NORTHERN TERRITORY GOVERNMENT

DEPARTMENT OF RESOURCES

PRIMARY INDUSTRIES ANNUAL RESEARCH REPORT 2009-10

October 2010

Bibliography:

Northern Territory Government (2010). Primary Industries Annual Research Report 2009-10. Department of Resources. Technical Bulletin No. 335.

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INTRODUCTION

Welcome to the Northern Territory (NT) Department of Resources' Primary Industries Division's 2009-10 Annual Research Report (ARR). The ARR provides a summary of new and continuing research work conducted during the 2009-10 financial year, together with results, where available, and general recommendations.

The research projects reported in this ARR cover two major areas: Animal Industries – cattle, pasture and buffalo production; and Plant Industries - crop, forestry and horticulture production. Some research covers both animal and plant industries, in such areas as diagnostics, biosecurity and product integrity. Much of this work is conducted at the Department's headquarters at Berrimah Farm, in Darwin, at its regional facilities in Alice Springs, Katherine and Tennant Creek and on its research farms. Work is also conducted on private properties in collaboration with owners.

The pastoral industry occupies about 55% of the NT and generates about 44% of the total production of its primary industries and fisheries. Beef production is the NT's premier agricultural industry, providing significant income and employment in regional areas.

There are about 300 active cattle stations in the NT, raising about 2 million beef cattle. In general, most NT cattle properties are managed as extensive, large-scale, low-cost operations. Some properties are more than 10 000 km^2 in area; the average property is around 3100 km^2. To enhance adaptation to tropical conditions, most properties in the northern (humid) part of the NT raise cattle with high *Bos indicus* (Asian breed) genes. Those animals are mostly destined for live export to South East Asia. In 2008-09 the live cattle export trade was worth about $160* million. On the other hand, cattle properties in the southern part of the NT, where the climate is less humid, generally raise animals with high *Bos taurus* (European breed) genes. Those animals are sent either to domestic markets in southern Australia or exported live to Asian markets, depending on best returns.

The Pastoral Production Group provides research, development and extension services to facilitate the sustainable development of the pastoral industry in the NT. The services, covering cattle and, to a lesser extent buffalo, include sustainable rangeland management, grazing strategies and improved pastures. They also attempt to improve breeder herd efficiency, enterprise management and profitability, live-weight gain, genetic content and ways to meet local and export market specifications. Moreover, technical services are provided to Asian trading partners to develop import markets in South East Asia. Also, assistance is provided to improve Indigenous economic development through the Indigenous Pastoral Program.

The project reports presented in the Animal Industries Section of the ARR describe current research on most of the productivity improvement issues described above. Depending on the stage of each project, some introduce recently commenced work, others provide updates on progress achieved so far and some, that have been completed, present extensive results and conclusions.

Similarly, the Plant Industries Group focuses on research efforts to maximise sustainable production of crops in the NT, mainly for the domestic market. It provides scientific and technical advice to increase productivity, efficiency and product quality; develop and maintain markets; strengthen business adaptability and enhance sustainability and natural resource management.

Mango production, which was worth $46 million, represented more than a third of the total annual production of plant industries, which was worth $118 million. Other major crops were melons worth $24 million, vegetables worth $20 million, nursery products (including cut-flowers) worth $19 million and field crops, including fodder, worth $13 million. The balance is made up of bananas, rambutan, table grapes and citrus (Figure 1).

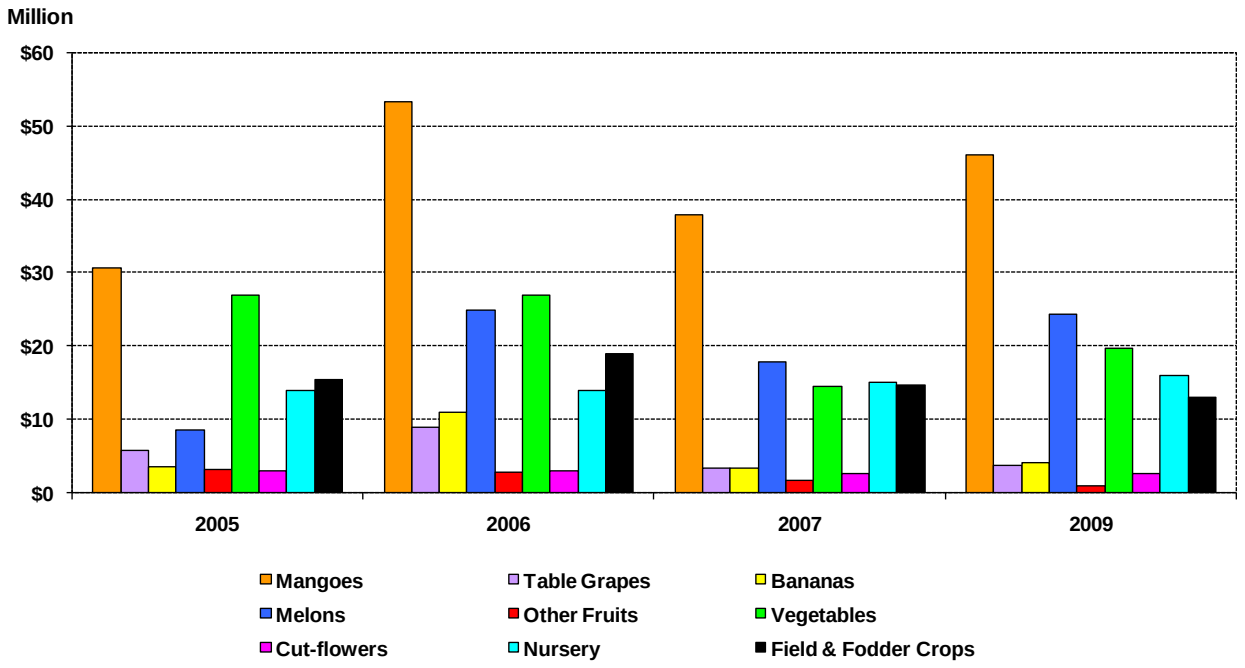

Figure 1. Gross value of NT plant industries 2005 – 2009

Horticultural crop production is continuing to grow, particularly in mangoes, passionfruit, snake beans and alternative Asian vegetables. Broadening market options for NT produce, particularly taking advantage of being "first on the market" seasonally is critical for industry growth, as is the development of new products to meet high value niche market demand.

Enhanced efforts in innovative integrated pest and disease management, focusing on natural plant defence mechanisms and insect chemical ecology, will help reduce the need for post-harvest chemical treatments, which are at present necessary in order to maintain market access.

A partnership with pastoral and plant industries together with other research providers will help in achieving these targets. With finite research and development funding, it will be necessary to focus on crops and animals that have a high market demand.

Projects related to plant industries are listed under the heading 'Plant Industries', irrespective of which departmental Group they come under. Similarly, projects related to animal industries are listed under 'Animal Industries', irrespective of which departmental Group they come under. To further assist readers to find projects of interest quickly in this ARR, two indexes are included at the end.

The ARR is published every year in October. Suggestions for improving the content, layout and structure of future editions of the ARR are most welcome and should be forwarded to technical.publications@nt.gov.au.

*Figures cited in this text are forecasts, which are sourced from: Department of Resources (2010). *Northern Territory Rural Industries and Fisheries Economic Outlook 2009,* which is available from the Department's website. At the time of publication, this document was located at: http://www.nt.gov.au/d/Content/File/p/annualreport0506/Economic_Outlook_2009.pdf

ANIMAL INDUSTRIES

Animal industries projects conduct applied research in controlled trials to discover solutions to problems that affect productivity and profitability of the industry and, where possible, to protect the environment and human health.

PROJECT: 21st Century Pastoralism

Project Officer: A. Bubb

Location: Alice Springs

Keyword(s): Desert Knowledge CRC, telemetry, cattle, technology

Objectives:

To provide research, development and extension services to commercial pastoral producers in the Alice Springs region and the broader desert regions of Australia as a component of the Desert Knowledge Cooperative Research Centre.

To increase the economic and livelihood opportunities associated with desert pastoralism by developing unique enterprise models, economic development systems and pastoral management systems that provide tangible benefits to producers.

Background:

The 21st Century Pastoralism Project attempts to increase the economic viability of desert pastoral enterprises. The project analysed the cost benefit of labour-saving devices, including telemetry systems. It has also created a remote livestock management system called 'WaterSmart', which evaluates commercially-available products for delivering and monitoring stock water.

The Cattle and Country Project completed evaluations of the Kimberly Indigenous Management Support Service (KIMSS) and the Indigenous Pastoral Program (IPP) and also reviewed Indigenous participation in the northern beef industry for Meat and Livestock Australia (MLA) and the Indigenous Land Corporation (ILC).

Method:

The cost of bore runs was determined at three stations in the Utilising Technology Project and telemetry monitoring systems were installed on all of them.

The development of a remote livestock management system (RLMS) is almost complete and is being commercialised through Precision Pastoral Pty. Ltd. Two RLMS units were purchased by the Department of Resources for research purposes.

The Cattle and Country Project completed evaluations of KIMSS and IPP and also reviewed Indigenous participation in the northern beef industry for MLA and ILC. The evaluations and review were based on extensive stakeholder interviews and visits.

Results:

The telemetry systems in the Utilizing Technology and 'WaterSmart' projects generated significant savings for the properties involved, reducing the cost of monitoring and maintaining stock water by 30 to 50%. The investments in the new technologies ranged between $30 000 and $70 000 and had a cost recovery period of six to 24 months.

RLMS was used in commercial operations at Napperby Station for data collection and mustering purposes. Drafting accuracy increased to 96% and 'walk-over-weighing accuracy' has also improved significantly.

The findings of the evaluation of KIMSS and IPP were presented to ILC in two individual reports. The Indigenous Pastoral Employment Review identified the lack of a clearly defined career pathway as the greatest impediment to long-term Aboriginal employment in the pastoral industry. An economic analysis of Aboriginal-focused training programs to increase participation in the pastoral industry has shown high economic returns to the NT. MLA is using the findings to form its future employment strategies for the northern beef industry.

PROJECT: Cell Grazing of Improved Pastures in Northern Australia for Increased Beef Production and Soil Carbon Sequestration

Project Officers: T. Schatz, D. Ffoulkes and P. Shotton

Location: Darwin

Keyword(s): cell grazing, carbon, beef production, pasture, cattle

Objectives:

To determine if cell (time-controlled) grazing of beef cattle in a monsoonal tropical climate results in improved beef production per hectare.

To determine the effects of cell-grazing on productivity and composition of a tropical pasture (buffel grass) in a monsoonal tropical climate.

To determine the effects of cell-grazing on soil uptake and storage of carbon.

To assess the benefits and costs of two different grazing management systems (cell vs. continuous grazing) on tropical pastures in parts of northern Australia where beef production is more intensive (such as the Douglas Daly and Katherine regions of the Northern Territory (NT)).

To evaluate the use of 'walk-over-weighing' and auto-drafting equipment in cell grazing systems.

Background:

It is claimed that cell-grazing (CELL) management systems produce more beef and are more resilient to climate change compared with continuous grazing because of better pasture productivity, increased soil carbon uptake and storage (carbon sequestration), resulting in better soil fertility. The cell grazing project at Douglas Daly Research Farm (DDRF) attempted to study the effects of set-stocking compared with CELL on animal and pasture production, pasture composition and sequestration of soil organic carbon (SOC).

Method:

The trial uses young animals shortly after weaning for about one year when they are replaced by fresh weaners. The trial started in late August 2009.

A 32 x 6 ha lattice of uniform buffel pasture paddocks are used on sandy Blain soils at DDRF to compare the benefits and costs of cell and continuous grazing on animal and pasture production and on SOC. Twenty six paddocks are used for CELL and the rest are continuously grazed at two stocking rates with three replicates for each stocking rate (see Figure 1).

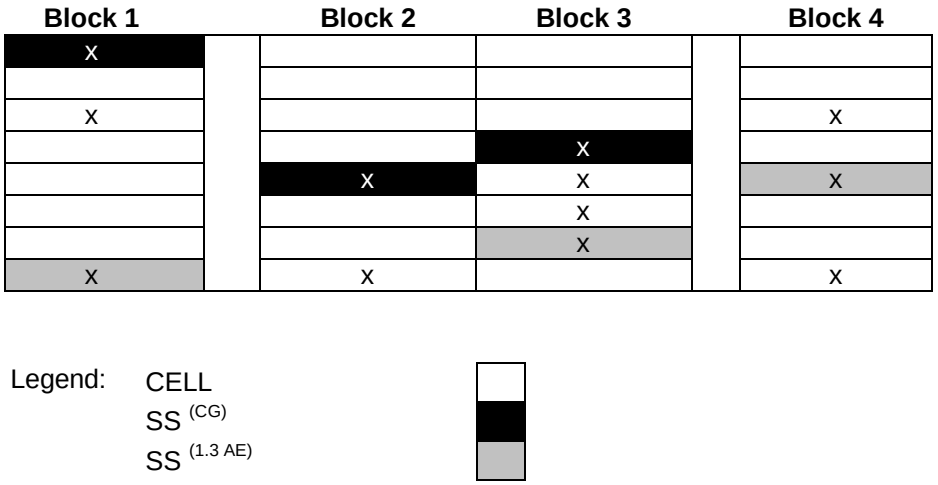

Figure 1. Paddock use and soil sampling sites (x) during 2010-14

One set-stocking rate (SSR) (1.3 AE) will remain at 1.3 animal equivalent (AE)/ha for the entire project (this is considered to be the safe long-term stocking rate for this pasture) while the other set-stocking cell grazing (SSCG) will be set at whatever the stocking rate is in the entire CELL treatment, i.e. the number of CELL animals divided by the total area of all the 26 CELL paddocks combined. In the first year of the trial, the stocking rate in CELL and SSCG was 1.13 AE/ha. In the second year, it was increased to 1.42 AE/ha. (Note: Each animal is equivalent to 0.85 AE during the post-weaning year).

In general, paddock movements for the CELL group, depending on paddock condition and season, will be as follows:

- Early wet (November-December): 60 days rest period; cattle moved every two to three days.
- Mid/late wet (January-March): 25 days rest period; cattle moved daily.
- Dry season (April-October): 90 days rest period; cattle moved every three to five days.

Live-weights and body score conditions of each animal are recorded every three months. In addition, empty (fasted) live-weights will be recorded at the start and end of each 12-month trial period and at the end of the dry season (usually in late November). Fat depth will be measured ultrasonically at the end of each year. A 'walk-over-weighing' system will be used to measure live-weights in CELL animals on a more regular basis to explore the potential of this technology as a management tool to identify and draft animals that have reached turnoff weight.

Before the trial started, test core soil samples from the 10-30 cm soil profile were taken from 10 randomly-selected paddocks across the project area to assess the variation in their physical and chemical properties. The bulk density of the soil (pre-dampened) was estimated using a piece of galvanised pipe with a volume of 385.8 cm^3.

At the start of the trial, soil core samples (benchmark) were taken at different depths (0-10 cm, 10-30 cm, 30-60 cm and 60-100 cm) from 26 paddocks to analyse SOC. The remaining six paddocks were excluded from sampling as they were slightly different in soil type.

During the trial, 10 soil core samples will be taken from each control paddock and from six randomly-selected cell-grazing paddocks (see Figure 1). Sampling from each depth will be carried out twice annually, at the end of the wet and dry seasons; samples will be bulked together for each paddock.

Soil core samples will be analysed for bulk density measurements and SOC. Core samples from the upper soil profiles of each treatment will also be analysed for carbon to nitrogen (N) ratio, calcium, sulphur (S), and micro-biota. Analysis will be done at CSIRO's Analytical Services Unit using chemical and mid-infrared spectroscopy and at the Soil Foodweb Institute for micro-biota assays.

BOTANAL (Tothill et al. 1992) is used to measure pasture yield and composition twice a year (in February and September) to coincide with the mid-wet and mid-dry season. In the first year, pasture assessments were undertaken in September 2009 and February 2010 to record total yield, plant composition, the percentage of bare ground and the basal area of grass and broad-leaf plants. Plant composition and bare ground were calculated by recording plant species found and estimated percentage of area covered by each species and by bare ground in 40 x 1 m² quadrants (from each paddock using four diagonal transects and recording at every 25 m). Total plant biomass was calculated by harvesting (cutting plants at ground level) 6 x 1 m² quadrats per paddock, oven-drying and weighing the samples.

Permanent sites were set up in the six set-stocked paddocks and in nine of the cell-grazing paddocks to assess changes in plant basal area. This involved setting up two permanent posts (20 m apart) and recording the basal area of grass tussocks on a 10-m transect midway between the two posts (having the permanent posts away from the site to be measured reduces the chances of the pasture being impacted by changes in behaviour caused by the posts). The basal grass area is measured by setting up a string line between the two posts and measuring the basal width of grass tussocks that come in contact with the string line on the 10 m transect. This allows the same sites to be measured in future to assess changes in basal area. The permanent sites are also used for fixed photo points to provide a visual memory over time.

Results:

Animal production

Only preliminary data from the first year group of animals is available at this stage. The live-weight gain in each treatment group is shown in Figure 2. Analysis of data has not been conducted yet; however, Figure 2 shows that weight gain was highest in the SSR 1.3AE and the SSCG groups.

However, it should be noted that weight gain in CELL animals was greatly affected by buffalo flies in December (due to a much higher stocking rate of 208 animals in a 6-ha paddock compared with eight or nine/ha in the set stocked group). If this had not occurred, it is likely that the weight gain in all groups would have been similar. It was observed that following rain in December, the behaviour of CELL animals was affected more by buffalo flies than in set-stocked animals. In fact, CELL animals were observed huddling together and milling around (leaving large churned up muddy areas in the paddock) trying to get relief from buffalo flies. Their average weight gain between 27/11/2009 and 06/01/2010 was 19 kg less than that in SSR 1.3AE animals and 27 kg less than that in SSCG animals. These were the differences in growth between the groups over the whole year. The SSR 1.3AE group gained 18 kg more than the CELL group and the SSCG group gained 23 kg more than the CELL group, over the whole year. The advantage in growth of the set-stocked group over the CELL group occurred entirely during this period.

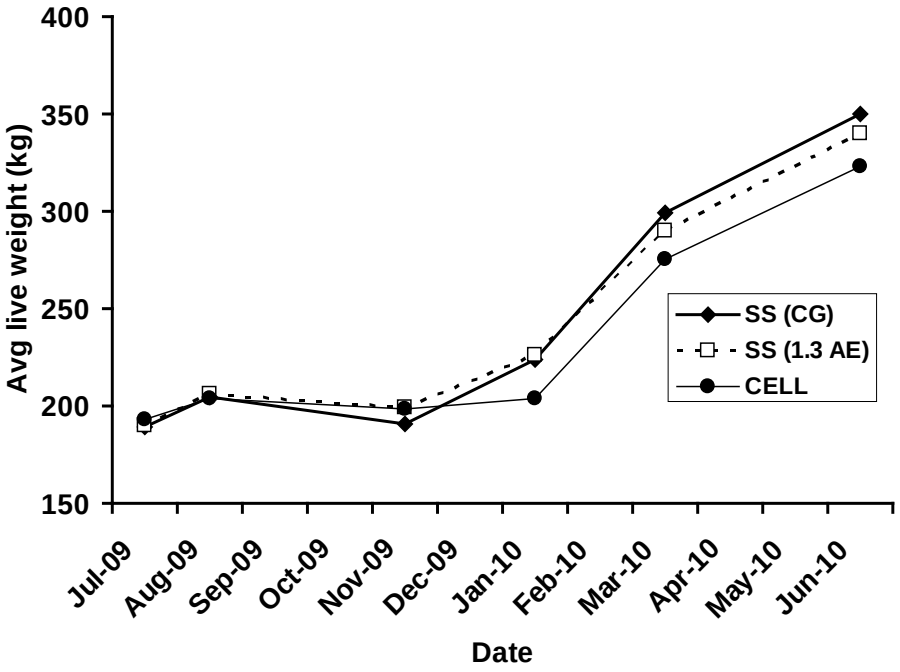

Figure 2. The average weight gain in each treatment group in the cell-grazing trial

Once the problem of buffalo flies became apparent in the CELL group, they were tagged with insecticidal ear tags. From then on, growth in all groups was similar. The results have highlighted the importance of buffalo fly control in cell-grazing systems.

Phosrite® lick blocks were supplied ad-lib to the set-stocked and cell-grazing cattle during the wet season and Uramol® blocks during the dry season. The consumption of Uramol® in the CELL group averaged 133 g/animal/day (over 108 days); the set-stock group consumed 135 g/animal/day in the heavier-stocked paddocks and 109 g/animal/day in the lighter-stocked paddocks.

The consumption of Phosrite® in the CELL group was 75 g/animal/day (over 164 days) while the set-stock group consumed 111 g/animal/day in the heavier-stocked paddocks and 107 g/animal/day in the lighter-stocked paddocks.

The CELL group was moved a total of 160 times between 01/09/2009 and 08/06/2010 and stayed in each paddock between 24 and 84 hours. From the start of the trial on 01/09/2009 to 21/12/2009 the cattle were moved twice a week. This was increased to every second day between 21/12/2009 and 09/01/2010 when rainfall and pasture growth increased. When pasture growth and condition was at its peak from 09/01/2010 to 08/04/2010, the cattle were moved daily. From 08/04/2010 to 08/06/2010 (when the first year group of cattle left the trial) the cattle were moved every second day as pasture growth slowed.

Soil carbon

The results of benchmark soil sampling of the project area are presented in Table 1. Bulk densities increased with depth while soil organic carbon and carbon stocks decreased with depth. The variation between the blocks of eight paddocks was significant (P=0.10) for soil organic carbon but was less significant for bulk densities (P=0.13) and not significant for carbon stocks (P=0.22). The block difference in soil organic carbon is probably due to charcoal found in one or two soil samples from a depth of 10 to 30 cm. The charcoal is likely to be from burning of trees at the time the land was cleared.

Table 1. Benchmarked soil and carbon properties at different depths and between blocks of paddocks across the project area

Depth	Bulk density (g/cm^3)	Soil organic carbon (%)	Carbon (kg/m^3)
0-10 cm	1.52	0.51	7.6
10-30 cm	1.60	0.29	4.5
30-60 cm	1.62	0.19	3.0
60-100 cm	1.64	0.12	2.0
Probability	*P*=0.00	*P*=0.00	*P*=0.00

Block	Bulk density (g/cm^3)	Soil organic carbon (%)	Carbon (kg/m^3)
Block 1	1.60	0.25	4.3
Block 2	1.59	0.31	4.3
Block 3	1.57	0.27	4.7
Block 4	1.61	0.28	4.0
Probability	*P*=0.13	*P*=0.10	*P*=0.22

Figure 3 compares the carbon stocks of Blain soils before the clearing of the Douglas Daly region in the 1970s (Day 1977) with the current carbon stocks at the project site. The undisturbed land then contained about 40% more soil carbon than today.

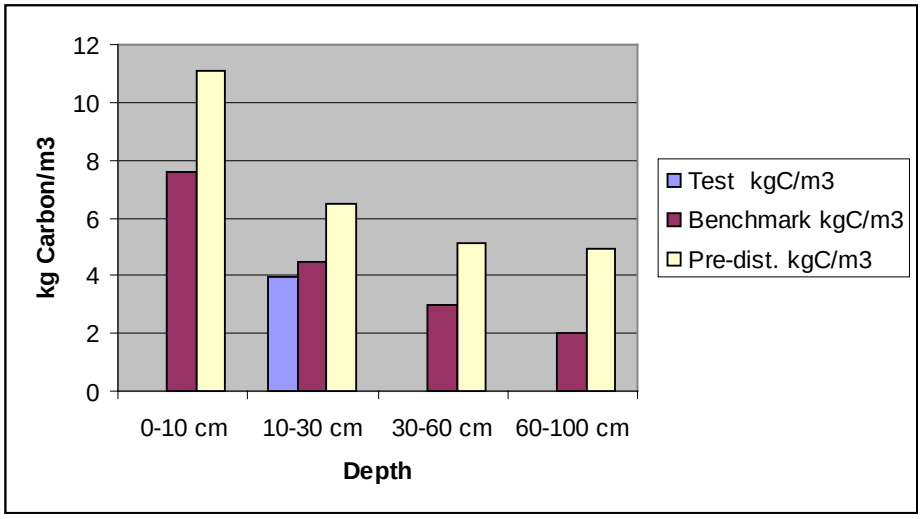

Figure 3. Subsoil carbon stock levels during pre-disturbance (1970s) and now (2009)

Pasture production

Buffel grass made up the majority of the pasture biomass of all paddocks. On average, the paddocks had 94% buffel grass in 2009 and 85% in 2010 (ranging between 74% and 99%). Buffel grass was found in nearly all quadrants assessed with the exception of those close to fence lines, corners of paddocks or near trees. Other pasture species present were sabi, pigeon grass, whitechloa and annuals with small amounts of Crotalaria, *Sida acuta*, Senna and Wynn cassia.

Plant identification was much easier during the February 2010 assessment when the plants were actively growing. During the wet season, patches of Crotalaria were noticeable.

The total average biomass in September 2009 was 3300 kg/ha and 4100 kg/ha in February 2010. The average paddock range was between 2200 and 4100 kg/ha in September and 3100 and 5100 kg/ha in February.

In February, April, May and June 2010, buffel plant tips were collected to ascertain pasture quality at different times of the year and to detect differences between the CELL and set-stocked paddocks. At that early stage, there were no differences between the set-stocked and CELL paddocks. N and S were the lowest major elements compared with recommended requirements for growing cattle; magnesium and sodium were also low. Crude protein (CP) was between 5.9 and 8.2% in the CELL paddocks and between 7 and 9.5% in the set stocked paddocks. The higher CP levels occurred in late February and the lower levels in May.

The assessment of plant basal area was set up in 15 paddocks where permanent sites were established to measure the change in the basal area of buffel and other plant species. The basal area in the first assessment varied between 1.5 and 5 m in the selected 10-m transects. No conclusions can be made at present on treatment effects on pasture.

Some paddocks were assessed to have a higher presence of weeds than others at the start of the project and were sprayed against broad-leaf weeds in the 2010 wet season (February – April) with Metsulfuron and 24-D. Spraying was carried out a few days after the paddock had been grazed to assist chemical contact with target weeds.

The total rainfall for the 2009–10 wet season was 1149 mm, which was about 50 mm below annual average. The number of rainy days above average for the season was 10, resulting 97 days of rainfall. The wet season did not start until mid November but finished late, receiving good rainfall in April and May. Rain in January and May was well above average, while in March it was well below average.

Reference:

Day, K. J. (1977). Fertility studies on three red earth soils of the Daly basin. Technical Bulletin No. 22, Department of the Northern Territory, Animal Industry and Agriculture Branch.

PROJECT: Helen Springs Producer Demonstration Site: Record keeping to Boost Production and Profit – Herd Segregation and Grazing Land Management on the Barkly

Project Officer: C. Duggan

Location: Barkly

Keyword(s): cattle, producer demonstration site, grazing land management

Objectives:

To increase the number of producers who collect objective information and apply it to decision making and property planning.

To enhance grazing land management practices in the Northern Territory (NT).

To improve herd performance.

To improve land condition.

To improve profitability.

To establish a successful producer demonstration site (PDS) at Helen Springs Station.

Background:

There is little appreciation in the NT's cattle industry of the value of record keeping for generating objective information to guide decision-making. Often it is considered too difficult to implement; or habits are too deeply-entrenched for its adoption. This makes it difficult to assess actual performance due to an inability to determine if animal and land resources are under or over-utilised.

Improved segregation and control of cattle can improve herd productivity, reduce labour costs and optimise carrying capacity and land condition. It also allows for more precise decision-making when combined with forage budgeting.

This project will attempt to demonstrate the ease with which information can be collected and incorporated into the decision-making process, leading to improved land use, herd performance and overall business success.

By highlighting the benefits of breeder segregation and linking it to sound grazing land management practices, it is hoped that producers in the region will appreciate the value of collecting objective information and applying it to decision-making and property planning.

The project has been reviewed and endorsed by the Barkly Research Advisory Committee. It has also been reviewed and supported by the producer group Barkly Landcare and Conservation Association. It is due for completion in 2012.

Method:

The project will collect both pasture and animal data at Helen Springs, which will then be analysed to examine the benefits of herd segregation, grazing land management principles and record keeping. Animal data, such as body

condition score, pregnancy status, foetal age, lactation status and weight is collected from all females in the segregation program. The data that Helen Springs regularly collects is not a specific requirement of the PDS. Pasture observations have been conducted on the 14 paddocks nominated for use in the segregation project. In each paddock, pasture yield was estimated every 500 m whilst driving through the paddock. As the project aims to demonstrate techniques that producers can utilise, easily-accessible roads were used to conduct pasture observations. Stocking rates for these paddocks have been calculated and once further information is obtained, more useful results will be published.

PROJECT: Senepol Crossbreeding

Project Officers: T. Schatz and T. Cowley

Location: Katherine

Keyword(s): cattle, Senepol, crossbreeding

Objectives:

To determine if crossbreeding with Senepol bulls is a viable strategy for producers with Bos indicus herds in the Northern Territory (NT) to produce animals that improve market options.

To compare the growth and meat quality of F1 Brahman x Senepol steers with that of pure Brahman steers.

To compare the performance of F1 Brahman x Senepol heifers with that of pure Brahman heifers over the heifer replacement phase (from weaning until the first calf is weaned).

To compare the performance of F1 Brahman x Senepol breeders with that of pure Brahman breeders.

Background:

The project will investigate if crossbreeding Senepol bulls with Brahman cows (current NT commercial genotypes) will produce offspring that perform well under NT conditions and have better meat quality than pure Brahmans. If that happens to be the case, the strategy would increase market options for NT cattle as they could be in demand in both the live export and Australian domestic markets.

The stressful environment in the northern parts of the NT (and also much of northern Australia) means that properties have to use cattle with a high *Bos indicus* (usually Brahman) content. However, Brahmans have a reputation for poor meat tenderness. While this has not been a problem in the live export markets, beef from high grade Brahman cattle is not as sought after in southern Australian domestic markets. During the last decade, the destination for nearly all cattle from the northern NT has been the Indonesian live export market. Having just one market makes properties with high grade Brahman cattle vulnerable should changes occur in that market. If the Indonesian live export market was to collapse and the high grade Brahman cattle from the NT were all sent to markets in southern Australia, it is likely that they would fetch lower prices due to perceived problems with their meat tenderness.

The ability to produce animals with better meat quality that is desired in several markets gives NT cattle producers more marketing options and more flexibility when market conditions change. They can then target whichever market gives the best returns at any point in time. This project has become even more relevant with recent developments in the live export market to Indonesia, which has meant that NT producers now have to find alternative markets for cull cows and heavier steers. Also, there has recently been renewed interest in re-opening

abattoirs in the NT; if this were to happen, it would be profitable for NT cattle producers to supply them with animals of good meat quality.

Considerable scientific research has shown that meat from *Bos taurus* cattle is more tender than meat from *Bos indicus* cattle (Crouse et al. 1995; Johnson et al. 1990; Sherbeck et al. 1995). Therefore, an obvious solution for high grade *Bos indicus* herds with a meat tenderness problem is to incorporate them with *Bos taurus* genes. However, this is not a simple process in northern areas of the NT, where pure British (*Bos taurus*) bulls often struggle just to survive in the harsh environment and their introduction in the past has largely been unsuccessful. To overcome these difficulties, it has been suggested to use tropically-adapted *Bos taurus* breeds, such as Senepol, which has been found to have good meat tenderness qualities (Olson 1999; Butts 1999). Its use could be an effective strategy to improve meat tenderness in tropical cattle. Another benefit of crossbreeding with Senepol is that all F1 offspring are polled or 'scurred', which avoids the need for de-horning.

The introduction of *Bos taurus* genes into Brahman herds in northern Australia has several potential advantages, such as hybrid vigour, potential for higher growth, better fertility and better meat quality; it also has some disadvantages, such as less resistance to pests and environmental stress, resulting in lower growth and fertility rates and higher mortality. This project is researching growth, fertility and meat tenderness in the progeny of F1 Brahman cows and Senepol bulls under NT conditions.

The Brahman x Senepol progeny will be compared with high grade Brahman cattle of the same age for the following parameters:

- Growth rate in steers in the post-weaning year on improved pasture in the Douglas Daly district.
- Steer performance in a feedlot.
- Meat quality and carcase measurements in steers at an Australian abattoir and meat science laboratory.
- Growth rate of replacement heifers up to two years of age on native pasture in the Victoria River District (VRD).
- Conception rates in maiden heifers.
- Re-conception rates in first-calf heifers.
- Breeder herd efficiency and other measures of performance in mature cows.
- Mortality rates.

The project will provide objective information to cattle producers in northern Australia who currently raise high-grade Brahman herds to assess whether crossbreeding Brahman cows with Senepol bulls will be a viable way of improving market options. The research should show whether mating Senepol bulls to Brahman cows will produce cattle that perform well under NT conditions and have improved meat tenderness compared with high-grade Brahmans. If that happens to be the case, the information would be useful for producers to:

- Produce cattle that have tender meat, which is in demand in several live export and domestic markets, thereby improve market options.
- Produce cattle with good tender meat for local abattoirs in the NT to reduce cattle sales to southern markets.

Method:

The research will comprise two phases. In the first phase, young F1 Senepol x Brahman animals will be bred and aspects of their performance up until three years of age will be studied. In the second phase, the performance of F1 Senepol x Brahman and high-grade Brahman cows will be compared when they are run together in the same paddocks and mated to Brahman bulls.

