rainfall and pumping of water from the creek or drain. Using bore water and town water there was no difference in plant growth, except for the cost. The most effective way to save water is to plant early preferably in April and slash the cane in November. This increases planting materials for the next year.

We trialled different planting methods. Conventional using normal 3 stick planter, hill up planting, flat surface and 1.9 wide conventional row planting. All planting methods were acceptable and tissue culture plantlets grew well but irrigation was very important. Irrigation was required at July/August plantings (due to drier weather) and needed to be installed properly and used immediately after planting and at timely/scheduled intervals (if no rain received) until cane was established. (see photo in Appendix 7)

Tissue Culture newly released varieties SRA1\textsuperscript{A}, SRA2\textsuperscript{A} and SRA3\textsuperscript{A} were planted using liquid fertiliser, with and without irrigation in April, relying on natural rainfall. Overall, it didn’t show much difference between the two treatments. The only difference was the varieties
performance based on soil type. SRA 3 is more vigorous compared to SRA1 and SRA2. The soil type Feluga series suited the SRA3A favourably. (see photo in appendix 8)

- Two different climate conditions, Kennedy/Warrami drier and Feluga/El Arish wetter conditions. The drier conditions required more irrigation and monitoring of scheduled watering. Less irrigation and watering was required at the wetter site.

- Based on our observations of soil types the sandy/loam soils required less minimal tillage to produce a finer tilt while in heavy soil required more tillage to produce a fine tilt before planting.

To develop a local propagation pathway with the skills and commercial interest to reduce the time between order and delivery as well as reducing unit costs by increasing volume.

- Mission Beach Tissue Culture Laboratory is a local business involved in our trial this year. Steve Lavis, owner has been conversing with SRA scientist (Prakash and George Piperidis) who are also part of this project. They are developing a petri dish and growing off the sugar cane plants to a hardening stage. We have managed to order 1000 seedlings of Q232A and 1000 seedlings of KQ228A. (Photo are available in appendix 9)

- Prakash and George have visited the Mission Beach Tissue Culture Laboratory and gave professional advice regarding procedures and protocols of growing tissue culture in the laboratory.

- The use of a local laboratory will decrease waiting times and will not be competing with other Productivity Boards ordering sugar cane tissue culture. If successful we will be able to trial different planting times according to projected weather outlooks for our region. We can also reduce the costs of freight and delivery. We can pick up locally and we do not need to pay to have tissue culture transported from Brisbane. Therefore, if the cost of tissue culture plantlets can be reduced and the uptake by growers increased we can hopefully decrease the cost of plantlets. If in the event of a cyclone we can still utilize the facilities of our off site laboratory to quickly produce plantlets for our region. This also works for other centres for example Mackay in the event of a major event they can utilise our local facility and the Brisbane facilities to quickly reproduce varieties.

- Brian Cridland, Mission Beach Foliage has previously separated planting material sent to us from SRA/Vitroflora Pty Ltd in flasks. Brian then grew them on to hardening stage in the nursery. We then took delivery and distributed to our members.

- We have additional expenses as we have to pay SRA/VitroFlora Pty Ltd for production of the sterile plantlets. If we order more than 2,000 it becomes cheaper and then again a reduced cost if order over 10,000 plantlets. We then need to pay freight to get them to the nursery for separation. We then have to pay again to collect them at hardening stage. Again the more that we have ordered the cheaper it becomes.

- Final cost to grower averaging $2.20 to $2.50 per plantlet (depending on quantity ordered).
Brian Cridland was very successful gaining planting material to the hardening stage at Mission Beach. He managed to harden and distribute 18,000 seedlings last year from growers’ orders including our demonstration trials.

Brian has increased his green house facilities in 2015 and can now handle between 15,000 – 20,000 tissue culture plants. The key focus is for future demand in the wet tropics region.

To try and reduce the cost of plantlets we are trialling Mission Beach Tissue Culture Laboratory another local business is involved in our trial this year. Steve Lavis has been conversing with Prakash and George from SRA who are part of this trial. They will be provided the cabbage (clean seed plant source) and the laboratory will dissect into sterile petri dishes and generate tissue. The plantlets will then be gowned on to hardening stage in their nursery. Therefore, cutting out the cost of providing material in plant form from the VitroFlora Pty Ltd step.

Steve has managed to generate tissue culture plants from petri dish to separation stage. They have now progressed to hardening stage. Plants should be available for delivery to TCPSL by April 2017. TCPSL will plant this first trial to observe the quality and survival of material. (see photo in appendix 9)

Steve will trial different sizing of pots and potting mix during hardening stage. Our target is to deliver the material within 7 months for growers to use for planting if this trial is successful. We are unsure of the costs of production at this stage.