First phase (2009–2015)

- Build up female numbers for later breeder studies.
- Study growth and meat quality of male progeny at Douglas Daly Research Farm (DDRF) and feedlot.
- Compare Brahman to F1 Senepol x Brahman - replacement heifer phase at Victoria River Research Station (VRRS).

Second phase (2015–2018)

- Compare the performance of F1 Senepol x Brahman and Brahman breeders.
- Run both genotypes of cows together in breeder paddocks (see Appendix 1).
- Use Brahman bulls in all paddocks.
- Concurrently study breeder performance under supplementation.

Methodology of different aspects of the research
During the trial period, all cattle will be mustered at least twice a year for recording weight, fatness, height, lactation and pregnancy status (if applicable). Horn status and coat colour of F1 progeny will also be recorded.

Studies on male progeny (Brahman vs. F1 Senepol x Brahman)
After weaning, all male progeny will be transported to DDRF where they will be processed (castration, dehorning etc.) and post-weaning studies will be conducted on growth on improved pasture. Some steers will be selected for feedlot studies at Katherine where their growth rates and maturity type will be assessed. Some steers will be transported to a feedlot in Queensland where their growth will be studied and then detailed meat quality studies will be conducted at the University of New England's meat science laboratory.

Parameters for assessment will include:

- Growth rate on improved pasture over the year, post-weaning.
- Fatness (measured by ultrasound) at the end of the post-weaning year.
- Growth rate at a feedlot.
- Maturity type (fatness at known age and weight).
- Carcase and meat quality assessments (shear force etc.).

It will also be possible to compare the two genotypes of steers right through the live export process and their performance in an Indonesian feedlot.

Methodology of meat quality studies
After grazing improved pasture at DDRF for about one year post-weaning, 40 Brahman and 40 F1 Senepol x Brahman steers (selected to represent a range of sires) will be transported to a feedlot near an abattoir in Queensland where they will be fed for about 60 days. They will then be sent to an accredited abattoir where they will be slaughtered to measure carcase characteristics. After aging for seven days, the appropriate muscle from each carcase will be removed and sent to a meat testing laboratory for tenderness studies.

This procedure would be followed in animals from the first two-year groups of weaners and may be repeated with the third year group if funds are available and the information appears potentially useful.

Studies on female progeny (Brahman vs. F1 Senepol x Brahman) – the replacement heifer phase
After weaning, heifers from both genotypes will be placed in paddocks at VRRS with equal numbers of each genotype in each paddock. The heifers will be mated for the first time at two years of age (with a limited joining period of 3.5 months from late December to the end of March) and their growth up to that time will be compared together with conception rates from their maiden joining. Brahman bulls will be used with both genotypes of heifers, which will be running together mixed equally in paddocks stocked at the same rate (animal equivalents/km^2). All

heifers will be vaccinated against botulism and will receive a 7-in-1 vaccination at weaning and at the second round muster after weaning; a Vibrovax vaccination will be done at the second round muster prior to joining.

The performance of first-calf heifers will be studied (re-conception rate and calf loss) and again both genotypes will be mixed together in paddocks. Brahman bulls will be re-introduced in late December and removed at the first round weaning muster.

The following parameters will be assessed:

- Growth rate on native pasture over the post weaning year.
- Weight gain to maiden joining and fatness at this time.
- Conception rates (within weight ranges) from maiden joining.
- Calf loss and mortality in first-calf heifers.
- Re-conception rate in first-calf heifers.
- Weight and fatness at the time when first the calf is weaned.

Studies on breeders (Brahman vs. F1 Senepol x Brahman)
Heifers will be moved to breeder paddocks once they have had their first calf. Again each paddock will have an equal mix of cows of both genotypes. Mating with Brahman bulls will be for a limited period from late December to the first round muster.

The following parameters will be assessed:

- Breeder herd efficiency (kg of calf weaned/100 kg cow mated).
- Weaning rate.
- Wet cow re-conception rate.
- Mature size (weight corrected for stage of pregnancy at the same age, fatness and lactation status).
- Calf loss and mortality.
- Resistance to pests.
- Ability to maintain body condition (fatness) under VRD conditions.

In consultation with the Katherine Pastoral Industry Advisory Committee, further studies will be conducted if necessary on the progeny of the F1 breeders.

Results:

Senepol bulls were mated to Brahman cows during the 2008-09 wet season. The first crop of calves was weaned in May, 2010. All males were transported to DDRF to grow on improved pasture (where their growth will be studied) and all females are at VRRS. Another two-year group of weaners will be produced.

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Sherbeck, J. A., Tatum, J. D., Field, T. G., Morgan, J. B. and Smith, G. C. (1995). Feedlot performance, carcass traits, and palatability traits of Hereford and Hereford x Brahman steers. *Journal of Animal Science,* **73:** 3613-3620.

PROJECT: Selected Brahmans - Improving Brahman Fertility through the Use of BREEDPLAN EBVs and Selection

Project Officers: R. Golding, T. Schatz and T. Cowley

Location:	Katherine
Keyword(s):	Brahman, fertility, estimated breeding values, scrotal circumference, days to calve

Objectives:

To continue improving herd performance through selection, culling and superior genes by using AI and herd recording for BREEDPLAN.

To conduct extension activities to raise awareness of the use of selection and estimated breeding values (EBVs).

To continue the sale of sires and increase the sale of semen.

To compare herd performance with industry herds - requires an increased herd size for valid statistical analysis.

Background:

The growth and development of the Australian Brahman herd has been described as the greatest livestock revolution in history. The use of Brahman genetics has transformed northern Australian beef properties from near bankruptcy to efficient and profitable enterprises, contributing billions of dollars from domestic and export income. The use of such technologies as artificial insemination (AI), embryo transfer and BREEDPLAN, has increased the rate of genetic gain in Brahmans from being a crossbreeding option to the largest pure breed in Australia (Croaker 2002).

While the Brahman has made remarkable productivity gains in northern Australia through its adaptation to tropical environments, it is recognised to have a lower fertility than *Bos taurus* breeds. While some of this can be attributed to environmental stressors, which are present in the north, herd reproductive performance ultimately determines enterprise profitability and with the cost of production increasing, it is imperative that herds maximise outputs through higher weaning percentages.

Over the past 23 years, the NT Government has carried out research to improve the fertility of the Brahman breed. In 1994, a herd was formed using females from research stations and bulls from the local area. A high selection pressure was imposed. This involved mating heifers as yearlings, a strict culling policy for females more than two years, which failed to become pregnant and selecting bulls based on testicle size. The research also utilised AI to introduce high quality genes based on EBVs into the herd. The herd joined the Australian Brahman Breeders Association and became a member of BREEDPLAN in 1996. Herd data since 1986 was recorded. From 2005, all

male calves were kept entire and bulls for breeding were selected as yearlings. As a result, all females are now mated to either bulls bred from within the herd or AI is used (in about 10%).

The success of this research is indicated by the low average days to calving and high scrotal circumference (reproduction traits) EBVs for Herd 4299, which is the best of all the herds on Brahman Group BREEDPLAN (Figures 1 and 2).

The success of this program in a small herd over a 14-year period shows that very rapid improvements in fertility are possible in large NT herds if the selection methods are adopted. The current project continues following completed work with selection based on EBVs and herd performance, in addition to increasing herd size while maintaining strict selection, extending the knowledge of selection practices used and sharing the gene pool through bull and semen sales. It is necessary to build up herd size to allow for a proper statistical comparison with industry Brahman herds. This project will also provide examples of, and work closely with, the northern $election Index project, which was completed in 2009.

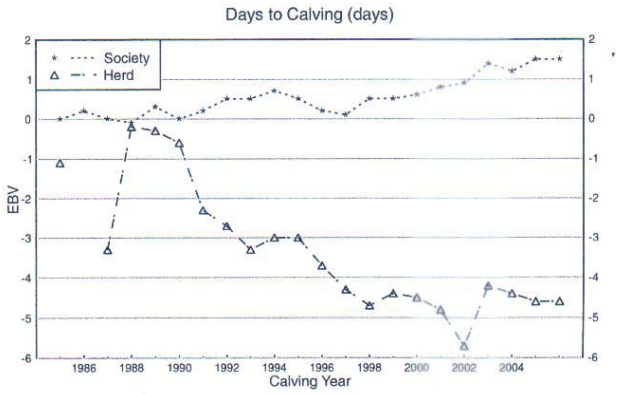


Figure 1. Days to calving EBV

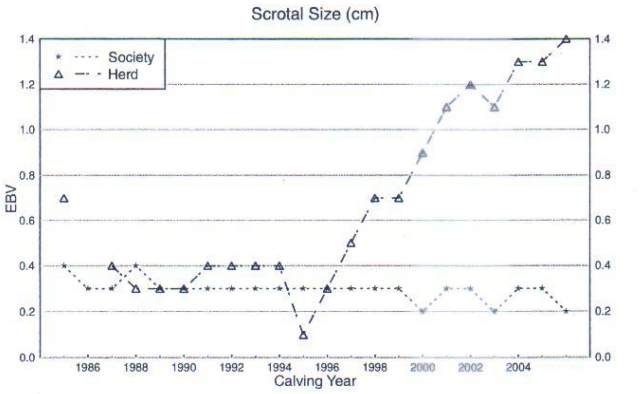


Figure 2. Scrotal size EBV

Method:

The selected Brahman herd has been based at Victoria River Research Station (VRRS) since 2002. Prior to that, the herd was located at the Douglas Daly Research Farm (DDRF). The replacement heifer phase still occurs at DDRF and the weaner heifers are transported back to DDRF shortly after weaning to grow and be mated there as yearlings.

Figure 3 illustrates the movement of cattle (females pink, males blue) in this trial and the data which is recorded and submitted to BREEDPLAN for national herd recording.

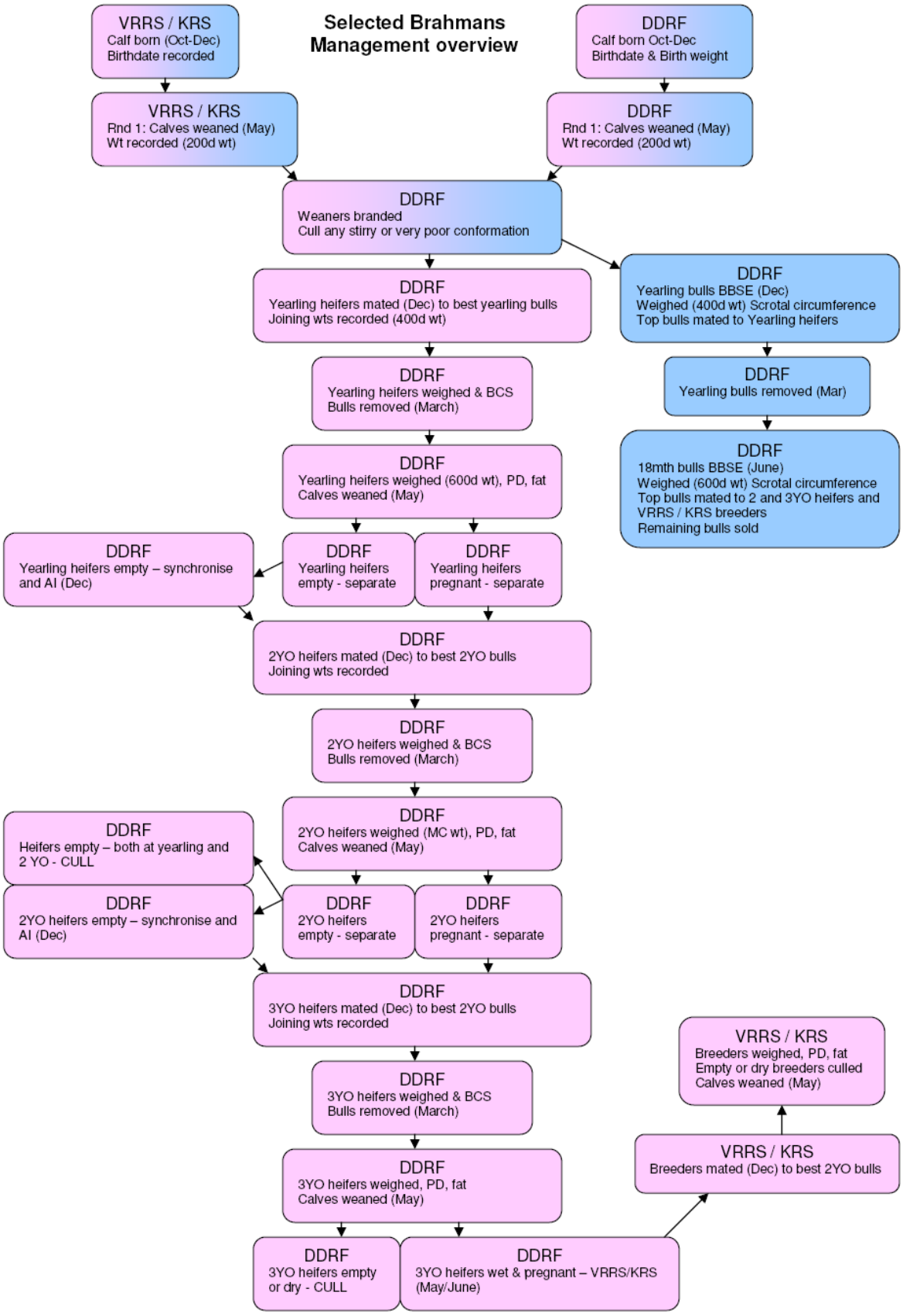


Figure 3. Selected Brahman herd management plan

The selection program in this herd involves the following:

- Cows were culled from the breeding herd if they did not raise a calf every year, although heifers that did not re-conceive following their first calving were not culled.
- Bulls bred within the herd were selected for use as yearlings on the basis of a selection index that placed emphasis on large scrotal size as yearlings, high 400-day weight, high percentages of normal sperm (at bull breeding soundness evaluation performed prior to their first joining as yearlings), as well as low age of their dam at calving and high dam "never miss a calf" scores.
- AI was used in maiden heifers to introduce different genes into the herd. Once EBVs for reproductive traits became available, the criteria used to select AI sires were low days to calving EBV and that they came from a herd which was known to cull non-pregnant cows each year.
- Heifers were mated for the first time as yearlings for a period of three months. At the start of the mating period, heifers were artificially inseminated once on the same day (following a synchronisation program with Crestar implants) and then bulls were introduced for the rest of the mating period. In years when sufficient numbers of heifers were pregnant from yearling mating, only pregnant heifers were kept as replacements

Results:

BREEDPLAN herd performance results were received after submitting the 2009-10 data. The results showed that herd fertility, measured by the "days to calving" and "scrotal size" EBVs, continued to improve compared with the BREEDPLAN group average (Figures 4 and 5).

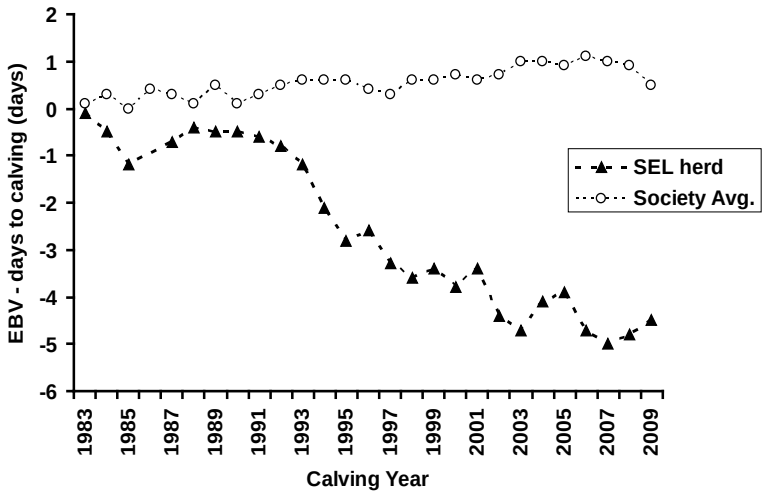


Figure 4. The change over time in days to calving EBV in the selected (SEL) herd and the Brahman breed society average

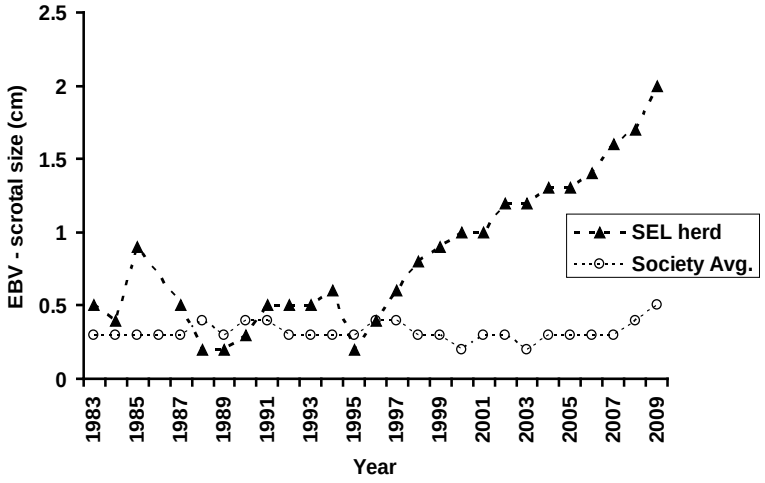


Figure 5. The change over time in scrotal size EBV in the SEL herd and the Brahman breed society average

Although the 2009-10 wet season started late and rainfall was patchy, pregnancy rates in the main SEL Brahman breeder herd at VRRS were high (75% in lactating cows) (Table 1). This is a very high pregnancy rate for lactating cows in these circumstances and is a result of the high fertility of the herd and management in a controlled mating system for a long period of time.

Table 1. Production figures for the SEL Brahman herd at VRRS

	Wet cows	Dry cows	Total
Number	72	9	81
Pregnancy rate (%)	75	89	77
Average weight (kg)	402.5	474.5	

It has been difficult to compare the fertility of the SEL Brahman herd with that of industry Brahman genotypes due to insufficient numbers for a proper statistical comparison and because the herd has been control-mated for many years while most commercial herds are continuously-mated.

However, in recent years, it has been possible to compare the fertility of the SEL Brahman herd with commercial Brahmans by comparing pregnancy rates of yearling mating of heifers from the SEL herd and from commercial (COM) herds at DDRF. More details of this work are available in Schatz et al. 2010.

Each year for three years, about 100 weaner heifers were purchased from a commercial property (a different property each year) and transported to DDRF shortly after weaning. Weaner heifers from the SEL herd at VRRS were also transported to DDRF and both groups of heifers were mated as yearlings from late December until the end of March.

Pregnancy rates were significantly higher each year in SEL heifers (Figure 6) and over the three years, pregnancy rates in SEL heifers were, on average, 35% higher. Note that only heifers in the same pre-joining weight ranges were compared with each other and that in each year the average weight of COM heifers was higher than the average weight of SEL heifers, although these differences were not significantly different.

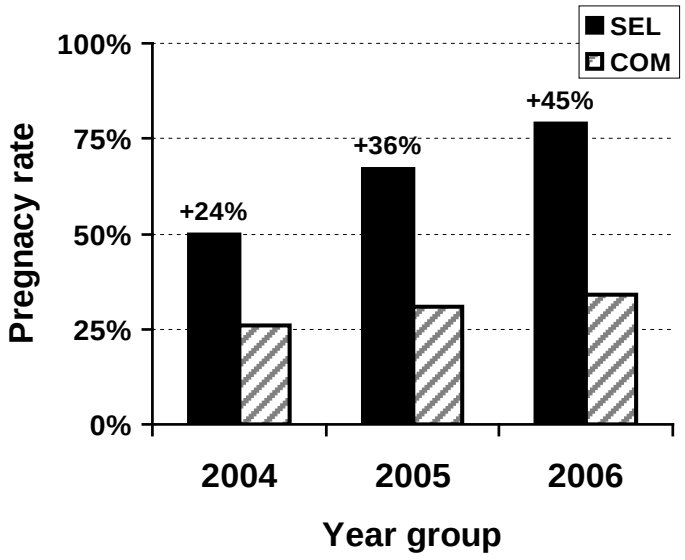


Figure 6. Pregnancy rates in yearling mating of heifers from SEL and COM

When heifers do not conceive from yearling mating, it is likely to be due to not having reached puberty by the end of the mating period. Brahmans are considered to be among the later-maturing breeds and in a recent study in northern Australia, the Beef CRC found that the average age at puberty of Brahman heifers was 750 days. Therefore, it is not surprising that this study found low pregnancy rates in COM Brahman heifers from yearling mating. It is likely that the reason for the significantly higher pregnancy rates in SEL heifers was lower average age at puberty and that more had become fertile before the end of a three-month mating period as yearlings.

A number of studies have shown that a lower heifer age at puberty is associated with improved fertility at later ages; yearling mating is seen as a way of identifying heifers that are inherently more fertile and will produce more calves over their lifetime. The results from this study show that SEL heifers were more fertile as yearlings than COM heifers, which implies that they were inherently more fertile. This suggests that the selection program that has been imposed on the herd that produced SEL heifers has been successful in improving their fertility. This is supported by data in Figures 4 and 5, which shows that two BREEDPLAN measures of fertility (EBVs for days to calving and scrotal size) for this herd have improved markedly over time relative to the average of the Brahman Breed Society.

These results show that improvements in Brahman fertility can be made through selection in northern Australia. This has substantial implications for the northern beef industry where cattle require a high *Bos indicus* content to perform under the stressful conditions. Reproduction is the most important trait affecting the profitability of beef enterprises, especially in tropical environments where reproductive rates are generally lower. Therefore, improving the fertility of Brahman cattle through selection has the potential to increase the profitability of cattle properties in northern Australia.

Reference:

Schatz, T. J., Jayawardhana, G. A., Golding, R. and Hearnden, M. N. (2010). Selection for fertility traits in Brahmans increases heifer pregnancy rates from yearling mating. *Animal Production Science,* **50:** 345-348.

PROJECT: NT Live-weight Gain Project

Project Officers: S. Streeter and T. Cowley

Location: NT

Keyword(s): cattle, live-weight gain

Objectives:

To analyse data from Beef CRC herds and stud herds from two major pastoral companies in northern Australia to determine the amount of live-weight gain variation in growing animals that can be attributed to genetic and environmental influences.

To identify and quantify the drivers of live-weight gain variation within and between ten groups of cattle on commercial properties in the Northern Territory (NT).

To identify the influence on live-weight gain variation of other difficult-to-measure causal factors such as foraging behaviour, parasites and some disease factors from a series of smaller scale nested experiments.

To report the potential differences in feed efficiency factors between high and low growth animals identified above in pen studies at the Katherine Research Station (KRS).

To develop a practical analytical toolkit and determine data requirements for investigating and identifying the drivers of live-weight gain performance in individual herds.

To develop strategies that can be identified using an analytical toolkit to reduce the number of poor performing animals and increase average herd performance.

Background:

It is proposed that the most cost-effective strategy for improving live-weight gain in cattle in the NT and many other extensive areas of northern Australia would be to address the large variation in performance within the herd and focus on lifting production in the poorly performing proportion of the herd. Measurements taken in extensively managed herds in the NT show an extremely wide variation in the rate of live-weight gain even within herds managed under the same conditions and containing animals without obvious breed differences. This variation represents an opportunity for significant improvement.

Method:

The project consists of three experimental phases.

1. Desktop study – analysis of Beef CRC and NT industry stud data (completed May 2008).
2. Field study – investigation of causal factors affecting live-weight gain on ten commercial NT herds (Barkly and Katherine regions) (current).
3. Nested studies – research station pen trials investigating feed efficiency and other difficult-to-measure factors contributing to live-weight gain variation measured in the extensive environment in Phase 2 (2009-10).

Field study

Data is collected from a single group of 250 steers on each participating property, from weaning to approximately 12 months post-weaning. Four observation events occur over this period: (1) weaning/processing, (2) two to three weeks post-weaning, (3) pre-wet season and (4) post-wet season. Table 1 lists the project variables to be measured at the animal level, group level and property level.

Table 1. Project variables described at the animal level, group level and property level

Variable category	Attribute	Variable	Units	Recording frequency
Animal-level	Identification	FID	3 digit number	Once
		EID	15 digit number	
	Sex	Sex	Single code digit	Once
	Branding year	Year brand	4 digit year number	Once
	Breed	Breed content	4 digit text	Once
	Observation Date	Date	date	Four times*
	Weight	Weight	kg	Four times*
		ADG	kg/day	Three gain periods
	Condition	Condition score	1 to 9	Four times*
	Maturity type	Hip height	cm	Four times*
		Fat depth	mm	post-wet season
	Genetics	Sire ID	6 digit number	Once
		Sire breed	4 digit text	Once
	Temperament	Flight speed	m/second	Four times*
	Tick	Tick score	1 to 5	Four times*
	Fly	Buffalo fly count	count	Four times*
	Lesion	Buffalo fly lesion score	1.1-2.5	Four times*
	Diet selection (FNIRS)	Faecal nitrogen	%	Three times
		Forage digestibility	%	
		ME intake	(MJ/100kgLW)	
		Dietary non-grass	%	
	Dehorn severity	Wound size	(cm^2)	Weaning
		Frontal sinus exposed	y/n	
		Bleeding rate score	0-3	
		Dressing applied	y/n	
		Tool	S,C,K,P	
		Hygiene score	0-3	
	Castration severity	Method (cut low, cut high)	L,H	Weaning
		Bleeding rate score	0-3	
	Dehorn infection	Healing score	0-5	Two weeks post-weaning
	Castration infection	Healing score	0-5	Two weeks post-weaning

Variable category	Attribute	Variable	Units	Recording frequency
Mob-level	Paddock	Paddock ID	3 digit text	Once
		Paddock location	latitude/longitude	
		Watering points	latitude/longitude	
	Climate	Rainfall	mm	Daily
		Average temp	ºC	Monthly
		Average relative humidity	%	
		Weather station ID	text	One-off
	Utilisation rate	Paddock size	Hectares	Once
		Total grazing area	Hectares	
		Stocking rate	AEs/ha	As required
		Total standing dry matter	kg/ha	Early dry season
		Pasture species composition		
	Diet quality (FNIRS)	Faecal nitrogen	%	Monthly
		Forage digestibility		
		ME intake	(MJ/100kgLW)	
		Dietary non-grass		
	Supplement	Feeding rate	(g/day)	As required
		Supp nitrogen	(g/day)	
		Supp phosphorus	(g/day)	
	Internal parasites	FEC	EPG	Four times*
		FOC	OPG	
		Species	%	
Property-level	Property name	Property ID	3 digit text	Once
	Management structure	Management structure	text	

*Four times: weaning, two to three weeks post-weaning, pre- wet season, post-wet season

Results:

Data collection for this project has not been completed yet. Some preliminary results are presented here.

Data collection for Stage 2 has been completed for “A” herds (Lakefield, Mainoru, Hayfield, Brunette Downs and Walhallow). Analysis has begun on “A” herd data. Average daily gain data for “A” herds has been summarised in Table 2.

Table 2. Summary of average daily gain (ADG) for “A” herds

Property (Region)	Mean weaning weight (kg) ± s.d. (n)	Mean dry ADG (kg/day) ± s.d. (n)	Mean wet ADG (kg/day) ± s.d. (n)	Mean annual ADG (kg/day) ± s.d. (n)
Hayfield (Katherine)	211 ± 44 (252)	0.06 ±0.12 (236)	0.28 ± 0.07 (221)	0.20 ±0.06 (226)
Lakefield (Katherine)	134 ± 23 (222)	0.05 ± 0.06 (208)	0.46 ±0.11 (193)	0.18 ± 0.05 (195)
Mainoru (Katherine)	160 ± 30 (285)	-0.04 ± 0.09 (179)	-	-
Walhallow (Barkly)	213 ± 36 (250)	-0.06 ± 0.11 (227)	0.55 ± 0.13 (206)	0.26 ± 0.06 (215)
Brunette (Barkly)	214 ± 50 (227)	0.31 ± 0.13 (216)	0.44 ± 0.10 (208)	0.39 ± 0.07 (211)

s.d. = standard deviation, n = sample size

Data collection for observation events 1 and 2 was completed on all 2010 properties (Legune, Helen Springs, Finniss River, Wyworrie and Wavehill), which consist of "B" herds, and will finish in round 1, in 2011. The weight data collected so far is summarised in Table 3. Outlier data has been eliminated from the summary analyses. Three stations presented animals that had been branded as calves. Hence observation 2 was not carried out and the impact of dehorning and castration could not be assessed.

Table 3. Summary of weaning weight, post-weaning weight and ADG for "B" herds

Property (Region)	Mean weaning weight (kg) ± s.d. (n)	Mean post-weaning weight (kg) ± s.d. (n)	Days	Mean ADG (kg/day) ± s.d. (n)
Legune	171	172	15	0.07
(Katherine)	±39 (202)	±26 (206)		±0.81 (202)
Wyworrie	176	180	24	0.17
(Katherine)	±36 (179)	±38 (180)		±0.33 (179)
Helen Springs	184			
(Barkly)	±29 (250)			
Wavehill	219			
(Katherine)	±38 (239)			
Finniss River	225			
(Katherine)	±35 (186)			

s.d. = standard deviation, n = sample size

Wyworrie Station presented a mix of branded and unbranded animals. Presumably the branded weaners were older and hence heavier at weaning. The difference in post-weaning ADG is shown in Table 4.

Table 4. Differences in post-weaning ADG for branded and clean-skin animals at Wyworrie Station

Branded status	Number	Weaning weight (kg)	Post weaning ADG (kg/day)
Branded	82	195	0.40
Clean skin	97	159	-0.02

Nested study - pen trial

A pen trial has begun at KRS to identify the relationships between insulin-like growth factor-1 and metabolites associated with growth and nutrient status of animals and post-weaning live-weight gain in cattle grazing low and high crude protein pastures. The trial will run for about 80 days and will be reported by March 2011. Seventy-two weaner males were selected based on their dry season weight gain from Lakefield Station (36 low growers and 36 high growers paired for weaning weight). There are two crude protein diets, low growth diet (Mekhong grass) and high growth diet (Cavalcade). The design is diet x ADG group (high vs. low grower), with six replicates. The weight differences after two weeks in the pen study are summarised in Table 5.

Table 5. Weight gain differences of the treatment groups in the KRS pen trial

Treatment group	Weight gain (kg)
High growers x Cavalcade	6.7
High growers x mekhong	4.9
Low growers x Cavalcade	7.8
Low growers x mekhong	4.2

The project is scheduled to finish in March 2012. More detailed reports will be produced in future. This project is funded by Meat and Livestock Australia.

PROJECT: Northern Australian Beef Fertility Project – Cash Cow

Project Officers: **M. McGowan[1], K. McCosker[2], G. Fordyce[3], B. Burns[3], D. Smith[3], T. Newsome[4], D. Menzies[4], P. O'Rourke[5], N. Perkins[6] and S. Jephcott[7]**

[1]The University of Queensland, School of Veterinary Science; [2]NTDoR; [3]QDEEDI; [4]Outcross Performance; [5]Queensland Institute of Medical Research; [6]Ausvet, Toowoomba; and [7]Chinchilla Veterinary Services

Location: NT

Keyword(s): cattle, fertility, peri-natal loss, cash cow

Objectives:

To define reproductive performance in a selected population of northern Australian commercial properties (study population) over three consecutive years using a range of measures.

To establish outcome measures for monitoring and comparing the reproductive performance of breeding 'mobs' and properties in northern Australia.

To define typical and achievable performance using the above measures in the study population.

To estimate variation in reproductive performance at the animal, 'mob', property and region level.

To identify causes of variation in the reproductive performance between animals, 'mobs', properties and regions.

To quantify the proportion of variation explained by identified risk factors.

To identify those risk factors which explain the greatest amount of the variation between 'mobs', properties and regions.

To develop a cost-benefit framework to estimate the economics of changing the major mob-level factors affecting reproductive performance.

To recommend a cost-benefit study to assess the impact on production of changing well-defined inputs and management practices that affect key risk factors.

To recommend extension priorities for changing well-defined inputs and management practices that affect key risk factors.

To recommend research priorities for inputs and management practices that affect key risk factors for which the impacts are not well defined.

To recommend the feasibility of establishing strategic ongoing reproductive performance monitoring 'systems' to enable the longitudinal evaluation of the impact of implemented changes in management practices and inputs.

Background and Method:

Please refer to the Annual Research Report 2008-09.

Results:

A summary of the collected raw data in 2008-09 is provided in Table 1. It should only be considered as preliminary results.

Table 1. Pregnancy rates in joiner heifers

Region	No. of mobs	Range (%)	3/4 of mobs achieved (%)	1/2 of mobs achieved (%)	1/4 of mobs achieved (%)
South Qld	18	39-100	72	84	93
North Qld	17	9-93	39	69	85
NT/WA	3	29-70	29	43	70
Total	38	7-100	48	78	88

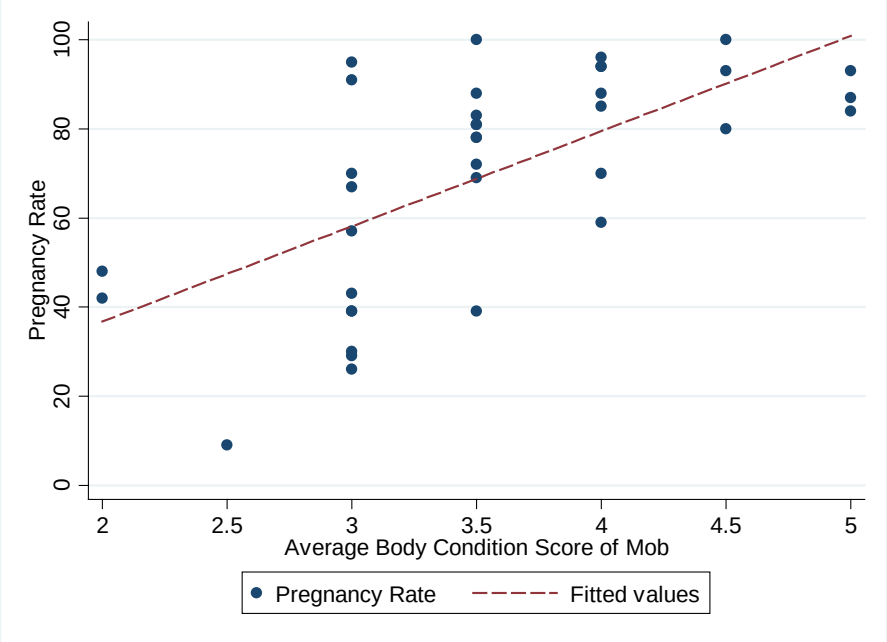


Figure 1. The effect of body condition score on pregnancy rates

Table 2. Pregnancy rates in breeders in 2009

Region	No. of mobs	Range (%)	3/4 of mobs achieved (%)	1/2 of mobs achieved (%)	1/4 of mobs achieved (%)
South Qld	50	38-100	83	90	95
North Qld	27	7-94	46	67	80
NT/WA	18	28-92	39	47	61
Total	95	7-100	56	80	92

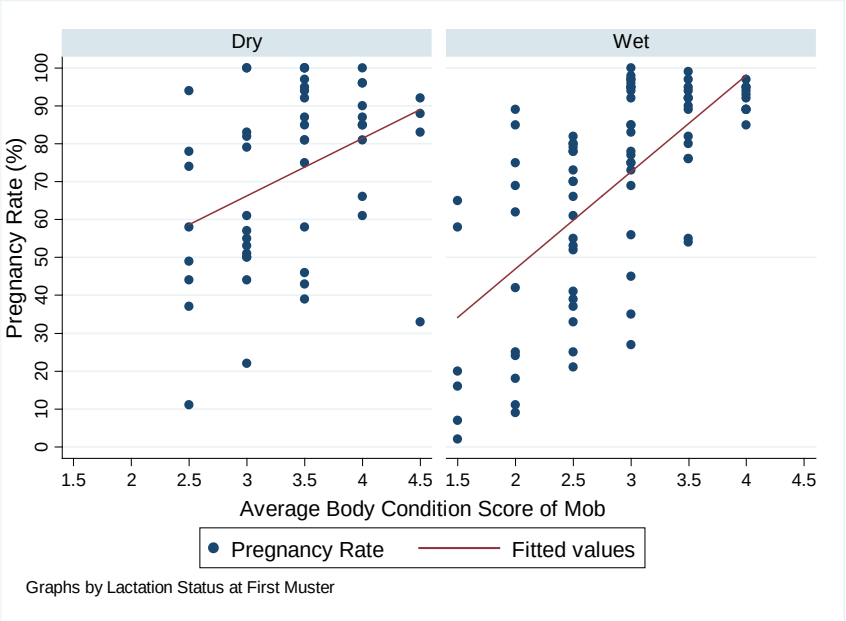


Figure 2. How body condition in breeders at first muster affects the likelihood of pregnancy by the second muster

Table 3. Pregnancy rates in first calf heifers in 2009

Region	No. of mobs	3/4 of mobs achieved (%)	1/2 of mobs achieved (%)	1/4 of mobs achieved (%)	Range (%)
North Qld	2	33	54	75	33-75
NT/WA	2	79	81	83	79-83
South Qld	10	81	86	88	64-93
Total	14	75	83	88	33-93

Table 4. Reproductive measures for first calf heifers

Reproductive measure	No. of mobs	Typical (median) mob	Range
Annual weaning rate	17	87%	61-96%
Annual weaner weight turned-off per breeder	12	172 kg	102-205 kg
Annual pregnancy rate	15	83%	26-93%
Losses between confirmed pregnancy and weaning	17	9%	0-23%

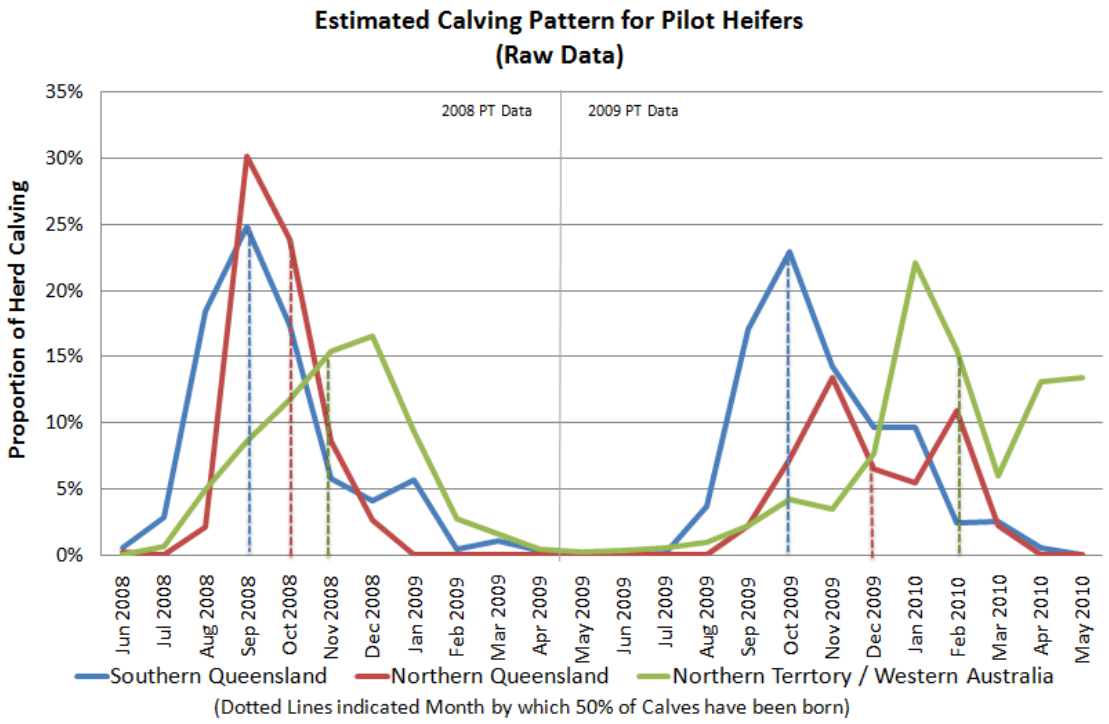


Figure 3. Predicted calving patterns using foetal age data grouped for the Southern, Northern and NT/WA regions. Dotted lines indicate the month when 50% of pregnancies were predicted to have reached full term.

PROJECT: Pasture Species Evaluation under Grazing at DDRF

Project Officers: P. Shotton, B. Lemcke and DDRF Staff

Location: Darwin

Keyword(s): improved pastures, pasture grasses, pasture legumes, cattle grazing, leucaena

Objectives:

To evaluate improved pasture species and mixtures under a continuous grazing regime on Blain soil at Douglas Daly Research Farm (DDRF).

To determine their persistence, productivity and contribution to weight gain performance in cattle.

To make pasture management recommendations for Top End livestock producers.

Background:

Promising pasture species and mixtures are evaluated under grazing by cattle at DDRF to determine their long-term potential in the Douglas Daly and other Top End regions of the Northern Territory.

Method:

Pastures are grazed in 4-ha paddocks by six Brahman weaner steers or heifers per paddock (1.5 animals/ha). This year only four of the latest established paddocks were used in the evaluation. Weaner heifers were allotted to paddocks on 15 September and remained in the grazing trial until the following June of 2010.

No top dressing fertiliser was applied to the paddocks this season.

During the wet season, paddocks 41, 43 and 51 were boom-sprayed with Metsulfuron plus 24-D for broad-leaf weed control. Paddock 42 was not sprayed so as to maintain the legume component of the pasture.

In past years, the animals were supplemented with Uramol® blocks ad-lib during the dry season and with Phosrite® blocks in the wet season; however, this year no supplements were given.

The cattle were weighed every second month to record live-weight changes and body condition scores. On 30 December, the cattle in paddocks 41 and 43 were moved into a nearby buffel grass paddock because of poor pasture regrowth and declining animal condition. These paddocks were rested and then re-stocked on 09/02/2010.

Pasture composition and yield were assessed twice during the year, first in the early wet season in December 2009 and then in June 2010.

Results:

The cattle allocated to paddocks 41 and 43 performed poorly before the end of the year. However, after the 40- day rest period, animals in the two paddocks grew well making good live-weight gains. The cattle in those paddocks grazed buffel grass and gained 0.4 kg/animal/day while animals in paddocks 42 and 51 gained 0.5 and 0.7 kg/animal/day, respectively over the same period. Although animals in paddock 51 had the highest live-weight gain overall and animals in paddock 41 had the lowest, the cattle from the two rested paddocks recovered well. It would have been interesting to see the results if the four paddocks were equally rested for the same period.

Table 1. Average cattle live-weight gains for each paddock (kg/animal/day)

Paddock	End of dry season 15/09/09 to 17/12/09 over 95 days	Early wet season 17/12/09 to 10/02/10 over 65 days	Mid wet season 10/02/10 to 12/03/10 over 30 days	Last two months 08/04/10 to 11/06/10 over 64 days	Total time 15/09/09 to 11/06/10 over 270 days
41	-0.14	0.36*	1.10	0.68	0.39*
42	-0.06	0.52	0.80	0.69	0.40
43	-0.15	0.39*	1.10	0.92	0.44*
51	-0.04	0.66	0.80	0.61	0.45

* Paddocks rested between 30/12/09 and 10/02/10

When live-weight gains are compared with past years' averages, the performance this year was well below the average in all four paddocks. This was probably due to the later start to grazing, the late start to the wet season and not supplying fertiliser or supplementary lick blocks thought the year.

Table 2. Average cattle live-weight gains from previous years (all paddocks)

Paddock	**41**	**42**	**43**	**51**
Paddock species	Tully /Wynn	Jarra / Wynn	Signal	Strickland /Wynn +
Past years av. wt (kg)	128	173	164	184
2009 - 2010 av. wt (kg)	105	108	118	121

Pasture composition and total yield were assessed in December 2009 and in June 2010. Paddock 41 had a large increase in Wynn cassia (now 45%) compared with last year's assessment of 15%. This has also reduced the proportion of Tully grass from 82% to 46%. Other grasses were mainly sabi and summer grasses; the main weeds were Sida acuta and Senna. The total biomass of the pasture was 5.3 t/ha (June 2010).

Paddock 42 also had increased Wynn cassia (from 20% to 30%) and less Jarra grass (77% to 60%); sabi was the other main grass species. The main weeds were Sida acuta and Senna. The pasture has a reasonable grass to legume ratio and despite no herbicide applications, has a relatively low weed infestation. The total biomass of the pasture was 6.8 t/ha (June 2010).

Paddock 43 consists mainly of Signal grass (75%) and other grasses (21%) sabi, summer grasses and button grass. Compared with last year, there was an increase in other grasses (5% to 21 %) and a decrease in Signal grass (95% to 75%). Prior to herbicide spraying in February, the paddock had a high population of broad-leaf weeds, mainly Senna with some Sida acuta. The herbicide application worked well, reducing weed numbers to a minimum. The total biomass of the pasture in June 2010 was 7.2 t/ha.

Paddock 51 consists mainly of Strickland Finger grass (81%), Wynn (*Chamaechrista* spp (16%) and other grasses, mainly sabi (2%). Broad-leaf weeds were less than 1%. The total biomass of the pasture in June 2010 was 9.5 t/ha. The pasture appeared to be selectively grazed with cattle targeting the shorter fresh growth and leaving the large thick tussocks of Strickland. A heavier stocking rate or "crash grazing" may have improved the performance of this paddock.

Figure 1 shows the average percent of the main grass species, all other grass species, main legumes and all broad-leaf weeds from the four paddocks during June.

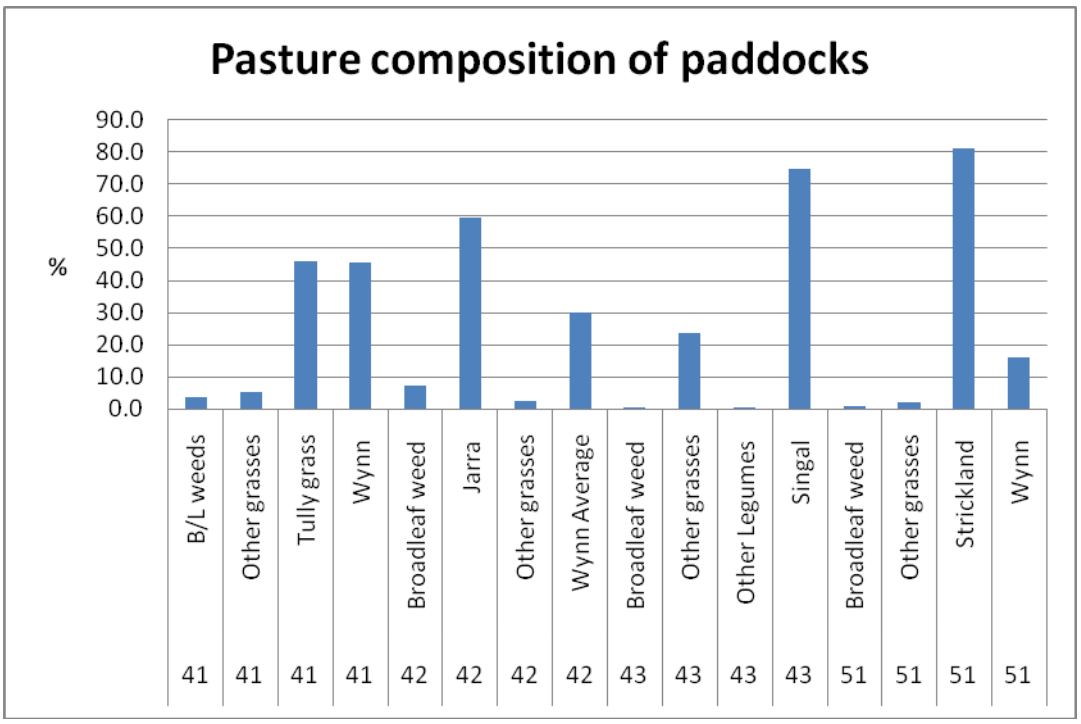


Figure 1. The proportion of plant groups in each paddock June 2010

Rainfall

The total rainfall for the 2009–10 wet season was 1149 mm, which was about 50 mm below the annual average. There were 97 rainy days during the season, 10 days above average.

The wet season did not start until mid November, but finished late, receiving good rainfall in April and May. Rain in January and May was well above average while in March it was well below average.

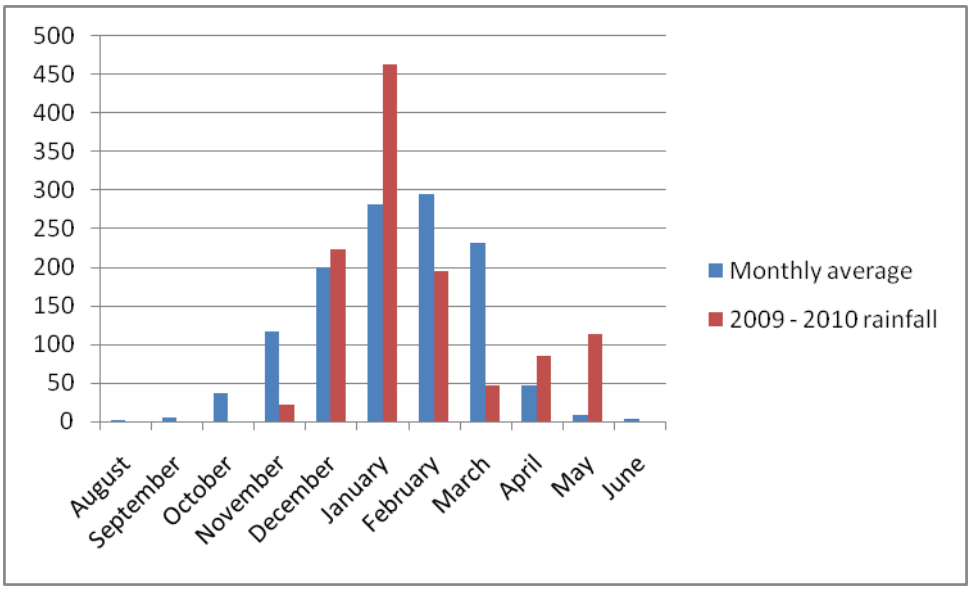


Figure 2. DDRF rainfall (mm)

Summary
Pasture grazing trials at DDRF were first established in the 1970s to assess the productivity of different pasture species and mixtures under continuous grazing to determine their long term potential in the Douglas Daly and other Top End regions. A report on the trial's results and outcomes is due for completion in November 2010.

PROJECT: Enhancing Productivity Improvements in the Water Buffalo Industry

Project Officers: B. Lemcke, E. Cox, A. Turner, M. Humphries and BHF Staff

Location: Darwin

Keyword(s): buffalo, breeding, artificial insemination, genetic analysis, buffalo register

Objectives:

To continue to develop artificial insemination (AI) synchronisation methods to achieve 50% conception rates from single inseminations.

To establish benchmark tenderness values for TenderBuff of various age/weight ranges.

To determine the quality factors of buffalo milk that set it apart from cow milk.

To continue to subsidise milk testing at the Millaa Millaa dairy to promote data gathering in a commercial herd.

To encourage the registration of buffalo producer herds in Australia

To monitor the performance of crossbred river buffalo at feedlots overseas compared with swamp buffalo.

To prepare a "Best Practice Manual for Water Buffalo in Australia".

Background:

The Department of Resources commenced a four-year RIRDC project (DNT 33A) in 2003. The results were reported in RIRDC Publication No. 08/189 "Australian Water Buffalo: Genetic and Reproduction Improvements". More funding was approved for the following four years to continue the study.

Method:

As suitable females become available, drug protocols are used to synchronise the onset of oestrus for fixed time inseminations. Italian and local semen is used to produce suitable genetic lines to improve productivity in both meat and milk production. Due to a limited number of imported bulls, AI is used. Northern Territory (NT) and interstate herds can participate in this project if they have suitable stock and facilities.

Comparisons between crossbred and swamp buffalo feed conversion efficiencies are planned locally and in overseas feedlots to demonstrate the value of crossbreds for meat production.

Milk testing for quality will be carried out to enhance marketing of buffalo dairy products.

Producers will be encouraged to participate in BREEDPLAN.

If abattoir facilities become available in the NT, TenderBuff studies will be continued to improve meat tenderness in different weight/age groups.

Contacts will be maintained with research and development organisations elsewhere.

Results:

AI was carried out in March, June and October, 2009 at Beatrice Hill Farm (BHF). The Ovsynch protocol was used in March and June with mixed results. In March seven out of 16 cows (no heifers) became pregnant, subsequently delivering seven live calves that were reared to weaning. In June, semen from COFA in Italy was used resulting for the first time in more than 50% pregnancies with the Ovsynch protocol. Out of 23 cows and three heifers, 15 cows and one heifer became pregnant resulting in 14 live calves in April/May 2010. Out of four recent first-calf producers, only one became pregnant. The calves have not been weaned yet. One pregnant 10 year-old cow was found dead due unknown causes just before calving.

Table 1. June 2010, Round 16

	Total count	**Conceived**	**Conception rate (%)**
Total group	26	15	57.7
Total cows	23	14	60.9
Total heifers	3	1	33
Wet cows	18	11	61
Dry cows	5	3	60
Wet cows with calves < 6 weeks	4	0	0

The results indicate a significant breakthrough in reproductive performance from the use of AI and appear to be entirely due to better quality semen now available.

In October, two protocols were used on 11 remaining cows and two heifers: seven on the out-of-season Cue Mate and six on the Ovsynch method.

Table 2. October 2010, Round 17

	Total Nos.	**Wet (W) /dry (D)/heifer (H)**	**Conceptions W/D/H**	**(%)**
Cue Mate	7	4/2/1	3/1/1	71.4
Ovsynch	6	3/2/1	0/1/0	16.7
Total group	13	7/4/2	6	46.2

The results demonstrate the usefulness of the “out-of-season” protocol at a time when climate, cow condition and day-length act against good conception rates.

It is hoped that a test run of 20 animals of two age groups (12 months difference) will be slaughtered at Oenpelli abattoir in the 2010 dry season to test for tenderness.

Eight milk tests were carried out at Millaa Millaa in 2009-10.

Table 3. Milk test results in 2009-10

	No. cows milked	Milk/cow (L)	Protein (%)	Fat (%)	Somatic cell count	Mean lactation (days)
Mean	53	5.1	5.06	9.23	52.7	103.8
Range	40-70	3.0-6.7	4.9-5.4	8.7-9.6	33-73	94-157

Barry Lemcke attended the 9th World Buffalo Congress in Argentina and presented a paper by him and M. Suarez entitled "Production parameters from different breeds of water buffalo in Australia", which was published in the Proceedings of the Congress *Revista Veterinaria,* Volume 21, Supp 1, April 2010.

PROJECT: Riverine Buffalo and Crossbreeding

Project Officers: B. Lemcke, E. Cox, L Huth, A. Turner, G. Jayawardhana and BHF Staff

Location: Darwin

Keyword(s): buffalo, breeding, swamp, riverine, crossbreeding

Objectives:

To determine the merits of crossbreeding and upgrading to riverine buffalo for the Northern Territory buffalo industry.

To distribute suitable progeny from the project to the industry for breeding or for supplying the TenderBuff or dairy trade.

To demonstrate sustainable buffalo production systems.

Background:

The aim is to produce purebred riverine buffalo from two directions to maximise and accelerate the process by using purebred cows to increase numbers from within the herd and by crossbreeding with swamp buffalo and then backcross to purebred through ¾, ⅞ and 15⁄16 generations back to purebred riverine.

It is expected that during the life of the project it will be possible to identify all mixtures of the two breeds that will best suit the meat, live export and dairy produce markets in Australia and overseas.

A four-year cooperative agreement was finalised in early 2007 with the Australian Dairy Buffalo Co in Millaa Millaa (north Queensland) to supply quantitative and qualitative data on the milk producing potential of the various crosses compared with the pure riverine buffalo. Most of that farm's stock was derived from the Beatrice Hill Farm (BHF) herd. The dairy supplies milk to the Vanella Cheese Factory in Cairns, which produces mozzarella, fetta, buffalino, yoghurt, and other products through its own outlets. Milk data is being incorporated in the genetic database and the Buffalo BREEDPLAN.

BHF also supplied stock to another dairy farmer in Maleny (Qld) in 2004, 2005 and 2008. That farm is now supplying milk to the local Maleny Cheese Factory, which is selling a range of buffalo cheese and yoghurts.

Heifers from BHF are also supplied to a local Darwin dairy owner and cheese maker to demonstrate the viability of dairy farming and cheese making in a wet/dry tropical environment.

Method:

All NT Government stock is held at BHF. Table 1 shows the bulls used in 2009.

Table 1. Bulls used during the 2009 mating season

Cow group	Bull(s) used in 2009	In 2010 (bull number)
Swamp	Sold to private interests	5861
F1	Sold to private interests	5861
3/4	5861	5860
7/8	5858	5880
15/16	5859	5879
River	Three rounds of AI in March 09, June 09 and October 2009	One round of AI in July 2010
All heifers	5875	5879 – purebred heifers from 20/04/2010; 5880- Xbreds from 01/06/2010; 5860 - Agisted heifers 20/04/2010

Since 2007, all yearling heifers have been mated separately and continuously rather than control-mated. This allows for earlier calving in heifers at two years of age instead three. Also, this was done because it is more difficult to use AI in heifers and because they are well grown as yearlings to cope with pregnancy. If left until the age of two years before mating, over-fatness may become an issue of concern at higher weights, particularly at calving.

Some Italian milking buffalo semen was imported for use in AI. Italian semen from COFA was imported in April, 2009. Nine new bulls are available.

Results:

Table 2. The composition of riverine, swamp and crossbred buffalo groups at BHF, June 2010

	Imported bulls	Locally bred bulls	Breeder cows	Yearling bulls	Yearling heifers	Male calves live	Female calves live	Calf deaths	Total
Purebred riverine	2	18	40	15	14	20	15	1	124
3/4			41		0	0	0	0	41
7/8			34		12	11	12	2	71
15/16			15		17	8	13	7	60
Total		18	130	15	43	39	40	10	296

Table 3. Calving during 2009-10 and pregnancy diagnosis (June 2010) for next season's calves

Breeder group	Calves born/cows mated = Calving rate (%) 09-10 calves	No. pregnant June 2010/ cows mated January 2010	Pregnancy (%)	No. pregnant + wet cows/ total wet cows = (%) wet cows pregnant
¾ cows + heifers	25/40 = 82.4	22/39	56.4	16/31 = 51.6
7/8 cows + heifers	28/40 = 56.4	14/34	41.2	5/19 = 26.3
15/16 cows + heifers	14/16 = 81	4/15	26.7	3/13 = 23
Riverine cows	22/35= 62.9 by AI	Round 1 6/13 Oct 09; Round 2 14/29 Jul 10	62.5 by AI	11/27 = 40.7
Total	89/131 = 67.9	60/123	48.8	35/90 = 38.9

Discussion:

There was again a significant loss of calves this year at 11.2%, up from an average of 8.8% over the last four years. The highest rate in that time was 12.8%, four years ago. This was mainly due to dingoes/wild dogs and mismothering by heifers. As a result, this coming season heifers will be kept separate from the rest of the breeder groups until they have weaned their first calves. The heifer calving group will be closely observed throughout the calving period and will be deliberately kept in an open nearby paddock for easy supervision against dingoes and for detecting mismothering.

Pregnancy from this year's mating has declined significantly, averaging over the whole herd at just less than 50%. This was unexpected, considering previous history. One observation is that the two worst producing groups had most of their calves in February, which is late in the mating season. This is possibly due to the late start of the wet season last year and the year before. There is an increase in both non-pregnant and dry and non-pregnant and wet cows this year. This is probably due to late calving. Another year is needed to determine whether it is a one-off event.

Another abnormal occurrence this year was the death of two adult buffalo cows due to dystocia.

Sales
There were no breeder sales this year except for swamp and F1 groups and their progeny to a local producer. All other turnoff animals were exported for slaughter. The recent weight limits imposed by Indonesia have caught many off guard, creating difficulties in finding a market for animals weighing over 350 kg.

The Darwin buffalo dairy has not yet commenced commercial production, but is expected to do so by the end of 2010. The main hurdles were regulatory in nature; most have now been resolved, albeit slowly. The dairy operator is gaining experience and is collecting feasibility data to determine the long-term viability of dairying in the Top End. The experience at Millaa Millaa suggests that buffalo dairying is feasible in the Top End provided there is sufficient quality feed. The advantage of raising buffalo is their ability to produce milk from grass without needing large quantities of concentrates or other supplementary feed.

PROJECT: TenderBuff Development and Supply

Project Officers: B. Lemcke, E. Cox, L. Huth J. Stevens and Beatrice Hill Farm Staff

Location: Darwin

Keyword(s): buffalo, TenderBuff

Objectives:

To promote and implement the TenderBuff quality assurance program for local and interstate markets.

To ensure a year-round supply of TenderBuff in the Northern Territory (NT).

Background:

The TenderBuff program was initially started to facilitate higher returns to the buffalo producer whose cattle numbers had been reduced following BTEC. TenderBuff was seen as a reasonable substitute for feral-derived eye fillet for the restaurant trade. It was also an opportunity to supply a much wider range of high quality cuts.

The project was conducted with the NT Buffalo Industry Council. It conducted quality assurance and branded carcases at the abattoir. The producer received $3.10/kg hot standard carcass weight (HSCW). The carcase must meet five specifications to receive the TenderBuff strip brand.

TenderBuff has lower cholesterol and fat than beef. These two factors can be used for positive marketing of the product as an alternative red meat. As riverine cross buffalo grow much faster than swamp buffalo, they can be turned-off at a much younger age and should therefore produce more tender meat.

Another problem to affect local TenderBuff production was the closure of Litchfield Abattoir in March 2007. It is hoped that the Gunbalanya Meat Supply Company at Oenpelli, run by the Indigenous Land Council, will be able to fill in this gap when the abattoir is refurbished sometime in 2010. All current production is sent live to overseas markets, mainly in Indonesia, at a lower price.

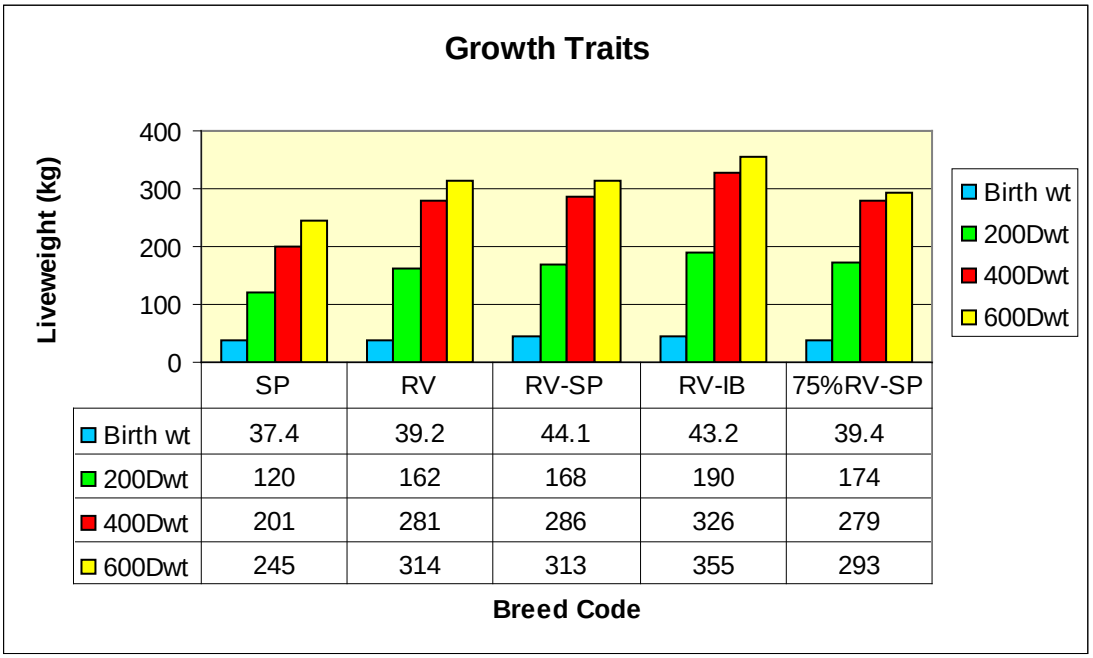
Method:

The current TenderBuff specifications are:

- 150-300 kg HSCW.
- 3-12 mm fat at p8 site.
- No permanent teeth.
- Electrically stimulated carcase.
- Muscle pH below 5.8 after 18 hours.

Results:

Data analysed by ABRI has indicated that there is a further 40% increase in productivity following the use of Italian semen in US riverine buffalo over and above the 40% of that due to the use of US riverine semen in swamp buffalo.



	SP	RV	RV-SP	RV-IB	75%RV-SP
Birth wt	37.4	39.2	44.1	43.2	39.4
200Dwt	120	162	168	190	174
400Dwt	201	281	286	326	279
600Dwt	245	314	313	355	293

Breed codes: SP= Swamp, RV= US River, IB= Italian

Figure 1. Growth rates in buffalo breeds

Eight animals were slaughtered at Oenpelli for the 2010 Royal Darwin Show. The abattoir facilities are limited due to insufficient rail height to hang full sides and the absence of equipment to electrically stimulate carcases. The abattoir is also isolated during the wet season making it difficult to slaughter all year round. Areas for wet season stockpiling and grazing are also very limited.

There is a rumour that an abattoir will be built in the Darwin area to counter Indonesian limitations on imports for an extended period of time. Such a move would be welcomed by the buffalo industry.

PROJECT: Multi-breed Composite/Brahman Breed Comparison

Project Officers: B. Lemcke, E. Cox, L. Huth, P. O'Brien, S. Reed, C. Hazel, Beatrice Hill Farm (BHF) and Douglas Daly Research Farm (DDRF) Staff

Location: Darwin

Keyword(s): cattle, breeding, Brahman, multi-breed composite

Objective:

To measure the relative growth, reproductive performance and carcase characteristics of the interbred progeny of some tropically-adapted multi-breed crossbred bulls mated to Brahman cows, compared with the progeny of Brahman bulls mated to Brahman cows in the Top End.

Background:

The argument for a more suitable animal than the Brahman for the Top End of the Northern Territory and its overseas markets has been around for some time, particularly for an animal suitable for a southern market should SE Asian markets fail at some time. A multi-breed composite is able to retain a larger amount of available heterosis compared with conventional two breed crossing systems. This project aims to compare the performance of a multi-breed composite under Top End conditions and assess its suitability as a possible replacement for the Brahman. A composite is better able to display the best attributes of a larger number of breeds and combine them through selection to produce a new “breed” for this environment. The project was initially conducted at DDRF, but the cows were run at Victoria River Research Station for six years. In 2009 the breeder herd was transferred to BHF in the Top End to finish off the project.

Method:

The mix of breeds in the composite is 56.3% Brahman, 12.5% Africander, 12.5% Tuli, plus 6.3% each of Shorthorn, Hereford and Charolais. This mix is 81% tropically-adapted and 19% unadapted *Bos taurus*. It is expected to retain 64% of heterosis in the second generation onwards.