Key focus is to reduce the unit cost of planting materials and produce strong and healthy plantlets at a reasonable cost for maximum uptake.

TCPSL has had two new bench tables built for tissue culture plants that we can care for plantlets until delivery to growers. These tables can store at least 3600 plants.

**Outputs and Outcomes**

This project has delivered many outputs that have helped to develop TCPSL staff skills, grower’s experience, increased knowledge about tissue culture, and some technical and practical ways of growing tissue culture, for our region.

We have developed a Planting Tissue Culture Procedure laminated information sheet including the SRA Information Sheet ISI3086 “Planting and managing your tissue culture plantlets in the field”. (See appendix 10)

This laminated sheet is given to growers prior to planting to ensure the ground is prepared to fine tilt and fertiliser is incorporated into ground prior to delivery – “Prior to Planting”. Plantlets are soaked in liquid fertiliser before planting and they have their irrigation ready for set up immediately after planting to ensure a good take up- “Day of Planting”. The procedures also include after planting requirements.

Tissue culture updates were included in our TCPSL Newsletter, issued quarterly. Presentations of the outcomes of the tissue culture trials were delivered at our Breakfast Meetings. We presented the key objects and the benefits of growing tissue culture to our members. A small bus of growers visited a MSF tissue culture trial on the
tablelands. Growers asked questions and observed the growth of advanced plantlets using a different irrigation system, underground watering with tape.

TCPSL purchased a tissue culture planter that we allowed members to use (engineering accredited). We had built a rack (6000 – 8000 plants) to transport tissue culture plants to our members. We also had built a bench top to store tissue plants in case of a natural disaster during the delivery time. We have purchased and installed irrigation supplies in our clean seed plot. Our intention is to use tissue culture every year to maximise propagation of new varieties.

We have demonstrated to growers how to plant and grow tissue culture plants. During our first demonstration we encountered unexpected problems. For example lack of water, weeds, not enough nutrient and other natural misfortunes such as rats and wallabies. Through experience we acquired knowledge and put into place strategies to increase health and production of the plants.

In our last year of the trial we increased the number of demonstration sites to include all districts with different soil types. With the increased awareness and exposure we were able to increase our plant orders from 5000 - 17000 plants. This showed increased grower interest.

Photographic evidence, workshop activities and Newsletter (see Appendix 11)

**Intellectual Property (IP) and Confidentiality**

Not applicable for this project

**Industry Communication and Adoption of Outputs**

We have communicated the trial outcomes from this project through:
- our quarterly newsletter to members,
- breakfast meetings with growers,
- industry reports and board reports ( Mill and TCPSL Board),
- magazine article in the SRA quarterly magazine “Cane Connection”,
- newspaper article “North Queensland Register”,
- growers walk at Mission Beach Seed plot
- TCPSL face book, growers who are signed up to this page.

What new information, if any, is available on the adoption of project outputs?
See Appendix

List any newsletters, fact sheets or any other media coverage.
See Appendix

Identify any further opportunities to disseminate and promote project outputs at seminars, field days etc.
Environmental Impact
Outline any new information on adverse/beneficial environmental impacts of conducting the project and/or implementing its findings.

Reduction of water usage: TCPSL trial on early planting in (April) to reduce the water consumption.

By planting tissue culture plantlets from April to June often doesn’t require any irrigation. The plantlets can establish good growth through rainfall and normal weather conditions. This will help to improve environmental impacts.

Timing of planting, fertilising and spraying this will also help to improve water quality in the water system.

Recommendations and Future Industry Needs
Include activities or other steps to further develop, disseminate, commercialise or exploit the Project Outputs and realise the industry benefits.

Diagram 1 Steps to order and grow tissue culture plants

Industry Benefit
- Access of clean seed
- Increase new variety populations available to growers
- Grower can order more seedlings if the cane is approved by industry
• Adopt new method of planting sugar cane and new technology
• Growers can maximise a small area to plant tissue culture and by next year they can plant new varieties in their commercial area.
  o Normal planting using long stick planter around 10 sticks per meter. Tissue culture plants, one plants gives 15 – 18 sticks.
  o Every 10m, the average cane stalk using long stick planting 100 - 120 sticks, while tissue culture stalk per 10 m around 250 – 270 sticks
• Reduce the risk of mechanical accidents and time planting
  o long stick planting requires
    ▪ couple of tractors, planter, fertiliser bag lifter, plant marker, plant cutter, cane trailer, cane stripper, and man power
  o Tissue culture
    ▪ tractor, long stick planter, tissue culture planter,
• If there a natural calamity in the Tully regions we can recover our planting materials quicker, same with new varieties.