The heifers are relocated to DDRF for yearling mating and the bulls for testing by the end on the dry season. The two breeder groups are run separately for mating from mid December when the bulls are introduced and removed in late March, then run together until calving (for ease of mothering-up).

Cows are culled if non-pregnant (except as yearlings or as lactating two-year-olds) and bulls are selected on growth rate, scrotal circumference and percentage normal sperm. Bulls are used for a maximum of three years in the herd, as yearlings with yearling heifers, as two-year-olds with two-year-old heifers (both at DDRF) and as adults with the breeding herd at BHF. They are then sold by tender to local producers. Some heifers are used on a yearly basis for artificial insemination with semen from outside the herd to reduce inbreeding and to introduce new genes into the mix. The herd is too small for use in inter-se mating exclusively.

Data is recorded in the HerdMaster recording package, the animals are registered with the Australia Brahman Breeders Association and also recorded in the Brahman BREEDPLAN.

Results:

All Brahman and composite cows selected for the comparison at BHF were pregnant before the 2009 calving season. Non-pregnant females were culled.

Table 1. Herd statistics

	Composite herd at BHF	**Brahman herd at BHF**	**Composite two-year-old heifers at DDRF**	**Composite yearling heifers at DDRF**
Herd size at start of mating (number)	102	59	48	49
Pregnancy rate 2009 mating (%)	100. All non-pregnant culled	100. All non-pregnant culled	35/48 = 72.9	27/49= 55.1
Calves born	102	59	35	27
Mean birth wt (kg)	26.4	25.5	27.9	23.0
Calf mortalities (number/%)	17/16.7	13/22	6/17.1	3/27 = 11.1
Calves weaned/% weaning rate	85/83.3	46/78.0	29/60.4	24/49.0
Calf growth rate to weaning (kg/animal/day)	0.73	0.74	0.84	0.71
Mean weaning wt (kg)/age (days)	181.6/207	169.9/195	192.6/196	154.7/185
2010 pregnancy rate	Not yet tested	Not yet tested	75/105 = 75.0% (two and three-year-old)	30/76= 39.5%
Adult mortality	Three (late wet)	1		

Calf death rates were higher than expected due to the late start of the wet, which was particularly stressful for Brahman heifers at BHF, where floodplain good feed was available but there were few trees in the main calving paddocks. Better seasonal conditions, such as the late finish to the last wet season, may help improve the performance of the animals next year.

Birth weights were not significantly different; however, lower birth weights in Brahmans were unusual, but were likely due to a higher proportion of heifers in the Brahman group. Generally, composites have lower birth weights.

Adult deaths were a mystery late in the wet. A possible cause may have been ephemeral fever as the cows were not in poor condition at the time. Brahmans in the BHF herd will be gradually increased.

PROJECT: The Use of Alternate Water Points to increase Pasture Utilisation

Project Officers: K. Scott and R. Cowley

Location: Barkly

Keyword(s): rotational grazing, cattle, water points, spelling, Rockhampton Downs

Objective:

To determine the feasibility of rotational grazing facilitated by water availability in a commercial environment and its effect on pasture composition and yield, animal production, animal behaviour and labour requirements.

Background:

Properties in the Barkly have relatively large, but poorly-watered paddocks, averaging 150-360 km² depending on district (Bubb 2006). Up to 32% of the region is more than 5 km from a water source (Fisher 2001). Given that cattle spend most of their time within 5 km from water (Fisher 2001), a lot of grazing land is relatively unused, while areas closer to water can be overused, leading to poor land condition. Hence, the development of watering points and paddock subdivisions are high priorities for many producers. However, the cost is limiting the rate of development for many.

As an alternative to traditional continuous stocking, other grazing systems, such as rotational grazing may provide an opportunity to increase production whilst also maintaining (or improving) rangeland condition. This project reports on a three-year, innovative rotational grazing trial conducted at Rockhampton Downs Station in the Barkly Tableland region of the Northern Territory. Rather than developing new fences to create multiple paddocks, cattle were rotated around a paddock by controlling the availability of water at each of several new and existing water points. The central concept of the strategy involved having only one water point operating at any given time.

Method:

An existing 538 km^2 paddock was subdivided to create a control paddock (278 km^2) and a treatment paddock (253 km^2). The control paddock employed a continuous grazing system (previously used at the site), while the treatment paddock employed the alternate watering rotational grazing system. Three existing water points were located inside the control paddock, whereas the treatment paddock utilised a network of both existing water points and new troughs giving a total of five water points.

Cattle were moved to new water points every six weeks, generally in an anticlockwise direction. Intensive supervision was required to achieve the desired rotational grazing. In the wet season, all bores were turned off following the first substantial rains that provided surface water. Cattle then relied on surface water and semi-permanent dams. Once surface waters had dried up, troughs were reactivated. In this way, all water points in the rotational grazing area were effectively spelled for up to three months during the wet season.

Pasture species at the site were recorded using BOTANAL (Tothill et al. 1992), in early (April-May) and late (October) dry season during the trial period in 2004 and 2006, but only in the late dry season in 2005 to quantify changes in pasture composition and condition. Animal production parameters were quantified annually between 2004 and 2006 at the first round muster in May. Collected data included breeder weight, weaning weight,

pregnancy status and foetal age, and lactation status (determined visually by several operators, although breeders were not 'hand stripped' for confirmation).

Results:

The trial demonstrated that a rotational grazing system can be implemented on commercial beef cattle properties. This represented a substantial mind-shift away from a continuous set stocked regime traditionally employed throughout northern Australia.

Infrastructure development, labour costs and the management of cattle initially present challenges to such a strategy. Cattle behaviour was difficult to manage, particularly in the first two years. Cattle would often congregate at dry water points, requiring frequent monitoring and management, making the trial very labour-intensive. Over time though, managers devised a procedure whereby the next water point was turned on when the current water point was turned off, on the day prior to moving the cattle. This significantly reduced the labour required to implement the system. Managers used the rotational grazing system to have cattle closer to yards at mustering, thereby saving a significant amount of time and money. The cost savings, achieved through more efficient management, could help offset the initial capital investment.

Infrastructure development (the installation of new water points) increased carrying capacity by increasing the watered area of the paddock. Higher stocking rates could be achieved in areas traditionally not grazed due to their higher yield. However, that bonus will be reduced over time if the system is not managed in a sustainable manner.

Land condition (measured by species composition, yield and cover) associated with new water points in the rotational grazing paddock appeared to follow a trajectory of degradation towards that shown around old water points with a long history of continuous stocking.

The previously un-grazed pasture showed some initial resistance to the effects of grazing following the introduction of rotational grazing and there were some signs of degradation over time. There was no significant reduction in total species richness during the three year trial and the abundance of the dominant perennial Mitchell grasses was stable except in areas immediately adjacent to water points.

The rotational grazing strategy did not improve existing land condition at old water points during the period of the study.

Problems in the design and implementation of the trial (data quality) did not allow for a reliable assessment of differences in animal production (live-weight gain and breeder performance) between the grazing systems.

At its completion, the trial gave station managers a better understanding of the possibilities of manipulating pasture utilisation and they expressed interest in applying that knowledge in future grazing management strategies.

A full report is now available on this project (see Scott et al. 2010).

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PROJECT: Rangeland Grazing Strategies for Improved Economics and Resource Sustainability

Project Officer: C. Materne

Location: Alice Springs

Keyword(s): grazing strategies, rotational grazing, pasture spelling, cattle

Objective:

To develop industry-acknowledged best practice guidelines for two grazing strategies in Central Australia that incorporate spelling, and contribute to improved economic viability and resource sustainability.

Background:

Best practice guidelines for Central Australian spell grazing strategies

Over the past decade, the ongoing cost-price squeeze has forced pastoralists to look at using their land more efficiently. There has been widespread interest by the industry in spelling practices, which hold potential for increased or sustained production with minimal impact on natural resources. However, this potential has been little documented, particularly on a practical whole-of-property/business scale. Heytesbury Beef established a major commercial trial of intensified land use in the tropical savannah region at Pigeon Hole Station. A project is also being conducted in the southwest rangelands of Western Australia (WA) to monitor the effects of various practices tested at the property level by pastoralists. In Queensland, a Meat and Livestock Australia project is exploring the environmental and economic benefits and costs of different grazing systems, particularly cell grazing. Thus, existing trials encompass summer and winter-dominant rainfall regions of the rangelands but leave a large gap in the non-seasonal and more variable sector of the rangelands, including western NSW, south western Queensland, Central Australia and the Murchison in WA. This project was developed to cover the gap.

The major benefits of spell-grazing practices include:

- Opportunities to spell country at critical times for pasture regeneration.
- Closer observation of the condition of the country to improve both the quality of animals produced for the market and resource condition.
- Better drought preparedness.
- Potential for carbon sequestration and the production of offsets for trading.

Potential risks of spell grazing include:

- Capital costs of setting up a more intensively fenced and watered system.
- Errors of judgment in leaving stock on country for too long (though offset by damaging less area in one go), probably riskier in regions with a more variable climate or fragile soils.
- Lack of access to markets at critical times.

Method:

Two grazing trials were set up in Central Australia, one at Mt Riddock Station and another at the Old Man Plains Research Station (OMPRS).

At Mt Riddock, steers were grazed through an eight paddock rotation. All paddocks consisted of buffel grass-dominated pasture that was annually spelled over the 'summer' growing period. Steers from each treatment were weighed at the beginning and end of the rotation period.

The strategy at OMPRS consists of a four-paddock rotation that allows for annual 'summer' spelling of the calcareous grasslands and biennial 'summer' spelling of the more resilient mulga country. Breeder performance was recorded annually.

In both trials, pastures were monitored pre- and post-grazing for yield, cover and composition. Animal grazing activity and behaviour were recorded using a defoliation index and a cattle activity index.

Results:

A report was submitted to the funding body, *Caring for our Country*. The project period ended on 30 June, 2010 and the final report was submitted. Technical reports on land condition and herd performance were drafted for both trials and include future project recommendations. Best practice guidelines for alternate grazing strategies in Central Australia have been drafted and will be published as an *Agnote.* Communication activities during the project included:

- A field day at OMPRS - "Being in the Grass Business". It was attended by 40 stakeholders representing 14 stations (38 000 km^2), three organisations, one business and three government departments.
- Five articles were published in the *Alice Springs Rural Review*:
    - "Differences in calf growth"
    - "More grass than cattle - what to do?"
    - OMPRS field day – being in the grass business"
    - "Rotational grazing – how Mt Riddock approached the year that was"
    - "Culling of low performance bulls, to improve herd efficiency"
- Four radio interviews:
    - Rainfall – what does this mean for growth?
    - 101 ways to graze
    - Grass can be more valuable than you think
    - 101 ways to graze (x2).

PROJECT: Enhancing Adoption Of Improved Grazing and Fire Management Practices in Northern Australia: Synthesis of Research and Identification of Best Bet Management Guidelines (also known as The Northern Grazing Systems Project)

Project Officers: D. Walsh, R. Cowley, S. Leigo and C. Collier

Location:	NT
Keyword(s):	sustainable pastoral management, pasture spelling, prescribed burning, infrastructure development, stocking rates, climate variability, cattle

Objectives:

To improve our understanding of the interactions and trade-offs, and identify cost-effective grazing land management options for:

- ***Improving animal production and economic performance.***
- ***Improving and maintaining land condition (vegetation, soil health and water quality).***
- ***Improving risk management in relation to climate variability.***

To integrate, enhance and extend key findings generated by the grazing and fire research projects across northern Australia.

Background:

This project forms the Northern Territory (NT) component of a cross-jurisdictional project, which aims to increase the adoption of innovative and practical best-practice grazing management by beef producers in Queensland, the NT and the Kimberley region of Western Australia. The Northern Grazing Systems (NGS) initiative is funded by Meat and Livestock Australia, state, NT and Australian governments. The Queensland Department of Employment, Economic Development and Innovation (which incorporates Primary Industries) manages the project, which is due for completion in June 2012.

The importance of infrastructure development (fencing, waters), stocking rate management, pasture spelling, and prescribed burning for sustainable pastoral management have been demonstrated at various field study sites. However, we are unable to predict how variations and combinations of these practices will affect the productive capacity and resource condition of grazing land in particular situations. In addition, the economic and practical implications of implementing these strategies at an enterprise level are often unclear. This is limiting the rate of adoption of practices to improve grazing and fire management across northern Australia.

The NGS project thus aims to integrate, enhance and extend key findings and knowledge generated from completed grazing and fire research across northern Australia.

Method:

The NGS project is being delivered in two phases. Phase 1, which was completed in September 2010, consisted of three activities:

- "Synthesis" – which reviewed, analysed and synthesised data and outputs from completed field research studies across northern Australia to develop additional insights, produce relationships that assist extrapolation of a range of environments and starting conditions, and generate a suite of best-bet management guidelines and strategies for different environments and scales of operation.
- "Regional assessment" – which sourced, collated and reported region-specific research data, herd and pasture management practices, and facilitated the input of local producers and other regional specialists in identifying and assessing best-bet management options.
- "Bio-economic modelling" – which modified, linked and applied existing simulation models to evaluate animal production, land condition and economic performance of the identified best-bet options in each region.

Phase 2, which commenced in late 2010, will implement, test and increase adoption of these practices through on-property demonstration sites, field days, forums, training workshops and MLA/other agency publications. The project will also identify major research gaps for further investigation in each region.

Results:

The Synthesis team identified four common and important land management issues impacting on the productivity, profitability and sustainability of the northern beef industry (McIvor et al. 2010):

- Matching animal demand to pasture supply on pastures in good condition.
- Managing pastures in poor condition.
- Woody vegetation thickening.
- Pastures too distant from water to be utilised by cattle.

The Synthesis team also identified the four grazing land management options that hold the most promise for managing these issues:

- Stocking rate management.
- Wet season spelling.
- Prescribed burning.
- Infrastructure development.

To gather local input and opinion on these issues and management options, two workshops are held in each region. The first workshops in Katherine and Tennant Creek were held in April 2009 and the first Alice Springs workshop was held in April 2010. Attendees included pastoralists, agricultural advisers and agency technical staff. The workshops documented current and best management practices related to infrastructure development, stocking rate management, pasture spelling and the use of prescribed fire for each region. "Representative" properties in each region were described by participants for use in bio-economic modelling. Since those workshops, a comprehensive summary of grazing management options (based on the literature and workshop input) has been produced (McIvor et al. 2010). Preliminary bio-economic modelling of different management options has been completed for the Barkly and Victoria River District regions and the findings were presented to producers and advisers for feedback at workshops held in April, 2010 (White and Walsh 2010a, White and Walsh 2010b). The Alice Springs region is currently being modelled and a second workshop will be held there to gather feedback on those results in late 2010. Producer demonstration sites will be held in each region between 2010 and 2012 to promote the most promising practices to increase industry adoption. Further bio-economic modelling will

also be undertaken to test the potential impact of predicted climate change on the performance of the recommended management options.

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PROJECT: Developing Sustainable Carrying Capacities in the NT

Project Officers: C. Pettit and R. Cowley (Sturt Plateau), K. Scott (Barkly) and C. Materne (Alice Springs)

Location: NT

Keyword(s): carrying capacity, cattle, grazing

Objectives:

To develop methodologies for the objective assessment of carrying capacity, including calibration of pasture growth models, for the Sturt Plateau, Barkly and the Alice Springs regions.

To develop methodologies to evaluate carrying capacity and make them available to pastoralists so they can make decisions on seasonal and long-term stocking strategies.

Background:

There is potential in the Northern Territory (NT) cattle industry to increase production through subdivision and intensification of land use. However, land intensification in other states has sometimes led to unviable small blocks and extensive land degradation, due to an over-optimistic assessment of land capability. Therefore, to facilitate sustainable development of the NT cattle industry, it is imperative to have an objective and transparent method for estimating carrying capacity, particularly where properties are being subdivided.

This project aims to calibrate the GRASP model to facilitate estimation of sustainable carrying capacities in important grazing pasture types of the NT (in the Barkly, Sturt Plateau and Alice regions). The GRASP model is calibrated through the collection of pasture, soil and meteorological data from small exclosures called SWIFTSYND sites. The exclosures have been set up on areas that represent different land systems and vegetation types in order to obtain a broad viewpoint across the region.

Method:

Field pasture growth measurements followed the SWIFTSYND methodology (Day and Philip 1997) with up to four measurements per year, usually following a reset (where pasture is removed by mow or burn) at the end of each dry season. The timing and number of measurements for the Alice Springs region depended on rainfall events. Pasture, soil and rainfall were measured at the sites. Field data was then used to calibrate the GRASP pasture

growth model for each site. Individual sites were then grouped by land type and average land type parameter sets were developed. Those land type parameter sets were used to model historical pasture growth, which allowed long- term median pasture growth to be calculated for different land types and regions.

Alice Springs

Six sites were constructed on four stations and the Old Man Plains Research Station (OMPRS) during 2004-05. They represented the following land systems: Alcoota (Alcoota Station), Ebenezer (Mt Ebenezer Station), Muller (OMPRS), Outounya (Umbeara Station), Renners (Deep Well Station) and Sandover (Alcoota Station). Harvests were undertaken when the predominant annual grasses had begun seeding (approximately 10-14 days after rain).

Barkly

Eleven monitoring sites were constructed in the Barkly region. They were located at Alexandria, Beetaloo, Brunette Downs, Helen Springs, Newcastle Waters, Rockhampton Downs and Walhallow stations. The land systems represented include Barkly, Creswell, Austral, Wonorah and Pollyarra.

Sturt Plateau

Ten SWIFTSYND sites were located around the Sturt Plateau on five different land systems. The selected land systems were Banjo, Larrimah, Sturt, Bulwaddy and Elsey. Together, the land systems represent 71% of the total Sturt Plateau region.

Results:

Alice Springs

Between six and 12 harvests were conducted per site between 2005 and 2010. Six sites were harvested in early 2010. Field data was collated for calibration of GRASP in September 2010.

Barkly

Between eight and 12 harvests were conducted on each site between 2005 and 2009 (Figure 1).

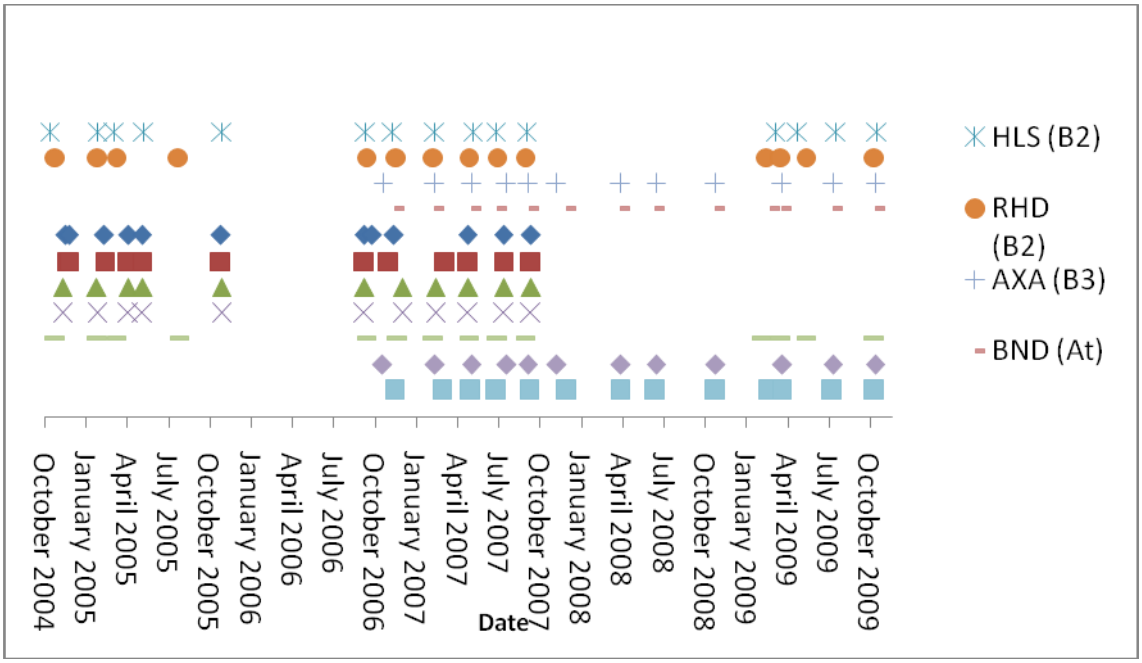

Figure 1. Field measurement and reset (mow or burn) timing for the Barkly SWIFTSYND sites between 2005 and 2009

Field data has been collated for the calibration of GRASP. GRASP land type parameter sets were finalised in August 2010.

Sturt Plateau

Field work was completed in 2007. All sites have been calibrated, land type parameter sets have been developed for the major land systems and soil types have been sampled. GRASP was able to closely simulate pasture growth for the sites. As an example, Figure 2 shows the fit of modelled pasture growth (blue line) to the measured pasture growth (red dots) for Bulwaddy land system on a site in the southern Sturt Plateau.

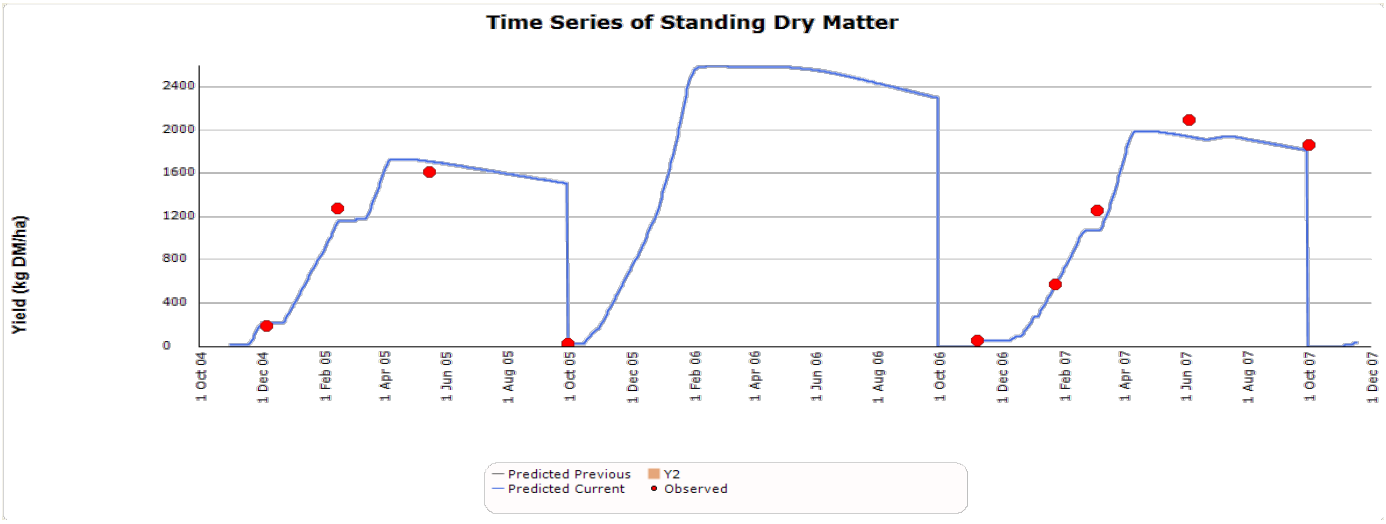

Figure 2. Modelled and actual pasture growth on a Bulwaddy land system in the southern Sturt Plateau

GRASP parameter sets were used to model median pasture growth for the different land types on 13 stations in the Sturt Plateau region and delivered through the Sturt Plateau Grazing Land Management Workshop in April 2010. See Table 1 for an example of modelled pasture growth for one land type and location.

Table 1. Sturt Plateau GLM modelled annual pasture growth for Bulwaddy land system in the western Sturt Plateau (kg DM/ha)

Tree basal area (m²/ha)	A condition	B condition	C condition	D condition
0	3200	2400	1400	600
1	3000	2300	1400	600
2	2800	2100	1300	600
4	2500	1900	1100	500
6	2200	1700	1000	400
8	2000	1500	900	400
10	1800	1400	800	400
12	1700	1300	800	300
15	1500	1100	700	300
20	1300	1000	600	300

Median rainfall 680 mm, soil type gravelly and deep loamy red earths.

GRASP was also used to model pasture growth and carrying capacity across the Sturt Plateau for the Katherine Research Station open day in February 2010. See Table 2 for modelled pasture growth and carrying capacity for different land types in the northern, central and southern Sturt Plateau.

Table 2. Variation in pasture growth and calculated long-term carrying capacity for major land types across the Sturt Plateau

Region of Sturt Plateau	**Alluvial cracking clay B condition**	**Good red soils A condition**	**Average red soils A condition**	**Gravelly shallow red soils B condition**
		Recommended utilisation rate (%)		
Northern	20	15	15	10
Central	20	15	15	10
Southern	20	15	15	10
		Median pasture growth (kg/ha)		
Northern	2400	2850	2250	2100
Central	2250	2850	2200	2050
Southern	1921	2700	2050	1750
		Long term carrying capacity (AE/km^2)		
Northern	16	15	12	7
Central	15	15	11	7
Southern	13	14	11	6

Assumptions in the modelling: pasture growth calculations are for average land condition and tree cover for these land types in the region, assuming 20% of the land area burns every year when pasture in October is at least 1500 kg/ha.

A full report on the Sturt Plateau pasture growth study will be written in late 2010.

Reference:

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PROJECT: Seasonal Burning of Mitchell Grassland on the Barkly Tableland

Project Officer: C. Materne and M. Hearnden

Location:	Barkly
Keyword(s):	Mitchell grassland, seasonal burning, prescribed burning, rangeland management, cattle

Objectives:

To measure the impact of low intensity, early dry-season fires and high intensity, late dry-season fires on Mitchell grasslands and woody plant species in Buchanan paddock at Alexandria Station.

To demonstrate the application of prescribed burning in the extensively-grazed Mitchell grasslands of the Barkly region.

Background:

The relationship between fire and Mitchell grasslands is not known due to a view that "the Mitchell grasslands are too valuable as a pasture to be burnt" (Thackway et al. 2007). However, more recently, observations by station managers and naturalists of Mitchell grassland recovering after wildfires have stimulated curiosity about whether seasonal fire is needed to maintain good condition in Mitchell grasslands of the Barkly Tableland. Scanlan (1980) found that burning under low soil moisture conditions followed by low rainfall was detrimental to the condition of Mitchell grasslands. Phelps and Bates (1996) demonstrated the use of spring fire as a tool for managing the undesirable *Aristida latifolia* (Feathertop wiregrass) in Mitchell grasslands grazed by sheep in Queensland. The aim of this trial was to better understand the importance of seasonal prescribed burning as a management tool for Mitchell grasslands under continuous grazing and its effect on cattle production on the Barkly Tableland.

Method:

This trial consists of two parts: An intensively-sampled plot trial to identify the response of Mitchell grasslands to burning at two different times of the calendar year and its effect on the encroaching woody vegetation; and a broader paddock-scale trial to demonstrate the use of fire as a pasture management tool.

The trial was conducted at Alexandria Station between 2001 and 2004. The trial site, dominated by *Astrebla pectinata* (Barley Mitchell grass) and *A. elymoides* (weeping Mitchell grass), is in the sub-tropical grassland climate zone and has a median July to June rainfall of 350 mm. The effects of early and late-dry-season burning on native trees and shrubs, pasture dynamics, cattle diet quality and grazing characteristics were investigated using a randomised three plot (25 ha each) block design with four replications in a paddock under continuous grazing with conservative stocking rates. Tree and shrub data was collected prior to treatment. Pasture yield, species composition and cover were collected from 50 x 1 m^2 quadrats on four parallel transects each 400 m long. Pasture quality data was collected using the 'grab sampling' technique (Ash and McIvor 1995) and analysed using wet chemistry methods. Cattle diet was analysed using near infrared spectroscopy (NIRS) on a paddock scale, replicated twice. Repeated measures ANOVA was used to test the interaction effect of treatment and time. *Post priori* comparisons were used to test for differences between treatments within different time periods.

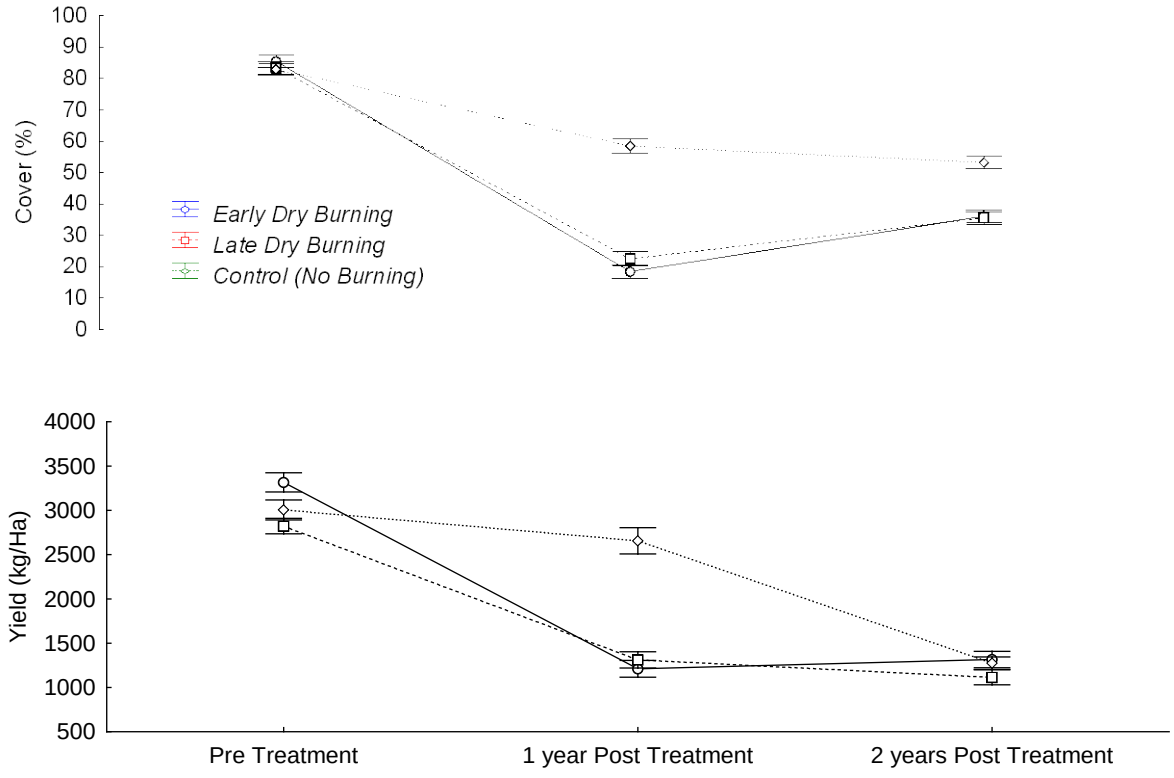
Results:

The financial year (July to June) rainfall totals during the trial were close to the median of 350 mm, although it became increasingly drier as the trial progressed. This followed consecutive above-average seasons (top decile) in 1999-2000 and 2000-01.

For all treatments, cover and yield (Figures 1and 2) progressively decreased over the trial period due to the decreasing seasonal rainfall ($F_{2,40}$=206, *P*<0.001, and $F_{2,40}$=316, *P*<0.001, respectively). Although no significant difference in cover ($F_{1,20}$=0.35; *P*=0.55) or yield ($F_{1,20}$=0.0; *P*=0.99) was recorded between the two seasonal burning treatments, burning itself significantly reduced both cover ($F_{1,20}$= 78.3; *P*<0.0001) and yield ($F_{1,20}$=35.9; *P*<0.0001) following the first growing season. Yield recovered but cover was still lower in burned plots after the second growing season ($F_{1,20}$=0.13; *P*=0.72, $F_{1,20}$=11.1, *P*<0.01, respectively).

Flora species richness was largely influenced by seasonal response ($F_{2,40}$=171.6; *P*<0.001). However, fire significantly increased species richness following the first growing season ($F_{1,20}$=7.8; *P*=0.01) (Figure 3). There was no significant difference between the two seasonal burning treatments after one year ($F_{1,20}$=0.4; *P*=0.05). The effect of fire on flora richness disappeared after two growing seasons.

Woody plant death rates increased following the burning treatments and were greatest under the late dry-season burns (Figure 4). Considerable re-sprouting occurred following burning, but consecutive drier than average years resulted in further deaths following the second year. The height of surviving woody vegetation was reduced on average by about 40% regardless of burning season (Figure 5).



Figures 1 and 2. Seasonal burning effect on ground cover and pasture yield under grazing, with 95% confidence limits (measured in April)

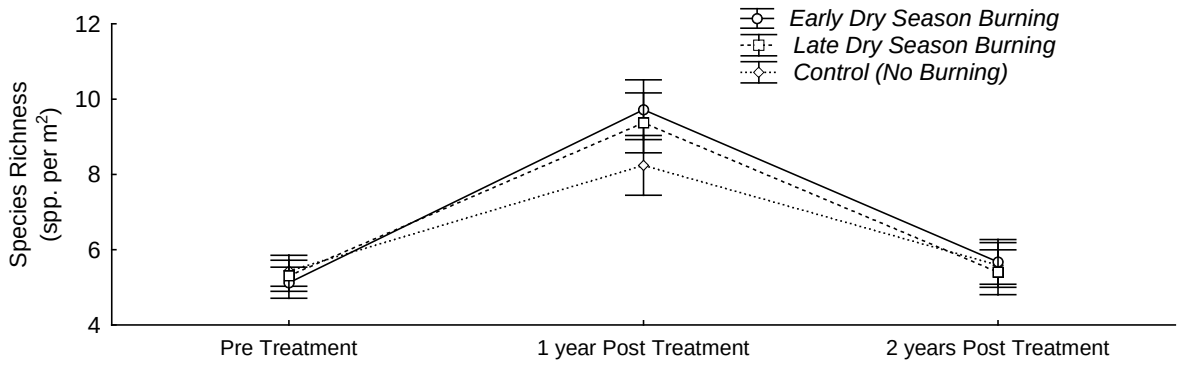
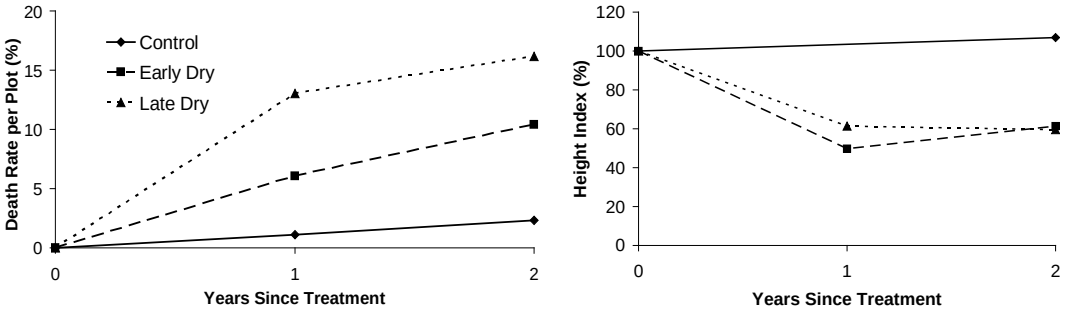


Figure 3. The effect of seasonal burning on species richness within a 1-m area over time with 95% confidence intervals (measured in April)



Figures 4 and 5. Woody vegetation death rate and height under different seasonal burning regimes

Cattle activity increased in all treatments over the trial period (Figure 6) due to the decreasing seasonal rainfall ($F_{2,40}$=124.9, *P*<0.001). Burning further significantly increased cattle activity ($F_{1,20}$=6.7; *P*=0.01) over at least two dry seasons. Short-term improvements in Mitchell grass feed quality over the first growing season were found following fire (Figure 7). However, NIRS sampling (Figure 8) indicated the effect on diet was relatively small and only at the beginning of the growing season before the annual grasses and forbs germinated.

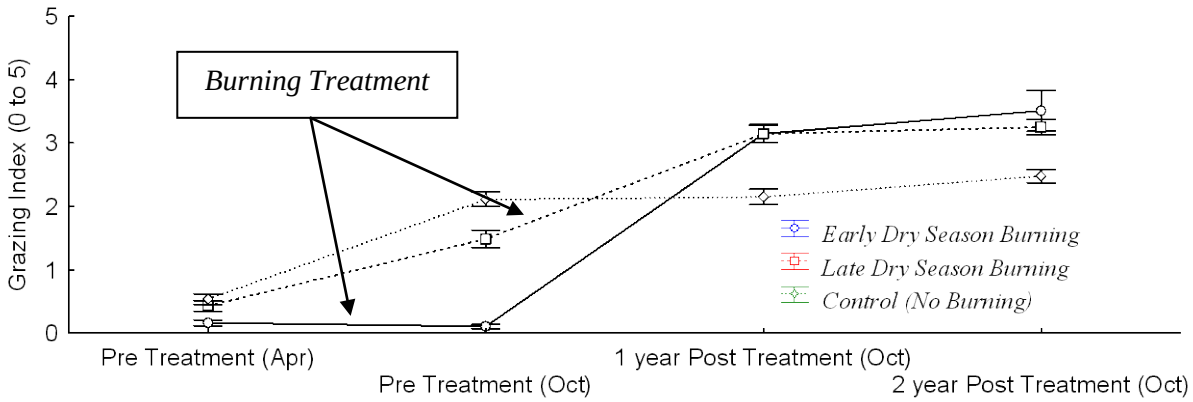
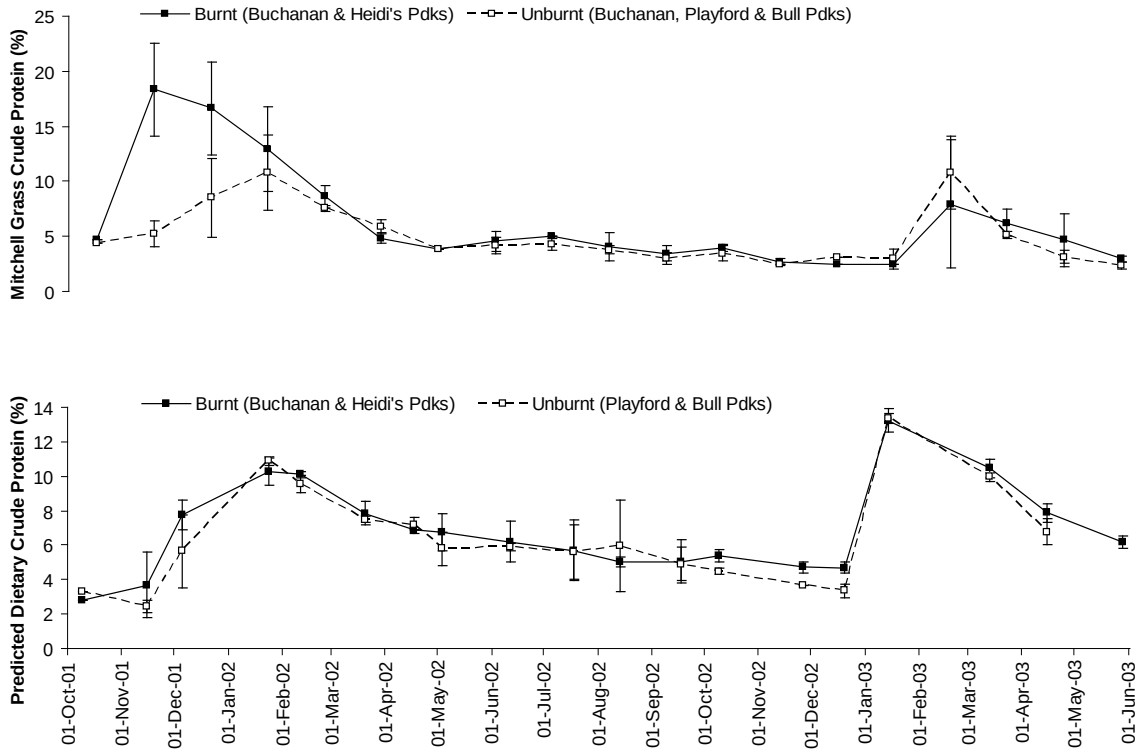


Figure 6. The effect of seasonal burning on cattle grazing activity, with 95% confidence limits



Figures 7 and 8. Crude protein content of late-dry-season burnt and unburnt Mitchell grass (*Astrebla pectinata* and *A. elymoides*) from grab sampling (analysis using the dry chemical method) and the predicted cattle diet quality from dung samples (NIRS analysis), with 95% confidence limits

Discussion:

The greatest pasture management risk with prescribed burning in Mitchell grasslands in more arid areas is a lack of follow-up rain, and the resulting short-term feed loss and production (Daubenmire 1968; Scanlan 1980). However, dry matter differences between the burnt and unburnt plots during drier seasons appear to be due to carry-over feed in the unburnt plots, rather than a lack of seasonal growth in the burnt plots. Following the second consecutive below-average growing season, no difference was identified in the available feed between the burnt and unburnt sites, similar to Scanlan's (1983) findings. With adequate rainfall, Scanlan (1980) found these grasslands can recover rapidly within one growing season, which is supported by observations in this study.

Following fire, Mitchell grassland cover appears to require three growing seasons to recover, a finding similar to that of Dyer et al. (2003) in the higher rainfall Victoria River District of the Northern Territory. Cover is considered an important land condition indicator (Materne 2005; Chilcott et al. 2007). However, personal observations, supported by research by Scanlan (1983), suggest the cover change was dominated by the litter component and not the more stable Mitchell grass basal area. Hence, the reduction in cover in this trial is not considered an indicator of declining land condition. This reduced litter cover is likely to have contributed to increased species richness because high litter levels tend to suppress germination and establishment of annuals.

Even though many woody species within the trial area are considered to be relatively fire-tolerant re-sprouters, fire still had a significant effect on survival and height.

Although short-term negative impacts were recorded following fire in Mitchell grassland on the Barkly Tableland, the results of this trial indicate that these grasslands are resilient to fire under conservative stocking, even under less than favourable growing conditions. Late dry-season burning has the potential to provide production benefits

and can be a useful and cost-effective management tool to remove rank pasture, reduce tree and shrub cover, influence cattle movements and improve short-term diet quality.

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PROJECT: Safe Utilisation Rates for NT Land Types

Project Officers: D. Walsh and R. Cowley

Location:	NT
Keyword(s):	carrying capacity, cattle, pasture utilisation, sustainable pastoralism

Objective:

To determine safe pasture utilisation rates for important pastoral land types across the Northern Territory (NT).

Background:

The pastoral industry in central and northern parts of the NT is currently undergoing rapid expansion and development with a focus on intensification. Dyer et al. (2001) predicted that intensification and development would continue in the northern beef industry due to the need to reduce the cost of production, increase the efficiency of production and maximise returns. This suggests that pasture utilisation will increase in northern and central parts of the NT, creating a need for research to determine optimum utilisation rates for high production without degrading the pasture resource (Dyer et al. 2001).

In the southern parts of the NT, where full development is close to completion, there is an apparent disparity at the property level between what is considered an economically viable herd size and recommendations about safe carrying capacities. This has recently been borne out during grazing trials conducted in the Alice Springs region, where large differences between scientifically-derived carrying capacities and actual stocking rates were documented (Kain 2008). Kain concluded there was an urgent need to work with the industry to document current utilisation rates, investigate the rates of utilisation in areas that have good land condition and determine sustainable carrying capacities for different land types. She also recommended that more work be done to determine whether there were factors influencing carrying capacity that are not currently taken into account in the methodology used to calculate it.

Currently, the only objective NT-tested knowledge on utilisation rates is from a limited number of case studies and grazing trials on black-soil land types in the Victoria River District (VRD). For black-soil pastures in good condition in the VRD, utilisation rates of 20-25% have been found to be sustainable. However, current Department of Resources (DoR) recommendations of a 15% utilisation rate for pastures on productive red soils, 10% for pastures on less productive red soils and 5% for spinifex pastures, remain untested. Hence, there is an urgent need to objectively assess utilisation levels of important pasture communities throughout the NT to guide sustainable development and management of the pastoral resource.

Method:

The utilisation rate is defined as the proportion of a year's pasture growth that is consumed by cattle. This project is using methods similar to those described by Johnston et al. (1996) to calculate utilisation rates from "benchmark" grazing properties and local grazing trials. The use of benchmark grazing properties is based on the assumption that paddocks in good land condition that have a long history of grazing must have been managed using safe utilisation rates (Johnston et al. 1996). By "looking back in time" and calculating what these utilisation rates have been, the recommendations on long-term sustainable utilisation rates for different land types can be improved.

DoR is working with producers who have paddocks with pastures in good condition and several years of accurate paddock stock records. Annual utilisation rates are then calculated using:

- Annual pasture growth – based on pasture type and watered area (growth is estimated using pasture models such as GRASP or AussieGRASS).
- Estimated consumption by cattle – based on the numbers, classes and approximate age/weights of cattle in the paddock (data provided by producers).

The goal is to make recommendations based on what is considered a safe utilisation rate in the context of annual pasture growth risk. In the northern NT, an appropriate safe utilisation rate might be based on the annual growth that would be expected in an "average' year. If, however, we base stocking rates in the southern NT on the growth expected in 50% of years, this would lead to overgrazing in five out of every 10 years and a high probability that pastures would not recover before the next dry period (Partridge 1999). Thus, in the southern NT, where the climate (and thus forage availability) is highly variable and the risk of poor forage growth is higher, we intend to make recommendations on a more risk averse basis (e.g. manage for utilisation rates that are safe in 70% of years, (Scanlan et al. 1994).

Results:

Alice Springs region

Average utilisation rates ranging from 78% to 124% were calculated for the three Alice Springs case study paddocks. These figures are not consistent with the good land condition noted during paddock inspections and are considered to be a gross over-estimate of the true utilisation rates being achieved. Figure 1 shows that when a plausible top feed component is included in the pasture growth estimate, utilisation rates consistent with maintaining good land condition are achieved. For example, when 200-500 kg/ha of top feed was included,

utilisation rates closer to 20% were observed, which is consistent with current recommendations for mulga land types (Burrows 1980; Chilcott et al. 2005).

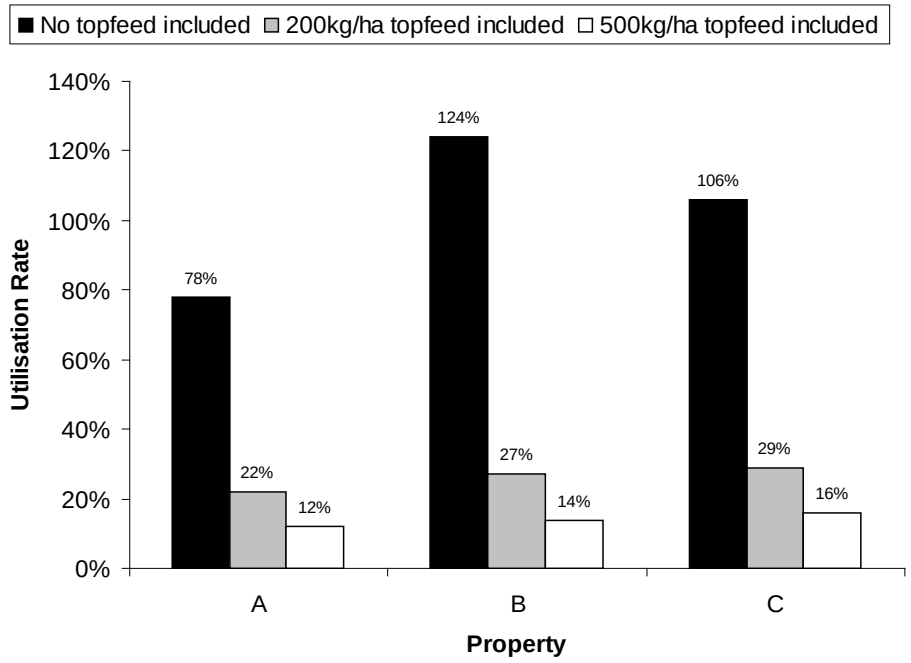

Figure 1. The influence of top feed on the utilisation rate in three case study paddocks in the Alice Springs region

With 200 kg/ha of top feed included, the average utilisation rate for the case study paddock on property A was the closest to the current recommendation of 20% (Burrows 1980; Chilcott et al. 2005). The average utilisation rate for the case study paddock on property B was 27%, but that paddock was dominated by sandy mulga country that is relatively resilient to grazing. A more precise estimate of top feed in this land type would allow us to identify the true utilisation rate being achieved in this paddock. The case study paddock on property C had the highest average utilisation rate (29%), which was reflected in the condition of some of its land types. The more robust land types in the paddock (e.g. mulga) tended to be in A to B condition but the less robust bluebush and saltbush flats tended to be in C condition (especially close to water). This indicates that the current utilisation rate is too high for the grazing-sensitive land types in this paddock, even when a top feed component is included.

Incorporating 200-500 kg/ha of top feed essentially doubles the pasture availability in relevant land types in the Alice Springs region. Therefore, the inclusion of top feed has a significant role in determining safe utilisation rates and carrying capacities. Top feed estimates for relevant land types in the NT are yet to be validated but the data supports the idea that the figure lies between 200 and 500 kg/ha (which is the figure commonly used in SW Queensland mulga pastures). For example, in the case study paddocks, the inclusion of 230 kg/ha of top feed would achieve the recommended average utilisation rate of 20% on property A, whilst the paddock on property B would require 320 kg/ha and that on property C would need 380 kg/ha.

Barkly region

Ten case study paddocks were studied in the Barkly region. The Barkly case study paddocks with lower utilisation rates tended to have more country in condition A, whilst paddocks with higher utilisation rates tended to have more country in condition B and C (Table 1). The exception was Paddock 2 on property D, which had an average utilisation rate of 33% but did not contain any condition C country. This was the smallest paddock in the study and contained only one (highly productive) land type. It had also experienced five early wet-season spells in ten years.

As expected, the paddocks with the lowest utilisation rates also had low stocking rates (Table 1). However, the expectation that high stocking rates and high utilisation rates would go hand in hand is not supported by the data. The highest stocking rates occurred in the four paddocks with mid-range utilisation rates (Table 1). All four exceeded the recommended stocking rates of 4.6 to 5.8 animals/km² suggested by Holt and Bertram (1981) and two of the paddocks also exceeded the stocking rate of up to 7.1 animals/km² recommended by Tothill and Gillies (1992). It would seem contrary to expectation that high stocking rates could be sustainable, but closer inspection of the data revealed that the four paddocks maintained relatively high average annual pasture growth (more than1200 kg/ha) over the study period. Annual pasture growth apparently dropped off sharply at utilisation rates above 32% but we intend to verify these results once the GRASP pasture growth model is calibrated for the Barkly in late 2010.

Table 1. Land condition, land type and stocking rate information, Barkly case study paddocks

	Land condition in study paddock	**Average utilisation rate (5 km watered area) (%)**	**Average stocking rate (5 km watered area) during the study (AE/km²)**	**Land systems in study paddocks (in order of largest to smallest area)**	**Recommended stocking rate (animals/km²) from Holt & Bertram (1981)**
Property D Paddock 3	Mostly A & B with small areas of C near waters	19	5.4	Barkly 3 Wonorah/Barkly 1 Wonorah	4.6-5.8 (Mitchell grass country)
Property D Paddock 7	Mostly B with some areas of A	20	4.7	Barkly 3	4.6-5.8 (Mitchell grass country)
Property D Paddock 6	Mostly A & B with small areas of C near waters	22	5.1	Barkly 1 Barkly 2 Wonorah	4.6-5.8 (Mitchell grass country)
Property D Paddock 1	Mostly A and B	23	6.8	Barkly 1	4.6-5.8 (Mitchell grass country)
Property D Paddock 4	Mostly A & B with small areas of C near waters	29	7.3	Barkly 2 Wonorah/Barkly 1 Wonorah Barkly 1 Yelvertoft Barkly 3	1.2 (hills)- 4.6 (broken Mitchell grass country)
Property E	Mostly B with areas of C near waters	30	9.2	Barkly 2 Barkly 3	4.6-5.8 (Mitchell grass country)
Property D Paddock 5	Mostly A & B with small areas of C near waters	32	7.1	Barkly 1	4.6-5.8 (Mitchell grass country)
Property D Paddock 2	Mostly A and B	33	5.2	Barkly 1	4.6-5.8 (Mitchell grass country)
Property F Paddock 1	Mostly B with some areas of A. Areas of C near waters and creek lines	45	4.9	Austral Kallala	4.6-5.8 (Mitchell grass country)
Property F Paddock 2	Mostly B with some areas of A. Areas of C near waters and creek lines	142 (heavily skewed by the very poor 2007/08 wet season) when this year is excluded, the average UR remains high at 40	5.1	Austral Wonardo	4.6-5.8 (Mitchell grass country)

Overall, the evidence indicates that stocking rates that result in an average utilisation rate of up to 30% appear to be sustainable on uniform, cracking grey clay soils supporting Mitchell grass in the Barkly region. However, this level of utilisation negatively impacts on less robust soil types and on preferred areas, such as creek lines and bluebush swamps in paddocks with a mix of pasture types. In general, the best performing paddocks in terms of land condition had utilisation rates of less than 25%. The only exception to this was Paddock 2 on property D, which had five early wet-season spells in ten years.

Katherine region

Utilisation rates were calculated for one paddock in the Sturt Plateau region for the period spanning July 2003 to July 2009. This relatively small paddock (30 km²) fell entirely within the Forrest land system and had a bore near its centre, which meant that the entire paddock was within 5 km of water. As expected, the average annual pasture growth was higher in the Katherine case study paddock (1600 kg/ha) than in the other regions. The land in the Katherine case study paddock was mostly condition A with some condition B close to waters and the average utilisation rate of 7% was low compared with the current agency recommendation of 15% for productive red soils. The average stocking rate during the study period was 3.8 AE/km², which is consistent with DoR recommended carrying capacities for gravelly red soils in the Sturt Plateau region. More paddocks in the Sturt Plateau region will be studied in late 2010.

Grazing trials

Utilisation rates are currently being calculated for all relevant DoR grazing research trials and will be included in the final report for the project at the end of December 2010.

This project is supported by external funding from the Australian Government's *Caring for our Country* Project OG082924: Land type specific sustainable practice guidelines for NT pastoral lands.

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PROJECT: Newcastle Waters Rotational Grazing Trial

Project Officers: K. Scott and R. Cowley

Location:	Barkly
Keyword(s):	cell grazing, Newcastle Waters, cattle, rotational grazing, pasture utilisation, stocking rates

Objective:

To compare the effect of rotational grazing (with wet season spelling) and continuous grazing in the Barkly region through an assessment of pasture composition, animal production and ease of application.

Background:

Newcastle Waters Station established a rotational grazing trial in 2002. The Department of Resources (DoR) conducted assessments of pasture composition and animal production in 2003-05, to determine whether such a strategy has advantages over continuous stocking in the extensive beef cattle properties of northern Australia.

Method:

Rotational grazing was employed in two paddocks divided into cells, while continuous stocking was employed in one control paddock. Brownies comprised four cells and Langlands comprised 10 cells. Cattle were rotated within the two cell grazing paddocks on a regular basis, though without firm 'rules' as to the direction of rotation or period of grazing. Assessments of pasture composition were conducted in the early (May) and late (October) dry season during the study period (2003-05). The weight gain of indicator weaners was assessed at the first and second round musters and live-weight gain was subsequently calculated.

Results:

A detailed report on this project is being produced. A brief summary of the findings include the following:

- Although initially set up as a cell grazing system, the inability to access paddocks during the wet season required them to be rested during the wet season and rotated between cells only in the dry season. Hence the treatment grazing strategy can be better described as a wet season spell with dry season rotation, rather than a cell-grazing strategy *per se*.
- Season and inter-annual variability (and fire in the last year of the trial) had a very strong effect on the grazing score (i.e. grazing activity), cover and yield.
- There was no significant effect of treatment (cell grazing vs. continuous stocking) on grazing score, cover and yield or species composition.
- There was no effect of treatment on diet quality.
- Live-weight gain was lower in cattle in cell-grazing paddocks than in the continuous grazing paddock, in both years (significantly so for the 2004-05 season).

Overall, the study suggested rotational grazing is a viable strategy in these rangelands. However, a full economic comparison still needs to be done.

PROJECT: Shruburn – Rangeland Burning Trial

Project Officer: T. Cowley

Location: Katherine

Keyword(s): fire, rangelands, woody thickening, pasture, cattle

Objective:

To investigate the efficacy of using fire to control woody thickening on pastoral land - specifically to identify fire frequencies and intensities that are effective in altering or maintaining woody vegetation structure.

Background:

Fires are an important component of grazing systems. Fire frequency has a strong influence on land condition and native woody plant cover and density, which in turn influence carrying capacity. Long-term impacts of regular fire frequency on land condition and woody cover are unknown. This project provides an important understanding of how fires can be used to manage woody cover and maintain carrying capacity. This is particularly relevant given the increasing rainfall over the last 20 years, which has been linked to increasing native woody cover in the region. Fire is the only practical tool to manage woody cover.

Climate change models predict that woody cover will increase under higher carbon dioxide levels. As for rainfall-driven woody cover increase, fire is the only tool that can practically manage it as a result of climate change.

This is one of the few long-term fire trials internationally and may assist global climate change modelling. For this reason, it has been identified as a high priority project for continuation in future.

Method:

The study site is on Victoria River Research Station in the central Victoria River District (VRD). It is composed of two trial sites on different soil-pasture communities within separate grazed paddocks. One site is a ribbon-blue grass (RBG) pasture on grey cracking clays dominated by *Terminalia volucris* (rosewood) and *Lysipyllum cunninghamii* (bauhinia). The other is an arid short (ASG) grass pasture community on red soil, which is dominated by *Eucalyptus terminalis* (inland bloodwood), *Eucalyptus pruinosa* (silver leaf box), *Carissa lanceolata* (conkerberry bush) and *Hakea aborescens*.

Each trial site is composed of 16 plots (100 m x 100 m) in a 4 x 4 design. Season of burn includes early dry season (June) vs. late dry season (October) and frequency of burning includes never, every two years, every four years or every six years. Each site has two plots for each treatment (see the plot map in Appendix 1).

Pasture sampling is carried out every two years to determine the impact of different fire regimes on pasture yield and composition. Woody vegetation assessments (specifically tree basal area and canopy cover) were carried out in 2009 (previously they were measured in 1999).

Results:

Woody vegetation
Site
The red soil site had double the canopy cover (18% vs. 9%, *P*<0.001) and tree basal area (TBA) (3 m^2/ha vs. 1.2 m^2/ha, *P*<0.001) of the grey cracking clay site, but significantly less ground cover (63% vs. 76%) and pasture yield (1385 kg/ha vs. 2012 kg/ha).

Burning season
On the black soil site both early and late dry season fires are effective in reducing woody vegetation compared with the control site, although late fires have been more effective. This suggests that in ribbon-blue grass pastures in the VRD, early dry season fires can be used to control woody thickening, avoiding the more dangerous late dry season fires. This finding is important because land managers are reluctant to light fires late in the dry season due to risks associated with controlling them.

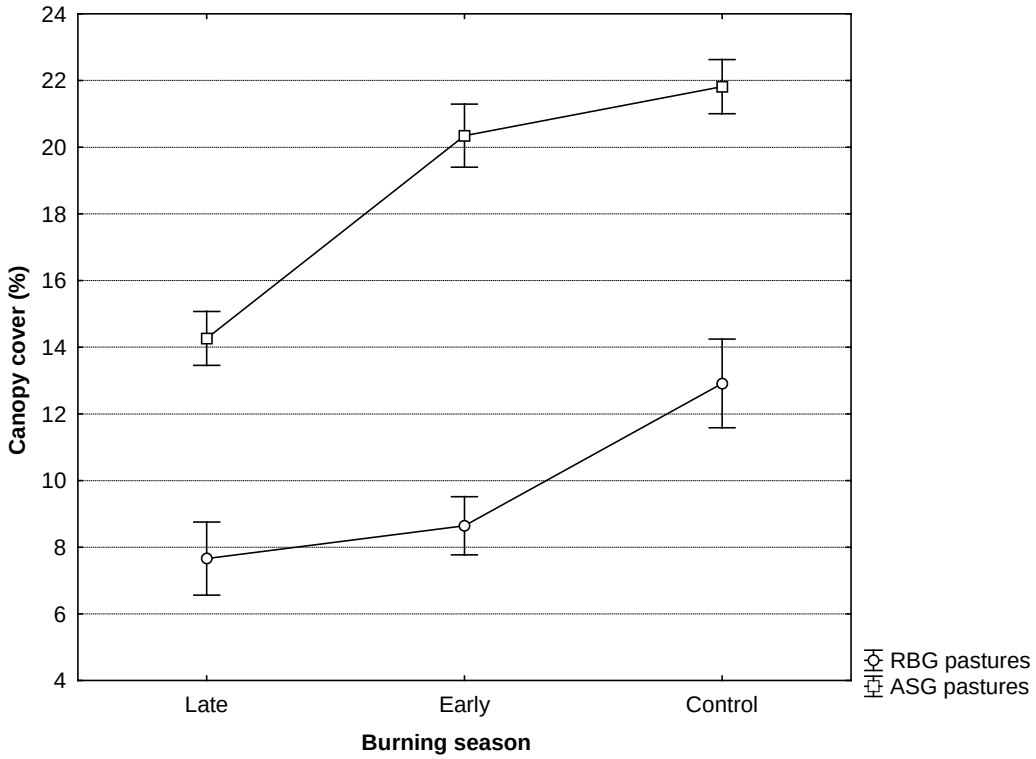

Figure 1. Effects of burning season on canopy cover

At the red soil site, the results showed that only late dry season fires significantly reduced woody vegetation ($F_{1,121}$=7.02, *P*<0.01).

Burning frequency
Burning frequency has had the same impact on woody vegetation on both pasture communities. Burning once every six years did not reduce woody vegetation compared with the control, while burning every two or four years was similarly effective in reducing woody vegetation (see Figure 2). This indicates that a burning frequency of four years is required to manage woody thickening on these pasture communities, which is more frequent than currently recommended (five to seven years).

Pastures

Ground cover was not affected by season of burn or frequency; however, pasture yield was. On both soil types, early dry season burning resulted in significantly less pasture yield (1400 kg/ha) than late dry season burning (1870 kg/ha) or no burning (1890 kg/ha). The differences were not significantly different ($F_{1,114}$=18.02, P<0.001). Grazing animals were not excluded from the burning plots. It appears that post-fire grazing on green pick after early burns has had a negative impact on pasture growth. Frequency of burning significantly affected only pasture yield on ribbon-bluegrass pastures - burns every two years resulted in greater pasture yield (2460 kg/ha) than four (1640 kg/ha) or six year burning frequencies (1820 kg/ha). At first glance, it appears that this was a consequence of reduced competition from woody species. However, since burning frequencies of two and four years yielded similar canopy covers, this does not appear to be a plausible explanation. The pasture data is still being analysed to determine the impact of different fire regimes on individual pasture species and groups. It should be noted that all treatments experienced a reduction in total pasture yield since the beginning of the trial, including the control. This suggests that grazing on the fire plots is having an influence. Further investigation is required to determine what is going on. Pasture sampling at the paddock level is being carried out for other research projects and will be used to determine whether it is a paddock-wide trend or limited to the fire plots.

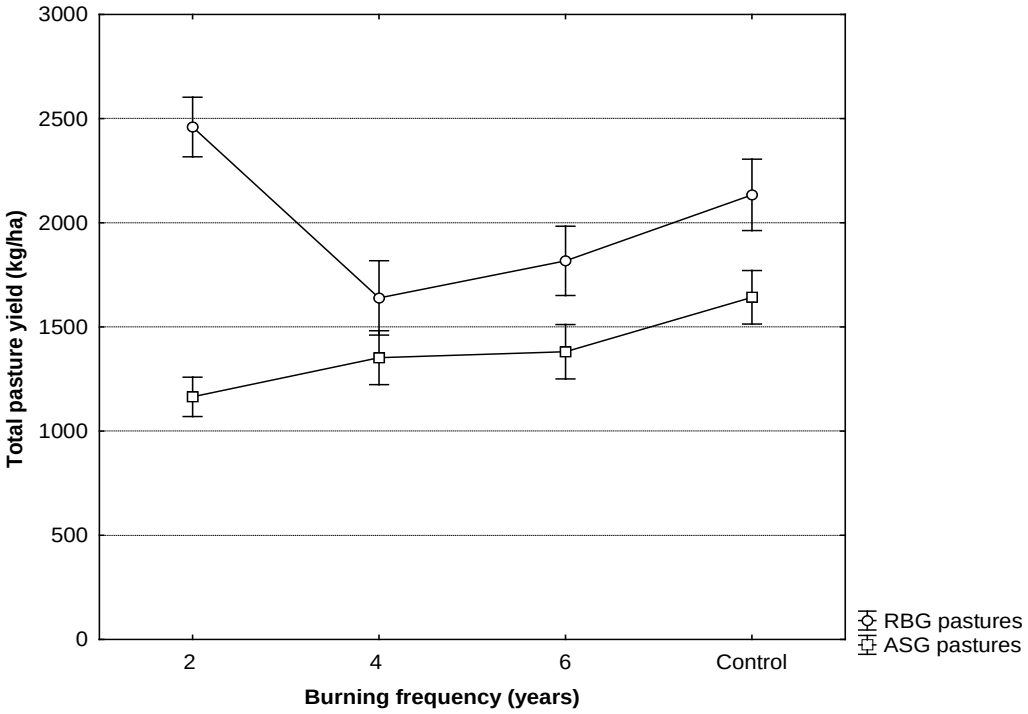

Figure 2. Impact of burning frequency by soil type on total pasture yield

Appendix 1: Shruburn trial layout – treatment allocations

Rosewood paddock

1 L6	**2** E4	**3** E4	**4** E6
5 E6	**6** L4	**7** L4	**8** 0
9 0	**10** L2	**11** L6	**12** E2
13 0	**14** E2	**15** L2	**16** 0

N ↑

Conkerberry paddock

17 L6	**18** 0	**19** E6	**20** L4
21 0	**22** E4	**23** E2	**24** L2
25 E6	**26** 0	**27** E4	**28** E2
29 0	**30** L4	**31** L2	**32** L6

N ↑

NB: E = early burn, L = late burn, the numbers indicate the frequency of burning.

PROJECT: Pasture Sustainability Monitoring at Kidman Springs

Project Officers: T. Cowley and C. Pettit

Location: Katherine

Keyword(s): rangelands, woody thickening, pasture, carrying capacity, cattle

Objectives:

To further monitor pasture at Victoria River Research Station (VRRS).

To monitor whether the Cowley and Bryce (2003) stocking rates are sustainable.

To determine the differences between paddocks regarding pasture quality and to monitor their seasonal changes.