**Nursery and tissue culture plants**

**Benefits**
• Good nursery operation and management helps to reduce the risk of materials being damaged and produces strong healthy plants.
• Nursery is the final step before tissue culture is planted (good hygiene, sufficient nutrients applied is important) to produce healthy tissue culture plants.
• A good seed always bears a good fruit.

**Land preparation**
In heavy clay soil preparation requires a couple of passes or rotary hoe to have a fine tilt before planting. In sandy loam soil this require less passes before planting. The photo below is an example of inadequate land preparation that tissue culture plants encounter problems at growing stage, it ends up a big lump of soil. Lumpy soil won’t store enough water and creates air pockets in the soil and is hard for the root system to grow and absorb the nutrients under the ground (Soil- root system contact). Good land preparation and the right amount of nutrients to the soil prior will help the plant to grow quick and remain healthy.

If the dirt is too dry it is okay to plant tissue culture as long as the irrigation system is set up immediately after planting. We suggest that before the tissue culture plants are transferred into the ground they are soaked in a large container with growth enhancer like SeaSol, Calpro and other liquid plant starters. This soaking will help to eliminate plant stress and dryness in the root system and promote healthy growth.

Several weeks after planting tissue culture requires spraying of a pre –emergence, stomp xtra and atrazine or dual gold and atrazine. (Rate please refer on page 8) to control weeds and to enhance plant growth.

Ten weeks after planting the irrigation system needs to be removed to allow mechanical hill up or cotton king to scratch the dirt and aerate the soil. If the cane is intended for planning material, we didn’t advise fertilising the cane in the second round in order to maintain straight cane that will stand up to eliminate lodging.

Tissue culture management is similar to commercial cane operations. The only difference is the early stage of planting. Tissue culture requires close monitoring and correct management while the irrigation is set up. The plant is too soft and young and requires the watering system to be monitored closely according to the current weather conditions and to ensure the watering system is working efficiently. Continual monitoring and investigation of the young crop to ensure it is free from pest and diseases. If the plant grows normally, 10 weeks after planting manage as a normal commercial cane.

Figure 2: Soil conditions not recommended for tissue culture planting.
This an example of dry soil and we setup irrigation immediately after planting. This sandy loam soil is easily managed before planting. If the dirt is finer before planting the plants have a better outcome and the root system will penetrate the fertiliser easily in the ground. The quicker the water is established to wet the surface, the plant will grow faster and will recover from stress, increase their root system and develop faster.

**Types of irrigation**

There are a couple types or methods to irrigate tissue culture plants, this all depends on growers’ availability to a water source and convenience. Below are the examples of possible water sources to irrigate tissue culture plants.

---

*Figure 3: This is an example of fine tilt soil that is recommended for tissue culture planting*

*Figure 2: Trickle tape set up ready for irrigation*
Figure 5: Gravity feed 1000-5000 litre tank, this grower tried using his vehicle to irrigate tissue culture plants. The photo below is a 1000 litre tank on the back of a Ute. The grower drives up and down the rows where tissue culture was planted. Some growers have a 5000 litre tank parked on the headland and use gravity feed to connect to the trickle type irrigation.

Figure 6: Another photo sourcing water from drain or creek. The grower has set a petrol motor to pump the water from the drain. We experience problem using drain water into trickle Type. No filtration to stop dirt entering trickle type irrigation. Our next alternative is to run the hose in the farrow and do flood water irrigation this will allow the water pressure form the pump to release easily to the ground, in doing this require monitoring every couple hours ensure that the pump and tissue culture plant not over water and pump working in the right speed and pressure.
Figure 7: Bore water, another source of irrigation. This grower who also grows banana and other crops has access to bore water. Majority of our bore water trials were successful, we didn’t encounter a problem with this option.

Figure 8: Town water was another option if the farm is closer to town and main roads. Town’s water has an easy access and can be easily installed to trickle type irrigation. If the irrigation pump is set up with an automatic timer this creates less work to manage tissue culture. Normally we sent the timer for early in the morning until 8 am and afternoon around 6pm until 9 – 10 pm it all depends on the distance and number of plants.
Different types of tissue culture planters

In Tully there are couple of planter types that were used to plant tissue culture. The photo and video below help us to determine which one is much better to use. From experience all the planters were useful and there was no difference between each planter.