Background:

Matching stocking rates to the estimated sustainable carrying capacity of paddocks is a management practice routinely recommended to achieve optimal production and still maintain land condition. The Department of Resources regularly provides advice regarding carrying capacity at land type, paddock and property levels. The methodology for calculating carrying capacities is based on current knowledge of sustainable utilisation rates and median pasture growth estimates derived from research sites on relatively uniform land types. However, little research has been conducted in paddocks with a mix of land types. This project monitors several indicators of pasture condition in order to further evaluate the carrying capacity methodology.

Furthermore, various cattle projects are being researched at VRRS. The pasture condition dataset provides valuable information on pasture quantity and quality in research paddocks, which is used to interpret cattle research trials. It provides an understanding of the relative seasonal conditions experienced during trials, as well as forage availability and quality in different paddocks.

Method:

Since 2003 pasture condition has been annually assessed at the end of the growing season in all breeder paddocks at VRRS. In each paddock, data is collected at fixed points, which are distributed according to the proportion of each soil type in each paddock. The Botanal method is used and data is collected on the variables listed in Table 1. Observer yield estimates are calibrated using material harvested, dried and weighed each day.

Table 1. Variables recorded

Variable	Definition
Observer	Initials of person making observations.
Paddock	Name of paddock.
Point	Point number. Range: 1-6
Site	Site number. Range: 1-5
Quadrat	Quadrat number. Range: 1-5
Yield	Estimate of total standing dry matter in quadrat (kg/ha).
Top four species	Record the four species which contribute the most to the total yield of the quadrat. Enter species codes from species list.
Composition of top four species	Estimate the percentage of the total biomass that each species makes up. The sum of these must equal 100%.
Total ground cover	% of ground covered by organic matter (e.g. plants, leaf litter, biological crusts, manure etc).
Defoliation	% of total biomass (e.g. plants) that has been removed due to grazing. 0%, 1-5%, 5-25%, 25-50%, 50-75%, 75-100%.
Soil type	Basic assessment of broad soil type within quadrat. Two choices: Red and black
History of fire	Has there been a fire in the last six months? Y/N
GPS position	Upload GPS position.

Results:

Figure 1 depicts the 2010 average total standing dry matter yields for each breeder paddock at VRRS in 2010. Differences between paddocks are largely related to soil type; however, an in-depth analysis of pasture condition trends, utilisation rates, stocking rates and animal performance from 2003 to the present will be completed in 2011. This analysis will determine whether the paddock carrying capacities derived by Cowley and Bryce (2004) are sustainable in the long-term and provide recommendations for refining the methodology for calculating carrying capacities in paddocks with a mix of land types.

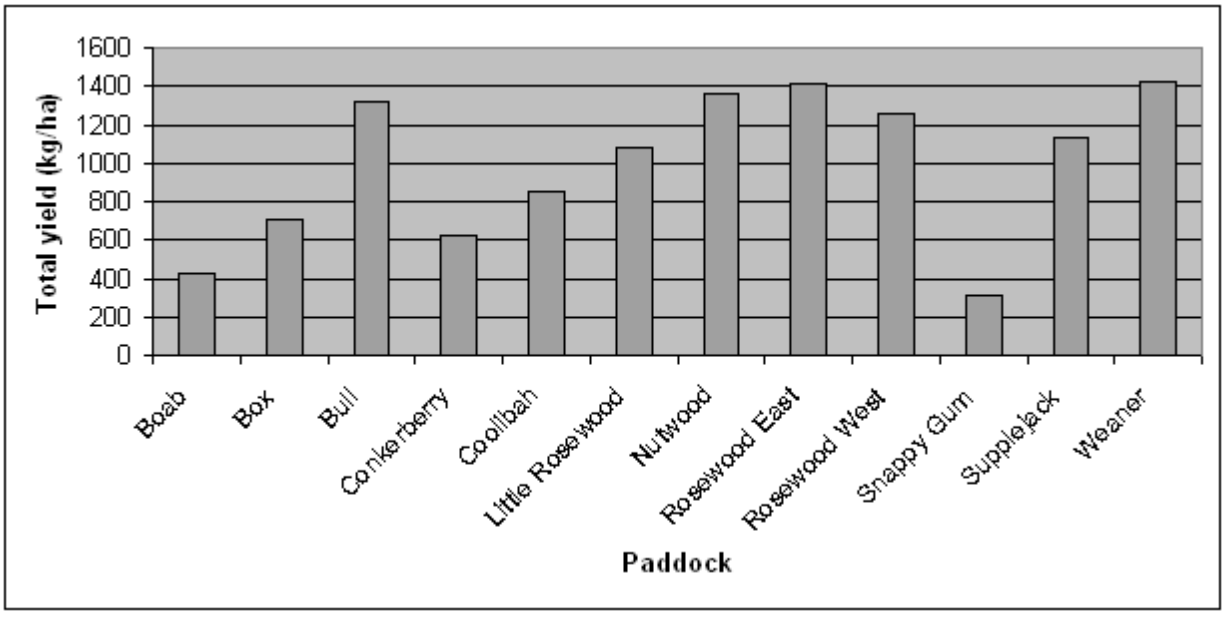

Figure 1. Average total standing dry matter by paddock at VRRS, 2010

Reference:

Cowley, R. and Bryce, D. (2004). Kidman Springs Carrying Capacity. Estimation using pasture growth models and estimates of animal intake. Department of Resources Internal Report.

PROJECT: Viral and Endogenous Retroviral Detection and Characterisation in Crocodiles

Project Officers: L. Melville, S. Davis, R. Weir and C. Shilton

Location: Darwin

Keyword(s): crocodiles, viruses, diseases

Objectives:

To define the best conditions for reliably producing primary cells lines from Crocodylus porosus.

To develop methods for the maintenance and cryopreservation of primary C. porosus cell lines.

To investigate the viral fauna of farmed and wild C. porosus in the Northern Territory.

To determine the feasibility and need for the development of continuous cell lines from primary C. porosus cell lines.

To investigate the role of endogenous retroviruses in runting and other diseases.

Background:

This project has been developed in response to findings from RIRDC project NAP05-16. One objective of Project NAP05-16 was to conduct a histopathology study to determine if there were any gross histological differences or pathological reasons for runting. No evidence was found to suggest that bacterial or fungal infections were causing runtism. However, lymphoid atrophy in several tissues (particularly the thymus and tonsils) as a secondary effect of viral infection could not be eliminated. Viruses are not well characterised in crocodilians because, until recently, no crocodile cell lines had been established and no crocodilian viruses would grow on other established cell lines. Development of a crocodile-specific cell line will allow the isolation and characterisation of crocodilian viruses as well as allow antigen preparation for serological testing and, eventually, the manufacture of specific vaccines. Furthermore, the characterisation of endogenous retroviruses (ERVs) could also provide evidence of infectious agents that have been associated with disease, pathogenesis, and immune system reduction, and associated disorders in other species. Endogenous retroviruses are copies (or remnants) of exogenous retroviruses that have integrated into a host cell genome at some stage. The present study aims to characterise ERVs and assess their levels of expression, particularly in runts. ERV characterisation is important in the context of aetiology and zoonosis of animal diseases. It will also allow potentially novel functional and non-functional retro-elements to be identified, whilst gene expression will establish which ERVs are functional and potentially transmissible. This information will allow associations to be drawn regarding the possible role of crocodile ERVs in causing runtism and other diseases.

Method:

Sixteen *C. porosus* cells have been established and maintained. The cell lines are from six tissue types: kidney (3), liver (1), subcutaneous tissue (1), heart (5), lung (4) and trachea (2). The growth conditions, growth media and

passaging procedures have been defined. All cells are grown at 30 °C in medium 199 plus 10% foetal bovine serum, 25 ng/mL epidermal growth factor and antibiotics (penicillin, streptomycin, amphotericin).

Diagnostic specimens have been collected from *C. porosus* submissions made over 2005-2010 and stored at -70 °C for later processing for virus isolation in *C. porosus* cell lines. The following protocol has been established for processing these samples. First, tissue is homogenised in viral transport medium and then filtered through a 0.45 micron filter to exclude any bacterial or fungal contamination. Clarified tissue supernatant is then inoculated into at least 2 different *C. porosus* cell lines growing in 25 cm^2 flasks. Cells are observed at three-day intervals for evidence of virus induced cytopathic damage. All cultures are passed into new flasks at 21 days and observed for a further 21 days until discarded if no evidence of viral growth is observed.

Results:

Cell lines can be maintained for extended periods without passage, up to four months. Minimum plating efficiency has been established for some cells lines (kidney, heart, trachea and lung) with the minimum plating density of 0.375-0.5 x 10^5 cells/mL to achieve confluence in seven days and 0.075-0.25 x 10^5 cells/mL to achieve confluence in 14 days.

Four hundred and seven samples have been processed for virus isolation. This has yielded 83 virus isolates. Ten virus isolates have been characterised by electron microscopy at CSIRO, Australian Animal Health Laboratory as belonging to the Herpesviridae family. Further characterisation of the remaining isolates will be given a high priority in 2010. The virus isolation program will also continue, targeting samples from submissions with suspect viral aetiology.

The sentinel hatchling crocodile program, which aimed to isolate arboviruses, was completed for 2009 with no further viral isolation. This program was resumed in 2010, with no viruses isolated to date. The genetic analysis work being conducted at Sydney University has led to the identification of a new endogenous retrovirus (ERV) type. Additional characterisation of ERVs from a large number of specimens is underway.

PLANT INDUSTRIES

Plant industries projects conduct applied research in controlled trials to discover solutions to problems that affect productivity and profitability of the industry and, where possible, to protect the environment and human health

PROJECT: Preliminary Evaluation of the Uptake of Foliar Nitrogen Applied to Mango Trees

Project Officers: D. Hamilton, C. Martin, M. Hearnden, M. Bennett and N. Hartley

Location: Darwin

Keyword(s): mango, nitrogen, foliar, ^{15}N,

Objective:

To determine how much nitrogen (N) from urea and potassium nitrate foliar applications was absorbed and contributed to total leaf N.

Background:

This study is the Northern Territory (NT) contribution to the Horticulture Australia Limited (HAL) Australia-wide project: "Delivering Mango Technology" (DMT). It is designed to improve mango productivity and was based on grower requests to investigate the use of N fertilisers to minimise wastage and increase profitability.

Mango growers in the NT use foliar application of N to boost productivity. This can increase fruit-set and yield of mangoes (Oosthuyse 1993; Yeshitela 2005). However, Nguyen et al. (2004) have shown that a high proportion of skin greenness and anthracnose disease on ripe mangoes was associated with foliar application of N. But foliar, rather than soil application, offers the benefit of rapidly boosting N levels in leaves when requirements for N are high. However, the amount of N uptake from foliar application has not been assessed.

Method:

Plant material and site characteristics
Details are the same as for the other DMT N trial conducted at the same orchard, which is described elsewhere in this publication.

Treatments
Separate solutions containing 2% (w/v) urea (10.0% ^{15}N enriched), 2% (w/v) potassium nitrate (KNO^3) (10.0% ^{15}N enriched) and distilled water each with 1% Triton X were applied once (day 1), or twice (day 1 and 2) separately to single tree plots. ^{15} N provides a direct measure of absorption of N. Four leaves per tree were dipped in each solution. Each leaf was located at a mid point on a shoot (approximately 30 cm long) on a separate branch on the north, south, east and west sides of the tree. The age of the dipped leaves was approximately two or six months, determined by the age of the shoot. The solution was weighed before and after dipping to calculate the application rate. Six single-tree replicates per treatment were used in a Latin square design. The treatments were applied within a week before the trees commenced terminal bud-burst with subsequent flowering.

Plant sampling and nutrient analysis

A day before treatment, leaves were sampled to determine background ^{15}N concentrations. Twenty-four hours after each treatment, the leaves were sampled again; at the same time, two untreated leaves (one on either side of the dipped leaf) were sampled to determine the movement of ^{15}N from the treated leaves.

To determine N uptake into the leaf, surface residue N was removed using the method of Silcox and Holloway (1986). All samples were ground through a 0.2 mm sieve. Samples for ^{15}N isotope ratio analysis were analysed by mass spectroscopy as described for the SANT experiment.

To estimate total uptake per tree, an estimate of leaf biomass was needed. This was achieved by counting one-half of the leaves of a tree of similar size, doubling it and determining leaf dry weight.

Results:

Before the treatments were applied, average leaf N ranged from 1.3 to 1.4%. The second dip of urea significantly increased the absorption of N compared with the second dip of KNO³. In contrast, the percent contribution of applied N to total leaf N doubled as a result of the second dip for both N fertilisers. The greater contribution of urea to total leaf N was due to its higher concentration of 0.92% N in the dipping solution, compared with the KNO³ concentration of 0.28% N.

Table 1. The proportion of applied N absorbed in the leaf and the contribution of applied N to total leaf N from one and two leaf applications of KNO³ and urea ($P \leq 0.05$)

Treatment	Applied N absorbed in the leaf (%)	Applied N's contribution to total leaf N (%)
KNO_3 (1 dip)	28.0 ab	1.3 a
KNO_3 (2 dips)	26.0 a	2.6 b
Urea (1 dip)	31.5 ab	5.3 c
Urea (2 dips)	39.8 b	10.2 d

Values followed by the same letter are not significantly different at P=0.05

Table 2 shows that the number of dips (applications) did not affect the level of leaf N percentage or total leaf N. The N absorbed from foliar application after two dips approximately doubled the amount absorbed after only one dip. The amount absorbed was also higher for urea. These results were expected because they were calculated using a leaf biomass (dry weight) of 12.6 kg and the figures in column 3 of Table 2 were multiplied by the numbers in column 3 of Table 1.

Table 2. The proportion leaf N and estimated total N in leaf biomass and the N absorbed from foliar N application after one and two leaf applications of $KN0_3$ and urea ($P \leq 0.05$)

Treatment	Leaf N (%)	Total leaf N (g)	Applied N (g) absorbed from foliar N
KNO_3 (1 dip)	1.4	176	2
KNO_3 (2 dips)	1.4	176	4
Urea (1 dip)	1.5	188	10
Urea (2 dips)	1.5	188	19

These results indicate that 26% to 39% of applied foliar N is absorbed into the leaf. Doubling the number of applications, 24 hours apart will double the contribution of applied N to total leaf N, whether it be from urea or KNO_3. Commercial applications of N may be less efficiently absorbed; however, repeat applications may significantly improve mango productivity.

References:

Nguyen, H., Hofman, P., Holmes, R., Bally, I., Stubbings, B. and McConchie, R. (2004). Effects of nitrogen on the skin colour and other quality attributes of ripe 'Kensington Pride' mango (*Mangifera indica L.)* fruit. *Journal of Horticultural Science & Biotechnology,* **79:** 204-210.

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Acknowledgement:

This project was partially funded by HAL, in partnership with the Australian Mango Industry Association and voluntary contributions by the industry. Thanks to David Joyce for providing the orchard for the trial and for other support. Thanks to all staff at Berrimah Farm who assisted with the work, in particular Sue Willoughby, Graeme Patch and Sean Bithel. Thanks to Dr David Huett of the Australian National University and several other experts in ^{15}N work who assisted with calculations and provided advice. The support from the NT Mango Industry Association and Queensland's Department of Employment, Economic Development and Innovation is appreciated.

PROJECT: The Effect of Leaf Nitrogen Concentration and Time of Nitrogen Application on Nitrogen Uptake and Growth Response in Mango Trees

Project Officers: D. Hamilton, C. Martin, M. Hearnden, M. Bennett and N. Hartley

Location: Darwin

Keyword(s): mango, nitrogen, ^{15}N, uptake, yield, quality, growth

Objective:

To determine how yield, quality and nitrogen (N) fertiliser uptake are affected by the time of N application and N levels in leaves at the time of application.

Background:

This study is the Northern Territory (NT) component of the Horticulture Australia Limited (HAL) project, "Delivering Mango Technology" designed to improve mango productivity in Australia. Growers requested an investigation on the use of N fertilisers to minimise wastage and maximise profitability. The work was conducted during one mango

season. In a review of the prospects for manipulating horticultural crops through N fertiliser application, Huett (1996) concluded that it was unlikely that manipulation could increase orchard productivity. The main highlighted reason was that new growth depends mainly on internal reserves of N in the tree. This study aimed to evaluate this premise and further investigate the role of N nutrition.

Method:

Twelve-year-old Kensington Pride mango trees, which were planted at a spacing of 7 x 12 m, were selected on a commercial orchard near Darwin. The soil was a Tenosol loam. Tree size was approximately 3.2 m high and 4.5 m wide.

The experiment was a randomised complete block, two-way factorial, with two leaf N treatments: high leaf N (HLN) and low leaf N (LLN) with two N applications and a control (post-harvest, December 2008; pre-flowering, April 2009; zero N - control). There were six replicates of single-tree plots with guard trees separating treatments within rows. HLN trees had an average leaf N level of 1.2% and LLN trees had 1.0 to 1.1% N.

N fertiliser was applied at 200 g of N/tree as urea containing added ^{15}N at 2.10 atom% excess (^{15} N content above atmospheric N_2). The fertiliser was dissolved in distilled water and watered in evenly in a 0.6 m radius from the trunk. Immediately after application, water was applied to minimise volatilisation losses.

Leaf samples were collected at approximately five and eight weeks after each N application to determine total N content and the percentage of N derived from the fertiliser (urea).

The proportion of shoot initiation was recorded on 30 tagged terminals/tree. Fresh fruit yield was recorded and fruits were sub-sampled for quality assessments. Yield efficiency was measured as kg fresh weight/cm^2 trunk cross-sectional area. Above-ground tree parts (one from each treatment) were removed from 30 September to 2 October, 2009. They were dried to determine fertiliser N uptake and recovery (of fertiliser rate used) in all aboveground parts of the tree.

Results:

Table 1. Significant effects ($P \leq 0.05$) of leaf N level, time of N fertiliser application and their interaction on various response parameters

Treatment	Parameter	Response
Leaf N level	Yield efficiency	High leaf N > low leaf N.
	Unmarketable small fruit (kg)	High leaf N > low leaf N.
	Shoot initiation (%)	High leaf N > low leaf N.
	Dry matter (DM) in fruit flesh (%)	High leaf N > low leaf N.
	Total soluble solids (TSS) in fruit flesh	High leaf N > low leaf N.
	N in fruit flesh (%)	High leaf N > low leaf N.
	N in fruit skin (%)	High leaf N > low leaf N.
	Total N in whole fruit yield (g)	High leaf N > low leaf N.
	Calcium in fruit flesh (%)	Low leaf N > high leaf N.
	Potassium (K) in fruit flesh (%)	High leaf N > low leaf N.
	Magnesium (Mg) in fruit flesh (%)	High leaf N > low leaf N.
	Total K phosphorus (P), Mg and sulphur in fruit flesh (g)	High leaf N > low leaf N for K, P, Mg & S.
Timing of N application	Fruit size: length of marketable fruit (mm)	Post-harvest N = zero N > pre-flowering N.
	N in fruit flesh (%)	Pre-flowering N > post-harvest N > zero N.
	N in fruit skin (%)	Pre-flowering N > post-harvest N > zero N.
	N in fruit flesh, seed and skin derived from fertiliser (%)	Pre-flowering N > post-harvest N > zero N.
	Proportion of yellow skin on fruit (%)	Pre-flowering < post-harvest < zero N.
Timing x leaf N interaction	N in whole fruit derived from fertiliser (%)	The increase from zero N application to pre-flowering N > for low leaf N.
	N in fruit flesh, skin and seed derived from fertiliser (%)	The increase from zero N application to pre-flowering N > for low leaf N.
Timing x leaf N x date interaction	Shoot initiation (%)	After application of N, % shoot initiation in high N trees declined relative to low N trees.
	Leaf N derived from fertiliser (post-harvest) (%)	In zero N no difference between high leaf N and low leaf N trees, whereas in post-harvest high leaf N > low leaf N.
	Leaf N derived from fertiliser pre-flowering) (%)	In zero N no difference between high leaf N and low leaf N trees, whereas for pre-harvest low leaf N > high leaf N.

Table 2. Whole tree above-ground uptake and recovery of fertiliser N

Leaf N and time of application	Whole tree N content (g/tree)	Whole tree N derived from fertiliser N (g/tree)	Fertiliser N to tree N (%)	Recovery of applied rate of fertiliser in the tree (%)
LLN Post-harvest N	594	6.7	1.1	3.3
HLN Post-harvest N	613	5.9	1.0	1.9
LLN Pre-flowering N	578	7.8	1.4	3.4
HLN Pre-flowering N	805	6.2	0.8	2.5

Table 3. The contribution of fertiliser N (%) to the total N content (g) in various above-ground parts of the tree (n=1)

Leaf N and timing	Trunk + stems	Leaves	Flowers	Fruit
LLN post-harvest N	2.8	1.4	3.2	3.3
HLN post-harvest N	2.1	1.6	2.3	2.3
LLN pre-flowering N	2.3	2.0	4.7	5.4
HLN pre-flowering N	1.3	1.0	2.8	3.0

Discussion

Yield efficiency was highest in HLN trees (Table 1) indicating that they had more photosynthesis per unit leaf area for fruit yield. This is supported by Dejong (1982) who reported a direct correlation between leaf N content and photosynthesis in peach trees. HLN trees also had a larger percentage of small (unmarketable) fruit (Table 1) indicating that more fruit was set than could be effectively supplied with carbohydrate. This is supported by the review of Lechaudel and Joas (2007) which stated that one of the most important pre-harvest factors affecting mango fruit growth was the availability of carbohydrates.

The proportion of yellow skin on ripe fruit decreased (increased proportion of green skin) the closer N was applied to flowering (Table 1). This is supported by the work of Nguyen et al. (2004) who showed that the proportion of green skin on ripe mango fruit was associated with tree leaf N levels. That is, N application close to flowering and higher rates of N application increased greenness. The importance of N to greenness was demonstrated in our results which showed that the percentage N in fruit flesh and skin increased the closer N was applied to flowering, and was higher in HLN trees (Table 1). This was despite the higher uptake of fertiliser N by LLN trees in all parts of the fruit (Table 1). There appears to be no previous study showing a possible link between the actual proportions of N in the skin on skin greenness in response to time of N application.

The estimated N uptake in the above-ground part of the whole tree was greater in LLN trees (Table 2). Similar results were found in citrus (Feigenbaum et al. 1987). Pre-flowering N application (closer to flowering) resulted in higher uptake of fertiliser N (Table 2). This was similar to that found in low-chill peach (Huett and Stewart 1999) and in pears (Sanchez et al. 1992). The pre-flowering application of N contributed a larger percentage of fertiliser N to total N in the fruit and flower parts, in contrast to the post-harvest application of N that contributed a larger percentage of fertiliser N to total N in the vegetative parts (Table 3). Stassen et al. (2000) found a similar result, but did not describe the portion contributed by fertiliser N application.

These results indicate a well-managed orchard with adequate leaf N levels can achieve a higher yield efficiency and improved fruit quality when N fertiliser is applied to meet the requirements of mango trees.

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Acknowledgement:

This project was partially funded by HAL, in partnership with the Australian Mango Industry Association and voluntary contributions from the industry. Thanks to David Joyce for providing the trial orchard and other support. Thanks to all staff at Berrimah Farm who assisted with the work, in particular Chelsea Moore, Sue Willoughby, Sean Bithel, Peter Bergin and Graeme Patch. Thanks to Dr David Huett and several other experts in the field of ^{15}N work who assisted with calculations and advice. The support of the NT Mango Industry Association and the Queensland Department of Employment, Economic Development and Innovation is appreciated.

PROJECT: Mango Root DNA Detection – a New Tool for Applied Research

Project Officers: S. Bithell and L. Tran-Nguyen

Location: Darwin

Keyword(s): mango, root detection, root distribution

Objective:

To evaluate the potential of a method to quantify mango root DNA in soil in order to facilitate root distribution research.

Background:

Although roots are responsible for all water and most nutrient uptake in mango trees, their function is poorly understood. Information on mango root profiles is essential for developing more accurate irrigation practices to maximise water use efficiency in this crop in the Northern Territory (NT). DNA-based root tools are capable of quantifying the distribution and contribution of fine roots normally lost during soil sieving and distinguishing between dead and live roots in other crops (Riley et al. 2009).

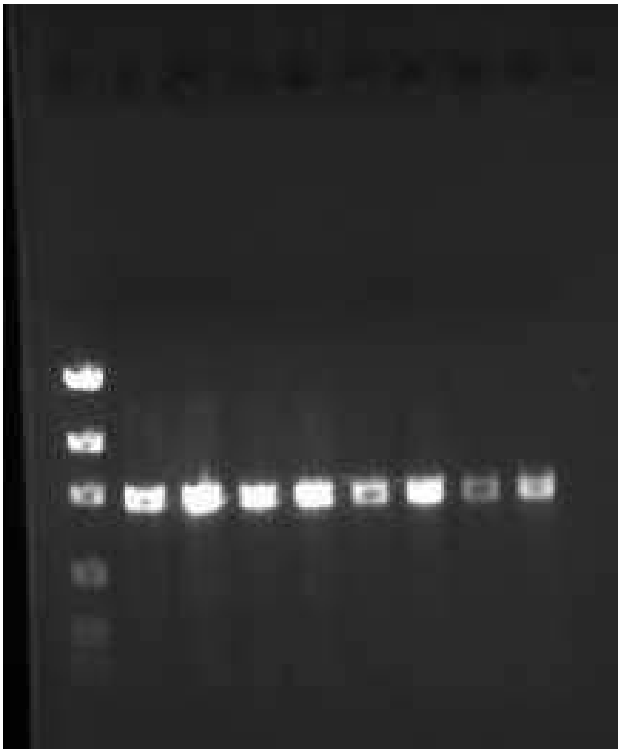
The NT Research and Innovation Board funded a trial for a 'proof of concept' to evaluate the potential of this method. The project was started in April 2010 and will be completed in 2011. The South Australian Research and Development Institute, which has expertise in the extraction and rapid quantification of plant DNA from large soil samples (300-500 g) (McKay et al. 2008), is a key collaborator.

Method:

DNA extraction techniques from mango root and leaf material will be evaluated, including root samples from a range of rootstocks, refinement of extraction techniques, trialling polymerase chain reaction primers and sequencing, determining conserved regions suitable for probe design, trials with mango roots mixed with soil and comparing with other plant DNA (from plants commonly found in mango orchards), in order to determine specificity. If this initial research is successful, soil samples from field-grown trees, such as mango rootstocks, will be quantified. Depending on findings, a review will determine our capability to develop a quantitative mango root DNA method for applied research.

Results:

Initial results are promising (Figure 1). The extraction of DNA from ten rootstocks was successful. Sequencing has focussed on conserved regions (ITS 1 and 2 and specific regions within) in order for the method to be used with a range of rootstocks. The sequence of local Nam Doc Mai root sample was analysed using the BlastN search engine on GenBank (http://www.ncbi.nlm.nih.gov/). Results indicated that it was 100% similar to a previously sequenced Nam Doc Mai variety, GenBank accession number AB071672. This has provided a suitable control to compare with a range of previously un-sequenced mango rootstocks. The identification of suitable sequences for a range of rootstocks is challenging due to the polymorphic nature of mango DNA (Yonemori et al. 2002). DNA has been successfully extracted from root samples of 15 non-mango species for use in specificity testing.



Lane number: 1 2 3 4 5 6 7 8

Figure 1. Amplified DNA extracted from a Kensington Pride mango leaf (lanes 1-4) and root material (lanes 5-8)

The project is still in the initial stages with further work to be completed on developing and trialling primers and probes, including evaluating the specificity and sensitivity of these methods, and detection and quantification of mango roots in soil samples. If these initial attempts are successful, it is planned to collect and analyse initial field samples from a long-term rootstock trial.

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PROJECT: Mango Fruit Borer (*Citripestis eutraphera)*: Diagnosis, Symptoms, Culture and Life History

Project Officers: B. Thistleton, L. Zhang, M. Neal and R. Meldrum

Location: Darwin

Keyword(s): mango, pests, *Citripestis eutraphera,* lepidoptera, pyralidae, artificial insect diet, insect culture, life history, morphological identification, molecular identification

Objective:

To identify a recently discovered pest of mangoes and develop a method for culturing the insect to observe its life cycle.

Background:

The mango fruit borer is the caterpillar of a small moth (*Citripestis eutraphera)* (Meyrick, 1933) (Lepidoptera: Pyralidae), which bores into mango fruit. The insect was originally described as *Phillotroctis eutraphera* from Indonesia (Meyrick, 1933) and has also been recorded from the Andaman Islands in India where it feeds on cashews (Jacob et al. 2004). In Australia, it was originally detected in two fruits from Darwin and Lambells Lagoon in 2008.

Method:

Diagnosis – morphological

Larvae were collected from two different locations, Berrimah and Lambells Lagoon, in August and October 2008. They were reared in captivity and the mounted adults were sent to the Australian National Insect Collection (ANIC) in Canberra for identification. Since the moth was a suspected Emergency Plant Pest, a second determination was required in line with national protocols. Dr Horak (Lepidopterist, ANIC) provided photographs of the male and female genitalia of this moth, which were emailed to Michael Schaffer, past curator of Pyraloidea, at the British Museum of Natural History (BMNH), London in March, 2009. Subsequent identification of moths collected from light traps or bred from surveyed fruit was made on the basis of genitalia dissections.

Diagnosis - molecular

DNA from one of the original confirmed *C. eutraphera* specimens provided by Andrew Mitchell (DPI NSW) was used as the reference positive control for diagnostic tests. The DNA was amplified in the Darwin laboratory and confirmed as identical to the sequence obtained by Andrew Mitchell. Two more *C. eutraphera* specimens were bred

from larvae in Java (Glenn Bellis, NAQS) and Lombok (Department of Resources ACIAR project) and DNA was extracted from the adults' legs. The DNA sequences were compared with the original specimen and confirmed to be 100% similar. The DNA analysis has therefore supported the morphological confirmation already made. Further details of this can be found in the report titled *Identification of exotic moths by DNA bar-coding* in this Annual Research Report.

Culture

Infested fruit collected from the field was kept in plastic containers to allow the larvae to complete their life cycle. Paper towels, vermiculite or sawdust were provided as a medium for pupation. After emergence, the moths were caged with mango fruit and provided with a 30% honey/water solution on absorbent wicks as food. Eggs on the mango fruit were allowed to hatch and the larvae burrowed into the fruit. Where large numbers of eggs were present, the larvae were transferred to other fruit for development.

Artificial diet

It was considered desirable to have a laboratory culture available for experimental work throughout the year. However, since mango fruit is not always available, an artificial diet was developed based on that previously used for *Helicoverpa* and cucumber moth in the Northern Territory and banana scab moth in Queensland (David Astridge, pers. comm.). The diet was poured into plastic containers and allowed to solidify. The larvae were then introduced to feed. Several different diets were tested with various nutrients and preservatives, including pulped mango from fruit frozen during the mango season.

Life history

Observations were made of the number and duration of the various stages. For duration, records were kept of time of egg laying, hatching, duration of the larval stage in mango fruit and the duration of the pupal stage. For the number of larval instars, it was easier to observe larvae feeding on thin layers of artificial diet, although the duration of the larval stage was longer.

Results:

Identity

Morphological and molecular studies confirmed that this species is the mango fruit borer (*Citripestis eutraphera)* (Lepidoptera: Pyralidae). This species was originally described from Java, Indonesia by Meyrick (1933)

Symptoms

In all fruit with larvae there is an obvious hole with frass and the fruit is often blackened around the hole. In many cases the fruit is also split; but it is not known if *C. eutraphera* caused the split or if it had laid eggs into an already split fruit. In varieties which have bunches of fruit together, the entry hole is often where the mangoes touch and the larvae join the fruit with silk. This damage would easily be seen in the field and damaged fruit are unlikely to be harvested.

Figure 1. Affected fruit

Culture

The larvae developed well on mangoes and the media was used for all the life history studies.

Artificial diet

Larvae died on the initial artificial diets, which were modified from *Helicoverpa* and cucumber moth diets with mango pulp added instead of pollen and rockmelon, respectively (see Table 1). It is thought that the larval death was due to the preservative used. When this preservative (methyl hydroxybenzoate – Nipagin M29908) was omitted, the larvae survived on the media but their development was slow and mould grew on the media within a few days.

The amount of nutrients was therefore increased in subsequent variations of the diet in an attempt to obtain development duration similar to that on mangoes. In addition, a different form of Nipagin (H3647) was sourced, which prevented mould development without killing the larvae. On the final two diets (numbers 5 and 6) mould did not develop, but larval development was still slow. Diet 5 contained pulped mangoes while diet 6 did not. The last generation on diet 5 failed to produce enough moths. Consequently, eggs and the culture of *C. eutraphera* were reduced to low levels. The culture will be replenished and studies of the artificial diet will be continued as soon as *C. eutraphera* can be collected from the field in the current season. In particular, Nipagin H3647 should be tried at lower quantities in diets 1, 5 and 6.

Table 1. Diet composition

Diet number 1a: Helicoverpa media (amended with mango pulp instead of pollen)		**Diet number 1b: Cucumber moth diet (amended with mango pulp instead of rock melon)**	
Cannelloni beans	75g	Distilled water	200 mL
Agar	15 g	Pentavite	6 drops
Yeast	25 g	Cannelloni beans	480 g
Ascorbic acid	5 g	Soluble starch	24 g
Nipagin M29908	2 g	Mango	200 mL
Acetic acid	1.25 mL	Cellulose powder	24 g
Bran	25 g	Sucrose	24 g
Wheat germ	22g	Brewers yeast	28 g
Mango blended	5 g	Ascorbic acid	2.8 g
Ascorbic acid	0.75 g	Ascorbic acid	0.66 g
Boiling water	750 mL	Nipagin, M29908	1.32 g
Formalin (40%)	0.5 mL	Agar	18 g
		Water	350 mL
All larvae tested on this diet died. It is thought that this was due to the preservative used.		All larvae tested on this diet died. It is thought that this was due to the preservative used.	
Diet number 2: Simpler diet without Nipagin		**Diet number 3: Diet enriched with vitamins without Nipagin**	
Mango (blended)	10 mL	Mango (blended)	10 mL
Agar	1 g	Agar	1 g
Distilled water	15 mL	Distilled water	20 mL
		Pentavite	1 drop
Citripestis larvae feeding on this diet developed slowly; so more vitamins were added in the form of pentavite for the next version. In the absence of Nipagin, mould developed on this diet.		It was observed that on this diet, growth of *Citripestis* larvae was still slow; so more nutrients were added in the form of beans and bran. In the absence of Nipagin, mould developed on this diet.	
Diet number 4a: Diet enriched with nutrients, version 1 without Nipagin		**Diet number 4b: Diet enriched with nutrients version 2 without bran and Nipagin**	
Mango (blended)	10 mL	Mango (blended)	10 mL
Agar	1 g	Agar	1 g
Distilled water	15 mL	Distilled water	15 mL
Pentavite	1 drops	Pentavite	1 drop
Beans	1 g	Beans	1 g
Bran	1 g		
These two diets were prepared to provide more nutrients to the *Citripestis* larvae. The second medium (b) did not contain bran so as to make it easier to see the larvae for biological studies. A problem encountered with all the media without Nipagin was the development of mould, which covered the entire media surface within a few days.			
Diet number 5: With mango and Nipagin H3647 preservative		**Diet number 6: Amended banana scab moth diet**	
Mango (blended)	100 mL	Wheat germ	30 g
Agar	10 g	Yeast	25 g
Distilled water	150 mL	Ascorbic acid	1.5 g
Pentavite	1 mL	Nipagin H3647	1.5 g
Beans	10 g	Ascorbic acid	0.5 g
Bran	10 g	Soy flour	42.5 g
Nipagin H3647	1 g	Water (for above)	230 mL
		Agar	15 g
		Water (for agar)	120 mL
Mould did not develop on this medium. However, while *Citripestis* larvae emerged while feeding solely on this diet, development was slow and staggered. Few moths emerged simultaneously and thus no eggs were obtained from these moths.		Mould did not develop on this medium. As with the other artificial media, the development of *Citripestis* larvae was slow. This diet was not tested fully as larvae were removed to diet 5 before they had completed their life cycle.	

Life history
The life history of *C. eutraphera* was recorded while they were maintained in an air-conditioned room at 28 °C.

Eggs
From 16/11/2009 to 24/12/2009, more than 200 eggs were observed. The eggs, which were laid on fruits or stalk, were about 1mm long, initially white in colour, turning to red the next day. Eggs hatched two to three days after being laid.

Larvae
The newly-hatched larvae were pale pink in colour and fed on the fruit surface or tunnelled into the fruit (most tunnelled in around the stalk). Some newly-hatched larvae were observed to feed on other unhatched eggs. After a few days, most of the larvae had bored into the fruit. The larvae fed in fruits for about 12 to 14 days and then left the fruit to pupate.

Pupae
From 05/10/2009 to 17/11/2009, 200 pupae were collected from 60 infested mango fruits. The emergence data is shown in Table 2.

Table 2. Pupae data

Time from pupating to emerging (days)	10	12	13	14	15	16	17	18	Total
No. of pupae emerged	2	2	6	64	52	14	8	2	150

Average pupal duration was 14 to 15 days and the percentage pupae emergence was 79%. The sex ratio (♀ :♂) of the adults was 81:77 (1: 0.95).

Adults
From 16/11/2009 to 24/12/2009, more than 80 male and female adults were released in cages and their longevity ranged between five days and 14 days. Female moths started to lay eggs two days after emergence, laying between 10 to 66 eggs.

Life cycle duration
From the above data the life cycle at 28 °C is shown in Figure 2.

Stage	Duration (days)	
Egg – initially white, then changes to red	2-3	
Larva	12-13	
Pupa	14-15	
Egg to adult	28-31	

Figure 2. The life cycle of *C. Eutraphera*

References:

Jacob, T. K., Veenakumari, K. and Bhumannavar, B. S. (2004). Insect pests of cashew in the Andaman Islands. Cashew 18 (4) Cochin: Directorate of Cashew Nut and Cocoa Development, 25-28.

Meyrick, E., (1933). *Exotic Microlepidoptera*. IV **(3):** 387-8.

Acknowledgement:

Thanks to Mark Hoult for assistance with records of the pest and supplies of mango fruit; Ken Raynor for supplying mango fruit; Dr Marianne Horak, ANIC, Canberra, for initial identification of the pest; the late Michael Shaffer, previously curator of Pyraloidea at the BMNH, London for confirmation of the identification; Stacey Anderson, Entomology Technician, NAQS – AQIS, Darwin for assistance with identification; Dr Andrew Mitchell, entomologist, NSW Department of Primary Industries for assistance with DNA studies.

PROJECT: *Citripestis eutraphera* - Seasonal Abundance and Distribution in the NT

Project Officers: B. Thistleton, M. Neal, L. Zhang and R. Meldrum

Location:	Darwin
Keyword(s):	mango, pests, *Citripestis eutraphera,* Lepidoptera, Pyralidae, light trapping, surveillance, seasonal abundance

Objective:

To record seasonal abundance and distribution of Citripestis eutraphera (Lepidoptera: Pyralidae) in the Northern Territory (NT).

Background:

The mango fruit borer is the caterpillar of a small moth (*C. eutraphera)*. This insect was recently detected in the Darwin area (see other reports in this publication). It was initially found on two widely separated properties. The mango fruit borer has been detected feeding on only mango in the NT. Overseas, it has also been detected on cashew (Jacob et al. 2004).

Method:

Seasonal abundance – light trapping
It was not possible to conduct surveillance for the pest by collecting mango fruit outside the mango season. So a light trap was set to see if adults could be attracted. The light trap was set up at Berrimah Farm (one of the original detection sites) on 28 April, 2009. Initially, the trap was set every night and later one night per week. Large numbers of a variety of moths were trapped and screened. Genitalia dissections were used to identify *C. eutraphera.*

Fruit samples
Samples of fruit were taken during the season to:

- Investigate if *C. eutraphera* was still present at the detection sites and determine the time of its first appearance.
- Obtain individuals for the establishment of a laboratory culture.
- Check the identity of larvae collected from other sites in the Top End.

Fruit was brought to the laboratory and larvae were reared to the adult stage for identification using external morphology and genitalia dissections.

Surveillance
The main aim of the surveillance was to determine the current distribution of the moth in the NT, based on presence/absence information. However, where possible, estimates were made from the numbers of trees examined and the number of fruit per tree, to assess the prevalence of the insect.

Results:

Seasonal abundance – light trapping

Light trap data is shown in Figure 1. It is evident that moth numbers became abundant only when mango fruit was present (August to November).

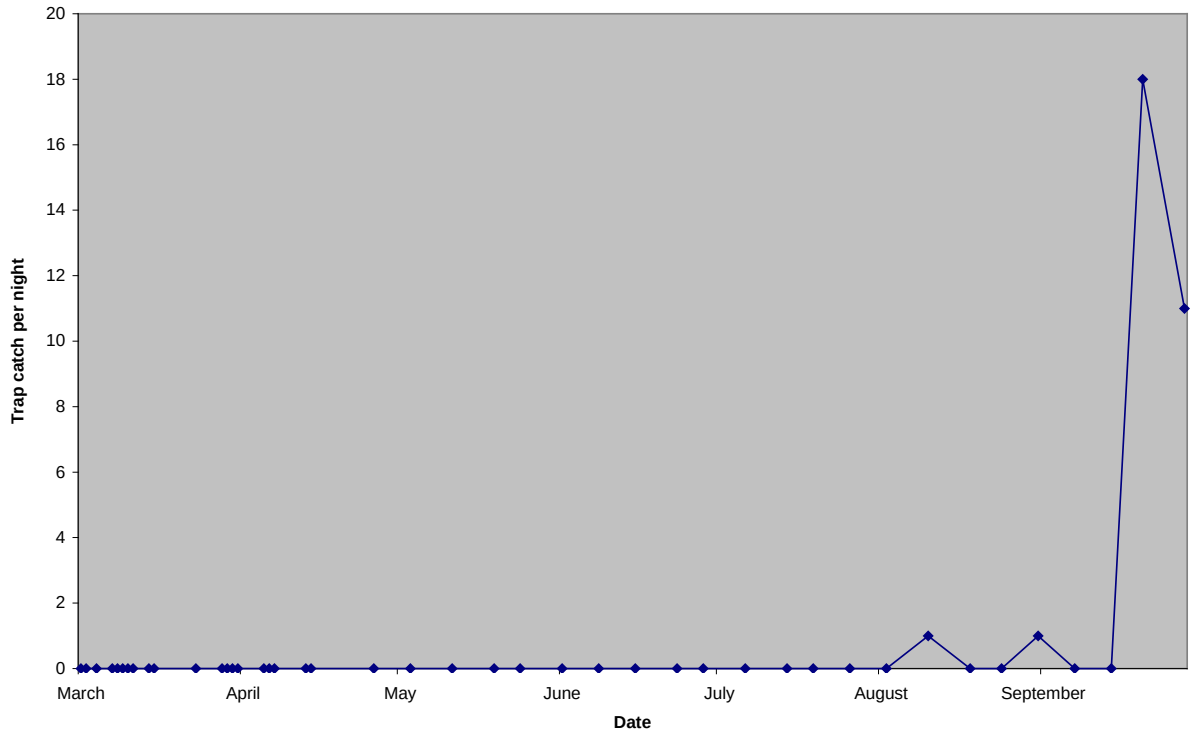

Figure 1. Moth numbers during the year

Fruit samples from Berrimah

Forty-two adults were bred from larvae collected from Berrimah Farm on 23 July. Of 1021 fruits examined between 25 June and 25 August, one from a tree had suspect *C. eutraphera* damage, while 540 fruits on the ground yielded 12 *C. eutraphera* adults.

In October, pest numbers increased (see moth trap results) and many fruits on trees displayed damage. Two of the more heavily-infested varieties (one tree of Akbar and one of Sensation) had a total of 282 fruits on them, of which 29 (10%) had suspect *C. eutraphera* on 23 October. There were no fruits on the ground for comparison as they had been eaten by magpie geese.

Fruit samples from Lambells Lagoon

Ten trees in each of four areas on a property at Lambells Lagoon (40 trees in total) were examined fortnightly from 19 August to 20 October. Out of 6277 fruits examined on trees and 777 fruits on the ground, four were found with larvae, all on the tree. One larva was identified as *C. eutraphera.* It had been collected on 19 August. It pupated, but failed to emerge. Another fruit from a tree yielded 13 pupae on 6 October, of which 10 emerged and were confirmed as *C. eutraphera.* Two fruits yielded one and 11 *C. eutraphera* pupae, respectively on 20 October.

So from a total of 7054 fruits examined, only four (0.06%) had larvae of *C. eutraphera*.

Other locations

Data from other locations is shown in Table 1, which indicates that the insect is widely distributed at low levels in the Darwin area. The insect has not been located yet in Adelaide River, Pine Creek or Katherine.

Table 1. Distribution of *C. eutraphera* in the Darwin area

Date	Location	No of trees examined	Range of fruit per tree	Estimated average fruit per tree	Total fruit examined[2]	No of fruit with *C. eutrphera*	Infested fruit (%)
25 June - 25 August	Berrimah	-	-	-	1,561	13	0.83
19 August - 20 October	Lambells	-	-	-	7,054	4	0.06
28 September	Casuarina	1	-	-	1	1	-
23 October	Berrimah[1]	2	-	-	282	29	10
23 October	Acacia Hills	200	30-300	100	20 000	11	0.06
28 October	Howard Springs	150	20-60	40	6000	17	0.28
28 October	Humpty Doo	9	50-300	200	1,800	2	0.11
29 October	Berry Springs	40	50-300	200	8,000	7	0.09
29 October	Casuarina	1	100	100	100	1	1.00
29 October	Nightcliff	10	10-100	40	400	1	0.25
12 November	Adelaide River	15	30-200	100	1500	0	0
12 November	Pine Creek	17	50 300	150	2550	0	0
13 November	Katherine	280	50-150	100	28 000	0	0

1. Samples at Berrimah were taken to collect larvae for the culture, rather than to provide prevalence data and are therefore focussed on heavily infested trees. In particular, the sample on 23 October was taken from two trees (Akbar and Sensation) which had high populations.
2. Actual numbers for Berrimah and Lambells Lagoon; others were estimated numbers from tree number and average fruit per tree.

Figure 2. Known distribution of *Citripestis eutraphera* in the NT (November 2009)

Reference:

Jacob, T. K., Veenakumari, K. and Bhumannavar, B. S. (2004). Insect pests of cashew in the Andaman Islands. Cashew 18 (4) Cochin: Directorate of Cashew nut and Cocoa Development, 25-28.

Acknowledgement:

Thanks to Mark Hoult for assisting with records of pests and for supplying mangoes, Ken Raynor for supplying mangoes and to various growers for allowing access to their properties for the surveillance work.

PROJECT: *Citripestis eutraphera*: Market Access Issues

Project Officers: B. Thistleton, L. Zhang, M. Neal and R. Meldrum

Location: Darwin

Keyword(s): mango, pests, *Citripestis eutraphera,* Lepidoptera, Pyralidae, market access, fenthion, dimethoate

Objective:

To test mortality rates in Citripestis eutraphera eggs and small larvae due to fenthion and dimethoate applied at currently approved rates for post-harvest fruit fly control for interstate market access.

Background:

The mango fruit borer is the caterpillar of a small moth (*C. eutraphera)* (Lepidoptera: Pyralidae), which bores into the mango fruit. This insect was recently detected in the Darwin area (see other reports in this publication).

Up to now, the pest has been found only in Darwin and its rural areas. Surveys have indicated that the prevalence of the pest in these areas is very low. The symptoms caused by large larvae on the fruit are very distinct, including the presence of holes with extruded frass, or splitting. Damaged fruit are either unlikely to be harvested, or would be graded out in the packing shed.

The eggs are very small, but can be seen with a hand lens. They are white or red. Small larvae cause feeding damage on the surface of the fruit. There is a risk that eggs and small larvae could be overlooked in the packing shed and could enter the supply chain, spreading to other mango-growing regions in Australia.

At present, no additional requirements have been imposed for the interstate movement of mangoes from the Northern Territory (NT) over what is currently required for treatment against fruit flies. However, it has been requested that further work be conducted on the efficacy of current fruit fly treatments against the mango fruit borer for consideration by the Domestic Quarantine and Market Access Working Group.

Fruit fly treatments are covered by Interstate Certification Assurance (ICA) 01, 02 and 03 (NT Government 2005 a, b, c) covering dipping and flood spraying with dimethoate, fenthion and low volume non-recirculated spraying with fenthion, respectively. In all cases, ICAs require wetting of fruit with an insecticide mixture containing 412.5 mg/L of fenthion or 400 mg/L dimethoate for a period of not less than one minute.

Method:

Only two preliminary tests have been carried out up to now. Since there were no mangoes available at the time of testing, it was decided to use *C. eutraphera* eggs laid on a non-host material. Many eggs were being laid on the fine mesh fabric on the honey/water feeders in the egg laying cages and it was possible to cut this mesh into small pieces containing batches of eggs.

An initial test was carried out with fenthion. The eggs used were laid overnight on Wednesday 28 April, 2010. The eggs were counted and dipped on Thursday 29 April, 2010 for 1 minute in a mixture containing 412.5 mg/L of fenthion. Once dry, the samples were stored in individual containers for assessment. The samples were observed in the days following the dipping date. Final assessment occurred on Tuesday, 4 May, 2010.

Results:

Some of the eggs fell off the mesh in the dipping tank. The assessment (Figure 1) was based on the remaining eggs which hatched or did not hatch.

These results show that only two of the 139 eggs dipped in fenthion hatched. However, one of these larvae died not long after hatching.

Table 1. The effect of fenthion on *C. eutraphera* eggs

ID number	Treatment	Red eggs	White eggs	Total eggs	Hatched eggs	Unhatched eggs	Damaged	Notes
1	Water control	10	0	10	10			
2	Water control	5	7	12	7	3		
3	Water control	1	6	7	5	1		
4	Water control	2	15	17	8	5		
5	Fenthion treatment	4	17	21	1	17		
6	Fenthion treatment	16	29	45		37		
7	Fenthion treatment	12	25	37		29		
8	Fenthion treatment	7	21	28	1	27		Hatched larvae were dead
9	Fenthion treatment	6	11	17		16		1 larva formed inside
10	Fenthion treatment	9	4	13		11		
11	No dipping	20	13	33	20	5	5	
12	No dipping	23	13	36	14	5	1	Includes dead larvae
13	No dipping	16	11	27	23	1	1	
14	No dipping	13	15	28	24	2		

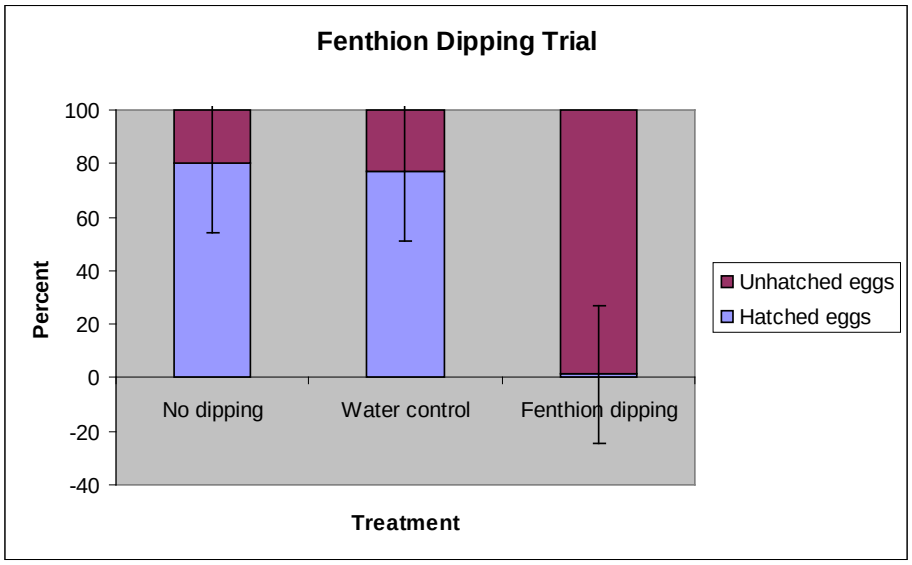

Figure 1. The effect of fenthion on *C. eutraphera* eggs

This is only the first test in a series which will look at mortality of eggs and young larvae when fruit is treated with either fenthion or dimethoate.

Fenthion and dimethoate do not kill fruit fly eggs directly but death occurs when the newly-hatched larvae feed on the treated fruit (P. Leach, pers. comm.). The fact that less than 2% of *C. eutraphera* eggs hatched in this test is very encouraging.

References:

NT Government, 2005a. Interstate Certification Assurance ICA-01: Dipping with dimethoate or fenthion. 31 pp.

NT Government, 2005b. Interstate Certification Assurance ICA-02: Flood spraying with dimethoate or fenthion. 33 pp.

NT Government, 2005c. Interstate Certification Assurance ICA-03: Low volume non-recirculated spraying with fenthion. 32 pp.

PROJECT: Managing Mango Leafhoppers and Other Sucking Insects in Mango Trees through Systemic Insecticides

Project Officers: S. Qureshi, B. Thistleton, M. Hearnden and M. Neal

Location: Darwin

Key words: mango, thiamethoxam, Imidacloprid, *Idioscopus nitidulus*, *I. clypealis*, drench, nymphs, sap-sucking

Objective:

To conduct a series of trials in the Northern Territory (NT) and Indonesia to manage leafhoppers and other sap-sucking insect pests and to assess their impact on mango yield and quality.

Background:

Mango, *Mangifera indica* L., a member of family Anacardiaceae, is an important commercial fruit of tropical and subtropical regions of the world (Barman et al. 2007). It is the most important commercial fruit crop in the NT (NT Government 2004).

Several arthropod pests could seriously affect mango production and marketing in Australia. They include two species of mango leafhoppers (MLHs) *Idioscopus nitidulus* (Walker) and *I. clypealis* (Lethierry) (Chin et al. 2002). MLHs are considered the most destructive pests of all mango varieties overseas and can potentially cause severe damage in Australia (Karim 1989; Anufrieu 1970; Corey et al. 1986). *I. nitidulus* is present in the NT.

The nymphs and adults of MLHs are sap-sucking insects, which generally feed on new shoots, leaves, inflorescences and, in some cases, fruit (Das et al. 1969). In addition, they excrete the exudate “honey dew”, which encourages the growth of sooty mould on the dorsal surfaces of leaves, branches, and fruit. This coating interferes with photosynthesis in the plant, resulting in the non-setting of flowers and dropping of immature fruit (Kudagamage et al. 2001).

MLHs have no natural predators within their range in Australia, leaving the judicious use of insecticides as the only management option. However, the indiscriminate use of insecticides may lead to the development of insecticide resistance in insects, disruption of agro-ecosystems, environmental pollution and create a serious threat to human health (Luckman and Metcalf 1982).

The aim of this study was to assess the efficacy of two systemic neonicotinoid insecticides: thiamethoxam (Actara®) and imidacloprid (Confidor®) through soil-drench and trunk-injection applications to control MLHs and other sap-sucking insect pests at Kensington Pride (KP) mango orchards in Darwin, and Arumanis and Gedong Gincu varieties in Lombok, Indonesia.

Method:

Trial 1

Design and treatments

Four rows of 10 mango trees each were selected on the eastern side of a Darwin mango orchard adjacent to the Stuart Highway (12° 25' 15.05"S; 130° 56' 18.00"E) in 2009. The wind direction during the dry season in the Darwin region is from the east, minimising spray and chemical drift from other trees in the orchard. There were five treatments in the study: two different doses each of thiamethoxam and imidacloprid, plus a control.

The treatments were arranged in a randomised complete block design. Each row was treated as a block and each tree as a replicate. There were five replicates in each row with a single tree buffer between each. There was a 10-m space between rows and between the 12-year-old KP trees.

A single dose was applied to all the treatment groups when all the trees had approximately 20% flowering, on 12 May, 2009 at the following rates:

1. Thiamethoxam, 6 g/tree of Actara® 25WG (equivalent to 1.5 g ai/tree) by soil drench, in 1 L of water.
2. Thiamethoxam, 12 g/tree of Actara® 25WG (equivalent to 3 g ai/tree) by soil drench, in 1 L of water - the recommended dose for red scales (*Aonidiella aurantii)* on citrus.
3. Imidacloprid, 7.5 mL/tree of Confidor® 200SC (equivalent to 1.5 g ai/tree) by soil drench, in 1 L of water.
4. Imidacloprid, 15 mL/tree of Confidor® 200SC (equivalent to 3 g ai/tree) by soil drench, in 1 L of water.
5. Control, 1 L of water by soil drench.

Figure 1. Applying Actara (systemic insecticide) into a mango tree via a soil drench

Soil drench application

The base of each treated tree was cleared of all dead and dry leaf matter and irrigated every day for one week before drench application to ensure the feeder roots were active. A 5 to 10 cm deep trench was made around the base of each tree, about 30 cm away from the trunk. Each treatment was prepared in 1 L of water and was applied to the trench of each tree with a watering can. Irrigation was subsequently provided for one week to all treated trees to absorb the insecticides.

Insect sampling

The number of MLHs (adults and nymphs) and associated species of interest (scale insects, plant hoppers, spiders, green ants and lacewing eggs) was recorded from the terminal 30 cm of one branch on each side of each tree: north, south, east and west (a total of four branches/tree). Records were taken one day before drenching (DBD) and 14, 28, 42, 56, 70, 84, 98, 112, 126, 140 and 154 days after drenching (DAD). Sampling was carried out during the early hours of the morning when the insects were least active.

Fruit sampling

Fruit (tennis ball size and larger) from each treated tree was counted on 19 October, 2009 by two observers, and the mean of the two observations was recorded. Where the difference between the two observations was greater than 15, the fruit was recounted. Fifteen fruits from all over each tree were harvested and assessed under laboratory conditions for visual quality, presence of insects and insect damage.

Trial 2

A similar trial set-up was used in trial 2 at a Berrimah mango farm, using 10-year-old KP trees. The systemic insecticides were applied by drench as well as by trunk injections at five injections/tree. The five treatments were applied at the following rates:

1. Thiamethoxam at 6 g/tree by soil drench, in 1 L of water.
2. Thiamethoxam at 6 g/tree by trunk injections (five injections/tree), in 20 mL of water/injection (i.e. 100 mL of water/tree).
3. Imidacloprid at 7.5 mL/tree (equivalent to 1.5 g ai/tree) by soil drench, in 1 L of water.
4. Imidacloprid at 7.5 mL/tree by trunk injections (five injections/tree), in 20 mL of water/injection (i.e. 100 mL of water/tree).
5. Control (1 L of water only by soil drench).

A similar method to that described in trial one was used for the soil drench application, and for insect and fruit sampling.

Figure 2. Applying Actara (systemic insecticide) by injection into a mango tree trunk

Data analysis

The counted fruits were analysed using a main effects analysis of variance to test for differences between treatments. *Post hoc* analysis of treatment effects with controls were compared using Dunnett's test.

The total number of observed adult and nymph MLHs, scale insects and spiders were analysed using a repeated measures analysis of variance to determine differences in mean numbers of each treatment level over time - DAD. Analysis of the residual values of the data indicated that no data transformations were required. *Post hoc*

comparison of treatments over time was calculated with contrast vectors comparing the control to each of the chemical treatments. *Post hoc* pair-wise comparisons between means were facilitated by calculating Fisher's least significant difference corrected using the Dunn-Sidak adjustment to protect the experiment-wise error rate at 5%. In the absence of an interaction over time, *post hoc* analysis of treatment main effects with controls were compared using Dunnett's test.

The equality of the proportion of fruit damaged by different groups of insect pests (pink wax scale, mealy bugs, caterpillars, ants, thrips and blemishes by other insects) and damage caused by formic acid, sap-burn and sooty mould within all treatments were carried out using binomial proportion tests.

Results:

Fruit counts

There was a significant difference in the mean number of fruit counted in each of the treatments (ANOVA, $F_{4,12}$, F=3.62, *P*=0.0370). Mean fruit number in the thiamethoxam 12 g treatment was significantly higher than that of the control. All other treatments were similar to the control (Table 1).

Table 1. Treatment means (+/- SE) for fruit counts with Dunnett's test *p* values for similarity to the control mean (means with an asterisk are significantly higher than the control).

Treatment	Mean count (+/-SE)	Comparison with control (*P*)
Control	72.5 (34.3)	
Thiamethoxam 6 g	76.3 (4.99)	0.7572
Imidacloprid 6 g	112.5 (8.3)	0.2594
Thiamethoxam 12 g	172.5 (19.55)	0.0097 *
Imidacloprid 12 g	116.8 (27.12)	0.2149

Mango leafhopper counts

Adults

The trend over time was a similar initial reduction in all treatments in the number of MLH adults after initial drenching, with numbers increasing from July to August and diminishing to mid October (Figure 3). Counts over time in the control treatment were significantly higher than in all other treatments (ANOVA, $F_{44,165}$=2.16, *P*=0.0003) while the chemical treatments indicate that higher doses seemed to reduce insect numbers more when relative abundance was higher (June to September).

Nymphs

Counts of MLH nymphs began increasing in most treatments by July corresponding with the increased numbers of adults (Figure 4). Mean numbers in all chemical treatments over time were clearly lower than those observed in the controls (ANOVA, $F_{28, 105}$=2.87, *P*<0.0001) and as a result of the adult response, the stronger chemical treatments were associated with a greater effect on mean numbers, particularly during the period of peak abundance from late July to September (see lsd. in Figure 4).

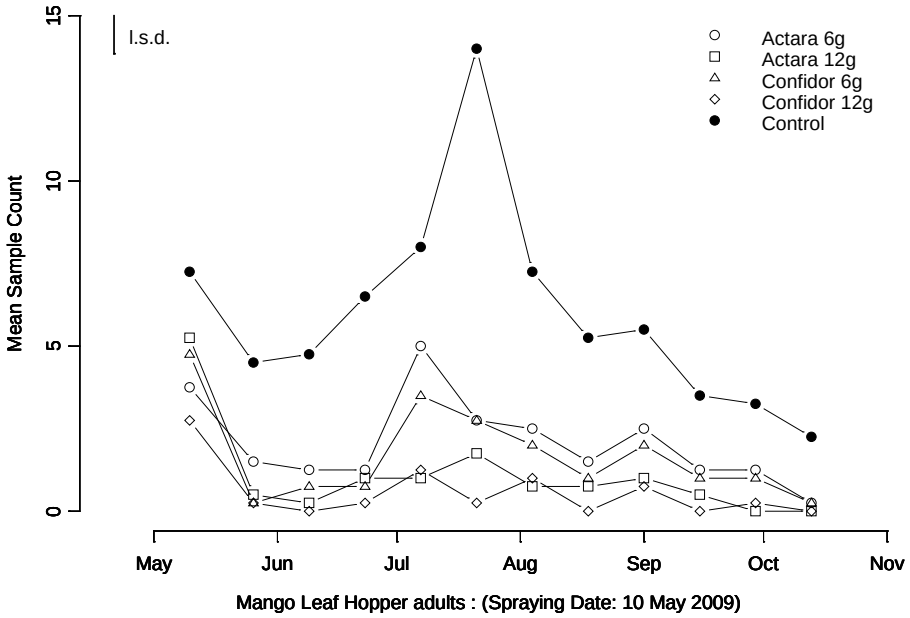

Figure 3. The mean number of adult mango leafhoppers observed during the sampling period in five treatments (thiamethoxam 6 g, thiamethoxam 12 g, imidacloprid 6 g, imidacloprid 12 g and control) from May 2009 to November 2009

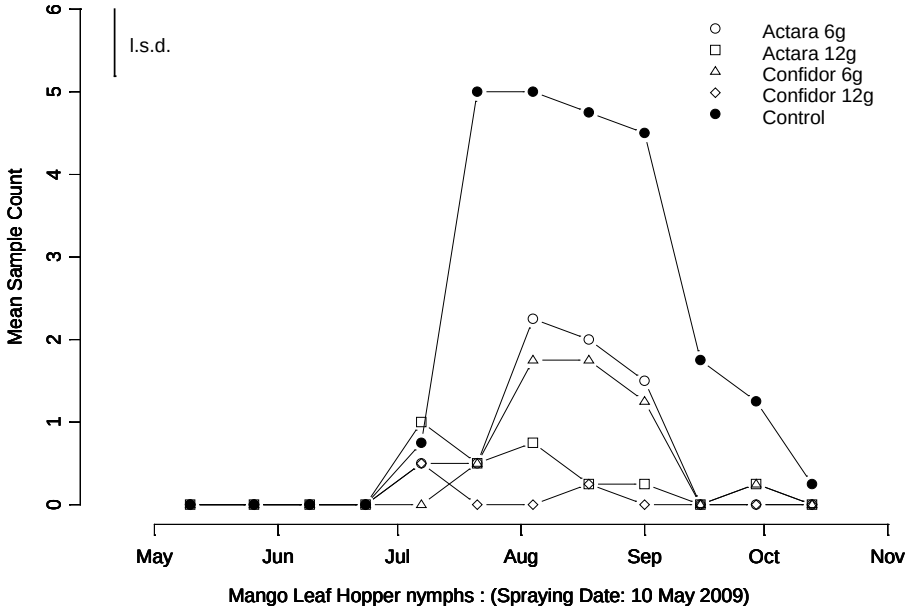

Figure 4. The mean number of nymph mango leafhoppers observed during the sampling period in five treatments (thiamethoxam 6 g, thiamethoxam 12 g, imidacloprid 6 g, imidacloprid 12 g and control) from May 2009 to November 2009

Scale insect counts

There was a significant difference in the mean number of scale insects during the trial (ANOVA, $F_{4, 15}$=17.58, *P*<0.0001); however, numbers were low and did not vary differentially between treatments over time (ANOVA, $F_{44, 165}$=0.70, *P*=0.9210). The mean numbers for all chemical treatments were significantly lower than in the controls (Table 2.)

Table 2. Treatment means for scale insect counts with Dunnett's test *P* values for similarity to the control mean (means with an asterisk are significantly lower than the control)

Treatment	Mean count (SEM=0.079)	Comparison with control (*P*)
Control	0.875	-
Thiamethoxam 6 g	0.333	0.0008 *
Thiamethoxam 12 g	0.020	<0.0001 *
Imidacloprid 6 g	0.333	0.0008 *
Imidacloprid 12 g	0.104	<0.0001 *

Spider counts

There was a significant difference in the mean number of spiders during the trial (ANOVA, $F_{4, 15}$=14.68, *P*<0.0001); however, numbers were low and did not vary between the treatments over time (ANOVA, $F_{44, 165}$=0.86, *P*=0.7080). Mean numbers of spiders in all chemical treatments were significantly lower than in the controls (Table 3).