No. 1 Planter

No. 2 Planter
The next planter was more for commercial farming where they can plant multiple crops.

No. 3 Planter
TCPSL considered buying this style of planter because it is simple and suited the purpose of our project. We demonstrated the use to growers and allowed growers to trial on their farm to see the different.

Figure 9: Planter Type
### Table 5: Cost analysis - planting cane using long stick machine, billet and tissue culture

<table>
<thead>
<tr>
<th>Equipment</th>
<th>Area</th>
<th>Cost</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Long stick planting in one acre of land</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>100m = 1000 stick</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Average 10 stick/meter</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1 x Tractor with 1 x cutter (contractor charges)</td>
<td>$165/tonnes</td>
<td>100m</td>
</tr>
<tr>
<td>1 x Tractor and 1 x stripper (contractor charges)</td>
<td>$25/tonnes</td>
<td>100m</td>
</tr>
<tr>
<td>1 x Long stick trailer and delivery (contractor charges)</td>
<td>$110/hr deliver</td>
<td>100m</td>
</tr>
<tr>
<td>4 x Staff involved in operation</td>
<td>$25/hrs</td>
<td></td>
</tr>
<tr>
<td>1 tonnes cane (Clean seed) = 1000 sticks</td>
<td>$110/tonnes</td>
<td>1 acre / 1000 plants</td>
</tr>
<tr>
<td>1 x Long stick planter</td>
<td>$250/ha</td>
<td>1 acre</td>
</tr>
<tr>
<td><strong>Total Cost charge around Tully area</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Billet planting in one acre of land</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>400m = 4000 sticks</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1 x Harvester</td>
<td>400m</td>
<td></td>
</tr>
<tr>
<td>1 x Tipler and 1 x Tractor</td>
<td>400m</td>
<td></td>
</tr>
<tr>
<td>4 x Staff involved in operation</td>
<td>$25/hrs</td>
<td></td>
</tr>
<tr>
<td>1 x Billet planter</td>
<td></td>
<td>1 acre</td>
</tr>
<tr>
<td>4 tonnes cane = 4000 sticks</td>
<td>$110 per tonnes</td>
<td>1 acre</td>
</tr>
<tr>
<td>Billet charge around Tully</td>
<td>$190/acre</td>
<td>1 ha</td>
</tr>
<tr>
<td><strong>Total cost of Billet</strong></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### Tissue culture aver stick 15 sticks per plants

<table>
<thead>
<tr>
<th>Equipment</th>
<th>Area</th>
<th>Cost</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 x Tractor to fertilise</td>
<td></td>
<td>$60/ha</td>
</tr>
<tr>
<td>1 x Tractor and planter</td>
<td></td>
<td>$60/hrs</td>
</tr>
<tr>
<td>66 x plants (TC) = 40 m</td>
<td></td>
<td>$2.20/plants</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Irrigation material and equipment</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Trickle type 3/4’ $206 (1600m), fittings ¾ $1.45 each, lay flat 2’ $275 /100m</td>
<td></td>
<td>$6 Trickle type $5 Fitting $3 Flat</td>
</tr>
<tr>
<td>Water usage</td>
<td></td>
<td>3-4 hours per day</td>
</tr>
<tr>
<td>4 x Staff involved in operation</td>
<td></td>
<td>$25/hrs</td>
</tr>
</tbody>
</table>

| Total cost tissue culture | | $379/40m |

**Table 6: Summary - Costs of different types of planting methods**

<table>
<thead>
<tr>
<th>Summary cost</th>
<th>Total per ha</th>
</tr>
</thead>
<tbody>
<tr>
<td>Long stick planting</td>
<td>$760</td>
</tr>
<tr>
<td>Billet planting</td>
<td>$909</td>
</tr>
<tr>
<td>Tissue culture</td>
<td>$379 excluding water usage</td>
</tr>
</tbody>
</table>

### Publications

Publications please see in the appendix
Appendix

Tissue culture plantlets - planted 30cm apart

Tissue culture plantlets - planted 60cm apart
Tissue culture plantlets - planted 80cm apart

Left 50:40mm pot size and right 105:25mm plot size
Plant from left 50:40mm (bigger) pot size and on right 105:25mm (smaller) pot size

Liquid fertiliser
Hill up planting vs flat surface planting

SRA 3 planted early (April). Plants relied only on rainfall (SRA 3 more vigorous than SRA1& 2)
First trial done at Mission Beach Tissue Culture Laboratory - Steve Lavis

Tissue culture planting and managing procedures - SRA information Sheet

Planting and managing your tissue culture plantlets in the field

**Planting**

Prepare soil to a fine tilth to ensure good soil/root contact. A seedling planter can be used if one is available, although hand planting small numbers is not a huge job. Plant them deep at the bottom of a

...can be treated with the same chemicals as the ratoons on your farm. Label rates of S-metolachlor plus atrazine have been applied successfully over the top after planting. For example we used Atralatex @ 2.5 kg/ha plus Dual Goal @ 1.5 l/ha for grasses and broadleaf weeds and also Sermpra @ 100 g/ha plus Activator @ 200...
Tissue culture planting and managing procedures – TCPSL information (reverse side)

PLANTING TISSUE CULTURE

PROCEDURES

PRIOR TO PLANTING
1. Ensure ground is prepared to FINETILT. (No. of plants x 0.6 = length of row required).
2. Incorporate fertiliser into ground preparation.
3. Pre - Purchase
   a. Standard trickle tape, holes 60cm apart,
   b. 2 inch lay flat, plus any connectors and hardware required,
   c. Lay flat connectors (alternatively if a small area a garden hose and
counters can be used),
   d. Liquid starter

DAY OF PLANTING
1. Tractor with 3 point linkage (High Rise not suitable) for Tissue Culture Planter.
2. Soak tissue culture in liquid fertiliser (seasol or equivalent) for up to 30 minutes
   prior to planting.

AFTER PLANTING
1. MUST water twice daily, preferably early morning and late afternoon, for 1 month.
2. Apply pre-emergent approximately two (2) weeks after planting. Stomp 440 or
   Dual Gold with Atrazine, NO GRAMOXONE.
3. Treat cane as plant cane thereafter.
4. At stooling stage, fill up ground along rows.
5. Treat like normal cane thereafter.

T:\Tissue Culture\GRA extension project folder (Tissue Culture)\flyer
TCPSL Newsletter sent to all members - quarterly

SEEDPLOT
The first inspections have been conducted on all seedplots. Details on germination rates, pest and disease and other notable information has been documented.

- Spray operations have been undertaken on headlands and clones that weren’t released.
- TCPSL 2016 Merryburn Seedplot was heavily damaged by climbing rats. Varieties most susceptible are Q250\(^{9}\), Q252\(^{9}\), SRA1\(^{9}\), Q200\(^{9}\) and Q247\(^{9}\).
- Baiting has already commenced in an attempt to control vermin.
- 2016-17 Seedplot
  - Euramo Seedplot (Damien Rigato) for 2017 has been planted with varieties: KQ228\(^{9}\), SRA3\(^{9}\), Q200\(^{9}\), Q208\(^{9}\), Q251\(^{9}\), Q252\(^{9}\), Q240\(^{9}\), Q231\(^{9}\) and one possible new variety for next year. This plot will be accessible for long stick, hand cutting and billet machine.

Tissue Culture Project
This is a SRA funded project to increase the adoption of tissue culture in the Tully region. TCPSL has been actively promoting tissue culture as an alternative source of clean seed for growers and a great way to adopt new varieties quickly as the grower is not constrained by their allocation at the clean seed plots.

The 2016 demonstration trial “Managing Impediments to Adoption in Tully” is due for completion in January 2017. Eight growers participated in the program with assistance given by the TSPSL team. In the adoption process, TCPSL assisted growers to plant tissue culture using a seedling planter and in the preparation of irrigation for the plantlets. It has been somewhat successful in adopting tissue culture practices with a small number of growers moving to tissue culture for all of their clean seed requirements. For the 2016 planting season, 17,150 plantlets were ordered. The most common being SRA1\(^{9}\), SRA2\(^{9}\) and SRA 3\(^{9}\). An order for 6,200 plantlets has been submitted by growers for delivery in 2017, with popular varieties comprising of SRA6\(^{9}\), Q200\(^{9}\) and Q231\(^{9}\).

Best Management Practices
Best Management Practices (BMP) is a Canegrowers run project supported by TCPSL. The idea of BMP is to show that the cane industry is a responsible and sustainable self-regulated industry that uses best management practices both for productivity and environmental outcomes. BMP is also a way of marketing sugar to the world, showing them it is a sustainable product. For those growers interested in becoming
Other Photographic Evidence

**Photo 5**: Setting up a water source for tissue culture

**Photo 6**: Nut grass issue in tissue culture plants
Photo 7: Group discussion with interested growers about tissue culture