Table 3. Treatment means for spider counts with Dunnett's test *P* values for similarity to the control mean (means with an asterisk are significantly lower than the control)

Treatment	Mean count (SEM=0.079)	Comparison with control (*P*)
Control	0.895	-
Thiamethoxam 6 g	0.417	0.0047 *
Thiamethoxam 12 g	0.063	<0.0001 *
Imidacloprid 6 g	0.250	0.0003 *
Imidacloprid 12 g	0.146	<0.0001 *

Fruit damage and defects

Significantly lower proportions of damage to fruit in chemical treatments (relative to the control) were associated with caterpillars (Chi-square = 25.4, df = 4, *P*< 0.0001), green tree ants (*Oecophylla smaragdina*) (*P*<0.0001), and other insects (*P*<0.0001). Sooty mould (Ascomycete fungi) was only observed in the control treatment. The proportion of damage associated with thrips (Russett) and pink wax scale were similar in all treatment groups. The proportion of damage associated with mealy bugs (*P*<0.0001) and sap-burn (*P*<0.0001) were significantly different between treatments but not necessarily associated with any increase in control trees. Similar results were found in the second trial in Darwin. Two other similar trials (Trial 3 and Trial 4) conducted in Lombok, Indonesia did not show any significant difference in the number of MLHs and other sap-sucking insects, and yield and quality of mango. This was possibly due to no irrigation applied or due to a different soil type (sandy soil), possibly leading to the chemical being leached away from tree roots.

Thiamethoxam and imidacloprid provided effective control against a number of insects, both in this trial and overseas (Patel et al. 2003; Godase et al. 2008; Bhaskar 2007; Palis et al. 1994; Verghese 2000; Kudagamage et al. 2001; Kumar and Giraddi 2001; Bhaskar et al. 2007). Both thiamethoxam and imidacloprid were found to be effective against MLH nymphs and adults. Work overseas has demonstrated that thiamethoxam has the further advantage of being effective against other sucking pests of mangoes, including mealybugs (in India and South

Africa: Murugan and Ramachandran 2001; Le Lagadec et al. 2009) and mango seed weevils (in South Africa: Louw et al. 2009).

Similarly, both thiamethoxam and imidacloprid proved effective against scale insects compared with the control in this trial. In contrast, only thiamethoxam was found to be very effective in a series of trials conducted in South Africa against the mango scale insect (*Aulacaspis tubercularis*) and mealybugs compared with imidacloprid and acetamiprid. Hence thiamethoxam is registered for use on mangoes for the control of scale insects and mealybugs in South Africa (Le Lagadec et al. 2009).

Both thiamethoxam and imidacloprid significantly reduced the population of spiders. Previously, the role of spiders in population regulation of pests was not fully known (Wheeler 1973) but they are believed to play a significant role in limiting some herbivore populations (Yeargan 1975).More recently, spiders are considered as beneficial arthropods due to their aggressive predatory nature against other insects and have the ability to move greater distances at a faster pace than other generalist predators (Bishop and Riechert 1990). Further research is essential to determine whether this effect is a consequence of reduced numbers of prey, or a direct effect of the chemicals. Further research could also quantify the beneficial status of spiders in a mango crop.

The number of fruits per tree at harvest was not significantly reduced in the treatments when compared with the control. On the contrary, trees treated with the higher rate of thiamethoxam returned a higher mean level of production. This resulted in a significant increase in fruit numbers. The quality of fruit at harvest was significantly better in the treated samples compared with the control. There was a significantly higher incidence of formic acid marks, caused by green ants on the control trees. Similarly, there were significantly (P<0.05) more sooty mould marks, caused by MLHs, in the control group compared with the thiamethoxam and imidacloprid groups.

Thiamethoxam was found to be effective in controlling sucking insects in mangoes, especially for MLH populations. Whilst it has the potential to enhance mango yield, its effect on populations of natural enemies (predators and parasitoids) requires further research. Thiamethoxam also has the potential to improve fruit quality with the reduced incidence of both sooty mould and formic acid marks with its use.

However, pest pressure was low when this trial was conducted. Further research is needed to confirm the effects of thiamethoxam on sucking insects of mangoes in different sizes of canopy and different tree ages, especially during times of peak pest pressure.

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PROJECT: The Efficacy of Fungicides in controlling Post-harvest Diseases in Mango Orchards

Project Officers: A. Daly and J. Liberato

Location: Darwin

Keyword(s): mango, post-harvest, fungicides, anthracnose, Mangifera, Colletotrichum

Objective:

To determine the efficacy of fungicides in controlling post-harvest diseases in mango orchards.

Background:

Anthracnose of mangoes (*Mangifera indica*) is widespread and is one of the most important diseases of this crop (Arauz 2000; Akem 2006). In Australia, anthracnose is predominantly caused by the fungus *Glomerella cingulata* (anamorph: *Colletotrichum gloeosporioides*). The pathogen infects leaves, flowers and developing fruit. The infection generally remains quiescent on fruit until harvest when ripening begins (Prusky 1996; Prusky et al. 2009). At that time black, sunken rotted areas develop on the fruit. If fruit ripening occurs during wet conditions or high humidity, severe infection is likely to occur (Arauz 2000). This situation is often a concern in the Top End of the Northern Territory, as it significantly reduces the saleable life index of the fruit and reduces market options for the producer.

Stem end rot of mangoes, another important post harvest disease, is caused by a number of (mostly) related fungi worldwide (Johnson 1993). In Darwin, the causal agent is *Lasiodiplodia theobromae*. It exists as an 'endophytic' colonist of mango trees, occurring within the stem and inflorescence tissues, including the fruit stalk and only invading the fruit following harvest (Johnson 1993; Johnson et. al. 1992).

Currently, fungicides are essential to control the disease in the field and post-harvest. Copper-based fungicides and mancozeb are commonly used as protective fungicides in the field. However, their effectiveness in controlling anthracnose appears limited during periods when the weather is conducive to disease. They do not control stem end rot. Azoxystrobin, a systemic fungicide, appears to be effective for controlling anthracnose (Sales et al. 2004; Sundravadana et al. 2006; Coates et al. 2009).

Calendar-based applications of copper-based fungicides and mancozeb are commonly utilised by growers during flowering and fruiting. In addition, one to three sprays of azoxystrobin have been recommended by its manufacturer. However, there is little information about the effects of the number and timing of sprays of

azoxystrobin on the control of disease post-harvest. The aim of this study was to evaluate the efficiency of azoxystrobin in controlling mango anthracnose and stem end rot under various fungicide application schedules.

Method:

Field treatments

The trial was conducted at an orchard with 11-year-old Kensington Pride mango trees at Lambells Lagoon (45 km south-east of Darwin) during the 2009 mango season. Three fungicides and 11 application schedules (treatments) were evaluated (Tables 1 and 2).

Trees with relatively even flowering were chosen and treatments were assigned employing a randomised complete block design. Each block comprised one row of mango trees and each plot one tree. Two rows each side of the experimental site as well as two mango trees at the end of each block comprised the external buffer. Internally, a tree row acted as a buffer between blocks, except between blocks 2 and 3 where the buffer consisted of two rows. Within blocks, there was a buffer tree between plots. The buffer trees did not receive any fungicide spray.

Treatments started when most inflorescences were in the stage between "full bloom" (flowers open on all panicle branches except for the tip) and fruit-set (swelling of the light green ovary in the centre of each flower). Suspensions of the fungicides were applied up to the point of run-off with a portable 2.3 mm nozzle Evolution M21 turbine gun connected to a 4.8 HP GX 160 Honda powered AR30 diaphragm pump operating at 40 bars.

At harvest, on 2 November, 2009, fruits on the trees were counted. From each replication, samples of 20 fruits of similar maturity (based on visual assessment of the cleavage, beak, shoulders and colour) were randomly harvested. For treatments 1, 2, 4, 5, 10 and 11, a second 20-fruit sample was harvested. The fruits were de-stemmed and de-sapped in the orchard.

Post-harvest

After harvest, the fruits were taken to the post-harvest laboratory at Berrimah Farm. For treatments 1, 2, 4, 5, 10 and 11, the second 20-fruit sample was treated with a non-recirculating spray of prochloraz at 247.5 mg/L[1] active ingredient for 30 seconds. All fruits were then kept at 20-22^{o}C until assessment, 14 days after harvest.

Fruits were assessed for incidence and severity of anthracnose using a modified 0-5 index scale based on Brodrick's (1978) and Cordiki et al. (2006): 0 (no disease), 1 (>0-1%,), 2 (>1-5%), 3 (>5-10%), 4 (>10-50%) and 5 (>50% of the fruit surface covered by lesions) and incidence of stem-end rot.

Statistical analyses were performed using the Statistica data analysis software system, version 8.0 (www.statsoft.com).

Table 1. Detailed information of the fungicides used in the 2009 season field trial

Common name	Trade name	Formulation 2	Active ingredient (%)	Manufacturer	Rate applied (dose/100 L water)
Azoxystrobin	Amistar®	250 SC	25	Syngenta	80 mL
Mancozeb	Manzate®	DF	75	DuPont	200 g
Cupric hydroxide	Champ Dry Prill®	WG	37.5	Nufarm	210 g
Prochloraz 1	Sportak®	EW	45	Bayer	55 mL

[1] Applied post-harvest; [2] SC = soluble concentrate; DF = dry flowable; WG = water dispersible granule; EW = emulsion, oil in water.

Table 2. Fungicide application schedules on mango trees in the 2009 season field trial[1]

Treatment	Azoxystrobin[2] sprayed in the field					Protective fungicides sprayed fortnightly	
	Full bloom/ fruit-set	Early season	Mid season	21 days pre-harvest	Seven days pre-harvest		
1 (Control)	-	-	-	-	-	-	-
2	-	-	-	-	-	mancozeb	5-Aug; 19-Aug; 2-Sep; 16-Sep: 30-Sep; 14-Oct; 21-Oct;
3	-	-	-	-	-	cupric hydroxide	5-Aug; 19-Aug; 2-Sep; 16-Sep; 30-Sep; 14-Oct; 21-Oct;
4	-	19-Aug	-	-	26-Oct	mancozeb	5-Aug; 19-Aug; 2-Sep; 16-Sep; 30-Sep; 14-Oct; 21-Oct;
5	5-Aug	19-Aug	-	-	26-Oct	mancozeb	5-Aug; 19-Aug; 2-Sep; 16-Sep; 30-Sep; 14-Oct; 21-Oct;
6	-	19-Aug	11-Sep	-	26-Oct	mancozeb	5-Aug; 19-Aug; 2-Sep; 16-Sep; 30-Sep; 14-Oct; 21-Oct;
7	-	19-Aug	-	12-Oct	26-Oct	mancozeb	5-Aug; 19-Aug; 2-Sep; 16-Sep; 30-Sep; 14-Oct; 21-Oct;
8	-	19-Aug	-	-	26-Oct	-	-
9	5-Aug	19-Aug	-	-	26-Oct	-	-
10	-	19-Aug	11-Sep	-	26-Oct	-	-
11	-	19-Aug	-	12-Oct	26-Oct	-	-

[1] Fruits were harvested on 2 November, 2009

[2] Azoxystrobin was always mixed in the tank with mancozeb

Results:

There was no significant difference among any of the treatments in fruit yield (number of fruits per tree) (data not shown). The incidence of stem-end rot ranged from 1.3 to 5.1% among the 11 treatments and there were no significant differences among them (data not shown).

Treatments 2, 3, 8 and 9 did not differ from the control in relation to incidence and severity of anthracnose. The field treatments combined with post-harvest application of prochloraz (treatments 2, 4, 5, 10 and 11) significantly reduced anthracnose compared with the application of prochloraz alone (Table 3).

Compared with the control, the reduction of anthracnose by fortnightly sprays of mancozeb or copper was not statistically significant, although mancozeb alone reduced the incidence of anthracnose by more than half. When fortnightly field applications of mancozeb alone (treatment 2) were combined with post-harvest application of prochloraz, the incidence of anthracnose declined significantly compared with the use of prochloraz alone, from 36% to 8% (see Table 3). Further evidence of the effectiveness of mancozeb was found using analysis of variance of linear contrasts, where the addition of fortnightly sprays of mancozeb to azoxystrobin plus mancozeb mixture sprays were more effective than two or three azoxystrobin plus mancozeb mixture sprays alone.

Two sprays of azoxystrobin plus mancozeb were not effective (treatment 8). However, when fortnightly sprays of mancozeb were combined with two sprays of azoxystrobin plus mancozeb (treatment 4), anthracnose incidence was reduced from 55% (in the control) to 14%. When three sprays of azoxystrobin plus mancozeb were used (treatment 9 and 10) the timing of application affected the results. Treatments 4, 5, 6, 7, 10 and 11 significantly reduced anthracnose. Treatments 4, 5, 10 and 11 were also tested in combination with post-harvest application of prochloraz, further reducing disease incidence to 0–3% (Table 3).

Table 3. The effect of fungicide applications on the incidence of anthracnose on Kensington Pride mangoes 14 days after harvest in the 2009 season

Treatments[1] in the field	Incidence (%)		Severity	
	Post-harvest treatment with prochloraz			
	Without	With	Without	With
7	3.75 c		0.05 c	
6	7.63 c		0.08 c	
5	8.33 c	1.25 b	0.10 c	0.01 b
10	13.75 cb	0.00 b	0.18 cb	0.00 b
4	14.14 cb	2.57 b	0.21 cb	0.04 b
11	18.42 cb	3.33 b	0.22 cb	0.03 b
2	25.00 cba	8.42 b	0.28 cba	0.10 b
9	26.67 cba		0.43 cba	
8	30.00 cba		0.36 cba	
3	43.95 ba		0.70 ba	
1 (control)	55.16 a	36.67 a	0.89 a	0.42 a
Variability coefficient	58.75%	75.31% [3]	72.02%	8.38% [4]

[1] For details about treatments, see Table 2.

[2] Means followed by the same letter in the same column are not significantly different (Tukey's test, $P<0.05$).

[3, 4] Previously the statistical analysis data for these variables was recoded as arc sin ($\sqrt{X/100}$) and $\sqrt{X+0.5}$, respectively.

Discussion:

Rainfall during fruit development is conducive to the occurrence of anthracnose. There was no rain at the trial site during the experiment; yet post-harvest anthracnose in control fruits was 55%. It is suggested that dew resulting from high humidity during most nights favoured infection of fruits. Previous studies have shown that *Colletotrichum gloeosporioides* var. *minor* requires the presence of free water on the mango leaf surface for sporulation of lesions, although a small amount of conidia (less than 10%) was produced when the relative humidity (RH) was between 95 and 97% (Fitzell and Peak 1984). Dodd et al. (1991) and Estrada et al. (2000) showed that conidia of *C. gloeosporioides* can also germinate and form dark appressoria at RH of 95% to 100% and at temperatures between 20 and 30 °C. During the 2009 fruit development period, except for ten days, when the maximum RH ranged from 96.2% to 99.4%, the RH in the orchard reached 100% at night, even though it was as low as 51.3% at 9.00 a.m. next day. This is evidence that the free water provided by the dew alone is enough to enable the dispersion of the pathogen and infection. This contradicts Fitzell and Peak's (1984) conclusion that rain was necessary to disperse conidia and to cause disease.

Although the incidence of anthracnose was relatively high, the severity was very low. Therefore, the treatments were not tested under high disease pressure. Even in that situation, the use of prochloraz alone in post-harvest treatment did not provide good control. It only reduced the incidence from 55% to 37%, which was still unacceptable by industry expectations.

Results from this study do not support fortnightly sprays of cupric hydroxide in the field for controlling post-harvest anthracnose. Although fortnightly sprays of mancozeb (treatment 2) in the field reduced anthracnose, its use alone did not provide an acceptable level of control and it may be even less efficient under higher disease pressure. Two sprays of azoxystrobin plus mancozeb were not sufficient and the effectiveness of three sprays of azoxystrobin plus mancozeb was time-dependent. The best results were obtained with two or three sprays of azoxystrobin plus mancozeb combined with fortnightly sprays of mancozeb. The additional post-harvest application of prochloraz markedly increased the control of anthracnose. When combined with fortnightly sprays of mancozeb, the timing of the azoxystrobin plus mancozeb mixture sprays appeared not to be critical under these experimental conditions.

According to Coates et al. (2009), it appears that the efficiency of sprays of azoxystrobin plus monthly sprays of mancozeb depended on disease pressure.

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PROJECT: Mango Defect Analysis 2009

Project Officers: C. Moore, S. Qureshi, I. Baker and M. Hearnden

Location: Darwin

Keyword(s): mango, post-harvest

Objective:

To conduct a defect analysis on mangoes from the Darwin and Katherine regions during the 2009 season.

Background:

An in depth fruit defect analysis was considered useful to meet research and extension needs in the local mango industry. The project aims to clearly identify the top post-harvest issues affecting fruit quality, irrespective of whether they are related to insects or diseases. Current research and extension in post-harvest quality is based on informed guesses, but is not necessarily representative of the whole industry. Furthermore, it was believed by industry representatives that current knowledge could not identify all post-harvest defects seen on fruit in packing sheds. Therefore, the project also aimed to investigate the extent and diversity of unidentified/un-named defects.

Method:

Seventeen mango samples were taken from Darwin rural and Katherine farms during the 2009 mango season. The Darwin region was sampled twice, once in September (Darwin 1) and again in November (Darwin 2). Of the mangoes sampled, all but two were Kensington Pride (KP); the others were Calypso.

Approximately 160 mangoes were collected for each sample. Ten fruits were taken randomly every 5 minutes from the packing line after treatment, but before grading.

The fruit was held at room temperature and was assessed individually. An initial grade was given to the fruit from 1 to 4 as follows:

Table1. Criteria for grading

Grade	Criteria
1	Defects less than 1 cm^2
2	Defects between 1 cm^2 and 3 cm^2
3 (juicing)	Defects more than 3 cm^2
4 (reject)	Open wounds, rots etc.

The fruit was then assessed individually for each defect type, based on the Queensland Government's 'Mango Quality Assessment Manual for Kensington Pride Fruit'. A given piece of fruit could have multiple defects at different ratings.

Results:

The results were assessed for proportions of fruit in each class (Figure 2) as well as a cluster analysis for damage using Ward's method. The actual data for all the orchards combined is recorded in Appendix 1.

Figure 1 shows the most common defects throughout the season in Darwin 1 fruit (sampled in September), Darwin 2 fruit (sampled in November) and Katherine fruit. There is a reasonable amount of variation between defects, locations and times. The most prominent defect was lenticel-spotting, particularly in Darwin, whilst in Katherine, the most common defect was cleavage-scarring. Overall, 13% of the downgraded fruit had damage that was preventable (sap-burn, foreign matter and mechanical damage). A further 10% of the damage was caused by insects, including that by russets, scales and chewing insects.

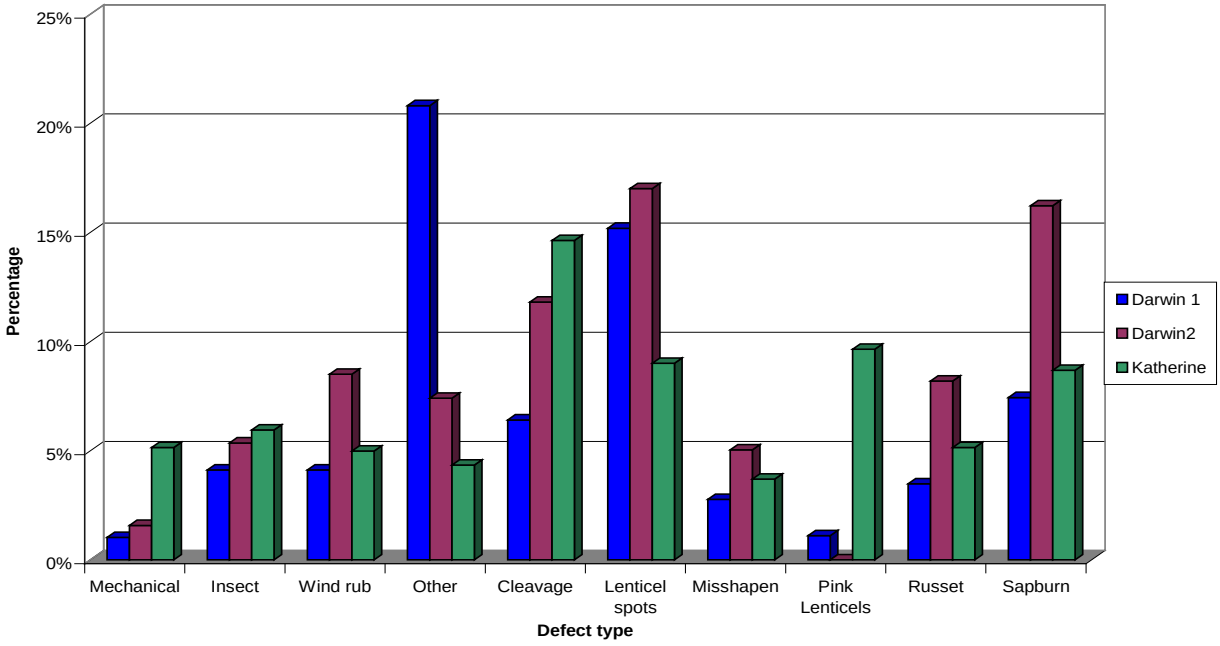

Figure 1. Major downgrading defects

Of the fruit assessed, 47% achieved first grade, compared with other classes. The average for Katherine was 48%; in Darwin 1, it was 50% and in Darwin 2, it was 41%.

Many of the common defects result from environmental factors and thus difficult to manage effectively. For example, lenticel-spots are believed to increase due to high humidity and rainfall or excessive irrigation (Cronje 2009). However, some defects, such as sap-burn and mechanical damage, are usually caused during post-harvest handling (Kernot et al. 2000) and can be prevented or reduced by proper management.

The proportions of the first three grades change according to individual orchards. The 'best' performers in terms of the proportion of first grade fruit were those growing Calypso mangoes. However, some orchards with KP fruit also performed exceptionally well. There were insufficient Calypso samples to compare the varieties directly to determine whether this was an effect of management strategies by individual Calypso growers, or an intrinsic difference between the varieties. However, it is certainly something that should be looked at in future.

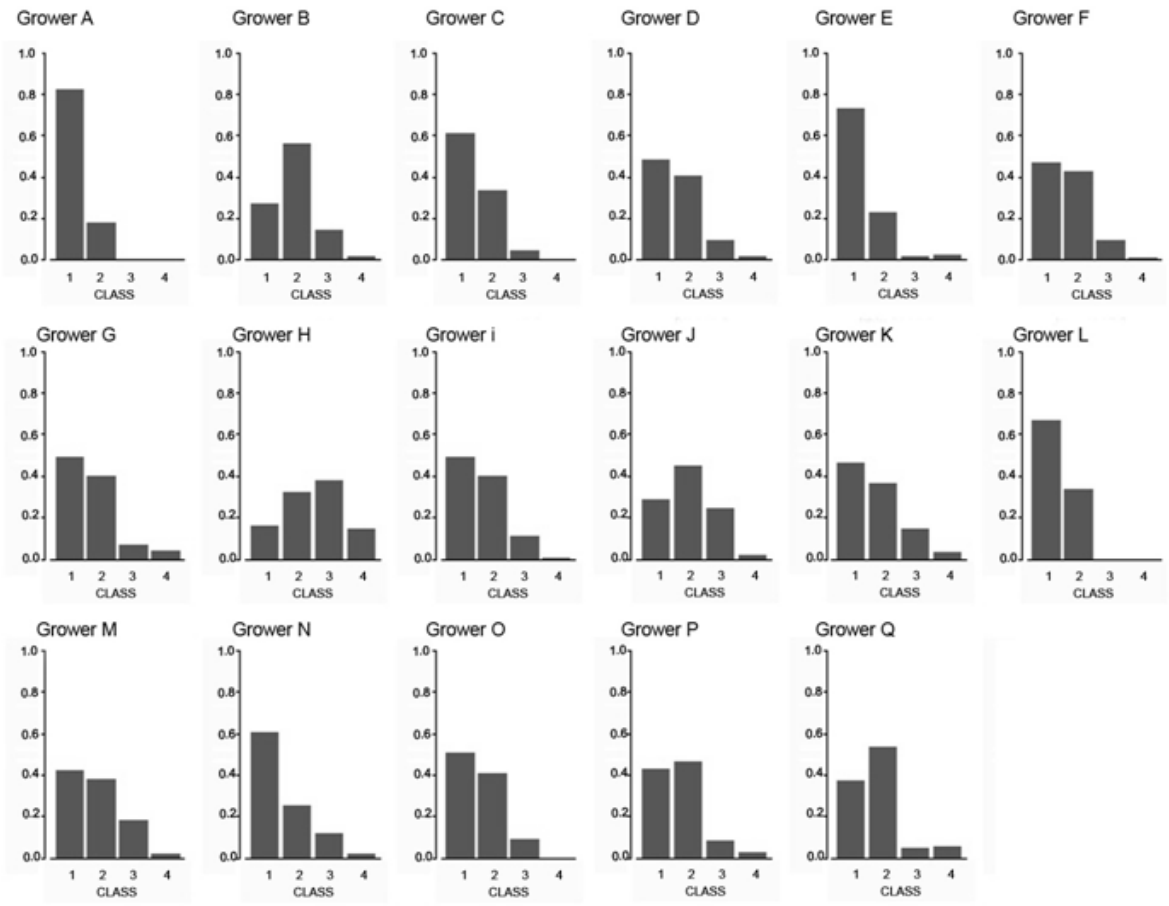

Figure 2. Individual plots of proportions of fruit in each class

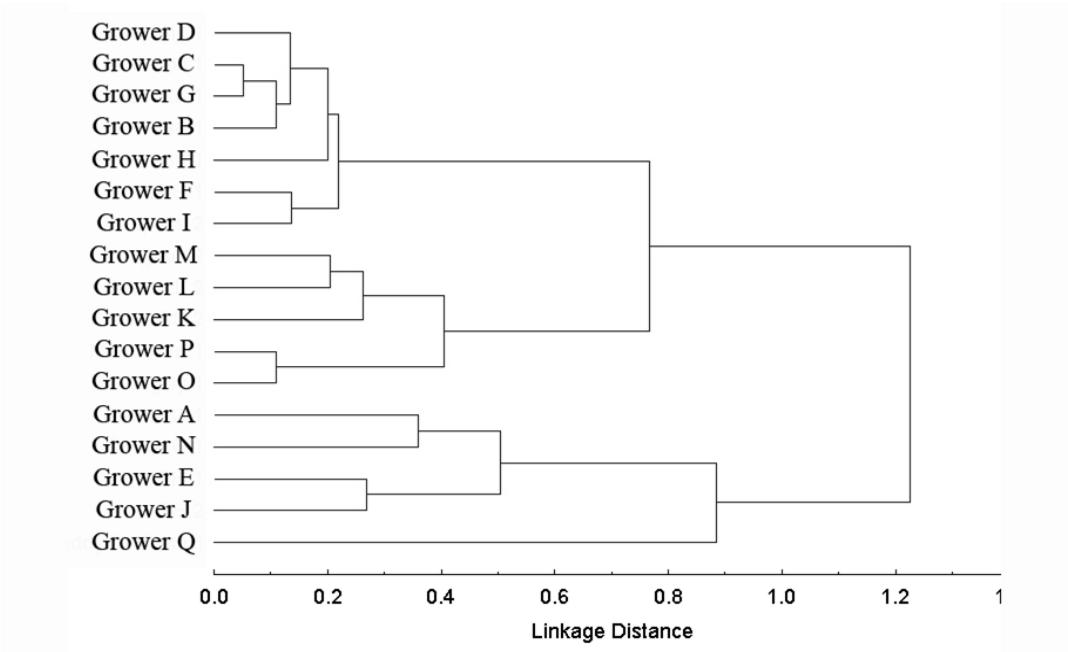

Figure 3. Tree diagram for damage (Ward's method; 1-Pearson r) excluding fruit quality

Further statistical analyses (Figure 3) of the results showed that although there were some similarities between certain growers within regions, there was enough consistency across the region. As such, there were greater differences between individual growers than between regions. For example, growers O and N are both from Katherine, but are clustered separately. The differences between orchards were greater than those between regions.

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Acknowledgement:

This project was funded by ACIAR and supported by the growers who contributed fruit for analysis.

Appendix 1: Industry defect analysis raw results

Rating	**1**	**2**	**3**	**4**
No. of fruit in each class	1193	964	294	70
Fruit in each class (%)	47	38	12	3
Wounds	2	1	1	12
Bacterial spot	0	0	0	0
Mechanical	137	43	7	5
Animal	1	0	0	0
Insect	46	77	45	1
Wind rub	141	108	29	0
Black tip	18	18	30	15
Green tip	32	9	3	0
nose crack	67	5	0	1
Other	439	259	77	1
Browning	128	49	3	0
Bruising	85	20	0	0
Cleavage	458	228	14	5
Dendritic spot	1	1	0	1
Foreign matter	13	8	13	2
Lenticel spots	700	333	23	0
Misshapen	58	70	20	0
Pink Lenticels	151	70	0	4
Russet	119	105	23	0
Sap-burn	250	197	53	1
Scab	0	0	0	0
Scale	159	5	5	0
Sticky fruit	1	0	0	0
Under skin browning	18	9	5	0
Greying	22	30	3	1
Sunburn	4	3	0	0
Stem bruise/ puncture	71	0	0	0

PROJECT: Testing and Deployment of *Khaya senegalensis* (African Mahogany) Clonal Material

Project Officers: **D. Reilly, R. Connelly, P. Bergin and G. Nikles and G. Dickinson of the Queensland Forestry Research Institute**

Location: Darwin, Katherine

Keyword(s): Mahogany, *Khaya senegalensis*, genetic selection, clone, genotype

Objectives:

To rapidly improve stem straightness and produce diverse, second-generation progeny.

To establish a series of clone tests to identify superior genetic lines for further deployment and commercial adoption.

To match Khaya senegalensis to suitable sites and determine optimal silviculture, nutrition, management and ways to improve the timber.

To improve the knowledge of staff in genetics and tree breeding.

Background:

The broad genetic base of *Khaya senegalensis* (Ks) in the Northern Territory (NT) comprises mainly of stands planted in the early 1970s. Primary breeding objectives are to rapidly improve stem straightness and reduce the occurrence of large branching, while maintaining the high quality characteristics of the timber through a joint study by Queensland, the NT and the Rural Industries Research and Development Corporation. Timber characteristics and drying requirements have been evaluated.

Method:

Selected superior trees were grafted and planted in December 2001 in a clonal seed orchard (CSO) and a gene re-combination orchard at Howard Springs and Berrimah Farm, respectively. In October 2003, 38 mature age trees were selected and harvested for a timber evaluation study of the species. Most of the trees were taken from Gunn Point and were among the original 96 selected for superior phenotypic characteristics, which were grafted and planted in the CSO. The remaining trees were selected from known provenances planted at Howard Springs and were of the same age. Of the harvested trees at Gunn Point, 11 contained mature seed pods, which were collected and planted at Berrimah Farm in family rows. With the addition of seedling material from Weipa in north Queensland and vegetatively-produced cuttings from selected trees at Howard Springs, the first hedge garden (HG) was established in March 2004.

Further additions have been made with coppice (stump re-growth) from some of the harvested trees at both orchard sites. More than 560 plants in the HG have the potential to yield cuttings that can be propagated and deployed as rooted cuttings (RCs) in clone tests. There are now 10 clone tests established at the following locations: the Coastal Plains Research Farm, the Douglas Daly Research Farm (DDRF) and the Katherine Research Station. Tests are also conducted on commercial properties on Melville Island in the north to the Douglas/Daly region and Katherine in the south.

This project continues to test clonal material of Ks across a number of sites and locations. The vegetatively propagated rooted cuttings are evaluated against each other and compared with seedling controls at DDRF on a free-draining Blain soil, using two seedling sources as controls: (1) Darwin street trees (DST) and (2) a provenance from Mali, called Favako. All other planted material at the site was propagated from cuttings accessed from the Berrimah Farm HG.

Table 1. The composition of *Khaya senegalensis* clone trial at DDRF

HG	Family ID	No. planted at DDRF
1-33	3	21
34-49	10	0
50-66	18	8
67-99	12	8
100-132	19	5
133-165	70	10
166-231	122	8
232-264	151	8
265-297	158	14
298-330	166	8
331-350	169	9
351-363	Weipa	2
430-528	Weipa	17
364-429	Howard Springs wildling	15
529-557	Stump coppice / Rooted Cutting	8
580	DST	19
595	Mali	20
Total		180

The trees were planted between 13 and 14 January 2010, containing five replicates (reps). Thirty three RCs had three ramets (copies of clones, derived from asexual propagation) and were therefore represented in three reps; 26 RCs had four ramets and were represented in four reps; 82 had five ramets and were represented in five reps.

The trial site at DDRF was surveyed and pegged on 22 December 2009 and pre-planting grass and broad-leaf herbicides were applied only to planting rows (1 m wide). The herbicides consisted of a mixture of Glyphosate 450 at10 mL/L and Starane at 8 mL/L with LI700 as buffer/wetter. Around 29 December 2009, the plantings were put out in rows with a single tyne ripper to a depth of 0.5 m. This ensured minimum disturbance to the site with the inter-rows intact with standing vegetation.

Table 1 shows that RCs from all families in the HG are represented, except HG 10 in this clone test, in addition to seedlings derived from DST (entry 580) and Favako (entry 595)] as controls. In replicates four and five, clone numbers with insufficient ramets were replaced with 'fills' of DST.

The single surround row in this trial was planted with excess RCs from previous years, with the exception of the western side where the RCs were of unknown origin. The trial is set out in five replicates with six incomplete blocks in each replicate, made up of 5 rows x 6 trees, resulting in 180 trees per replicate. Rows are 4 m apart, with 2 m between trees.

All plants, clones and seedlings were raised in Queensland forestry tubes prior to planting out. Initially, the RCs were propagated in 'hyco' trays, consisting of cells of about 93 mL volume, in a mix of perlite and coco-peat, and placed in a misting house.

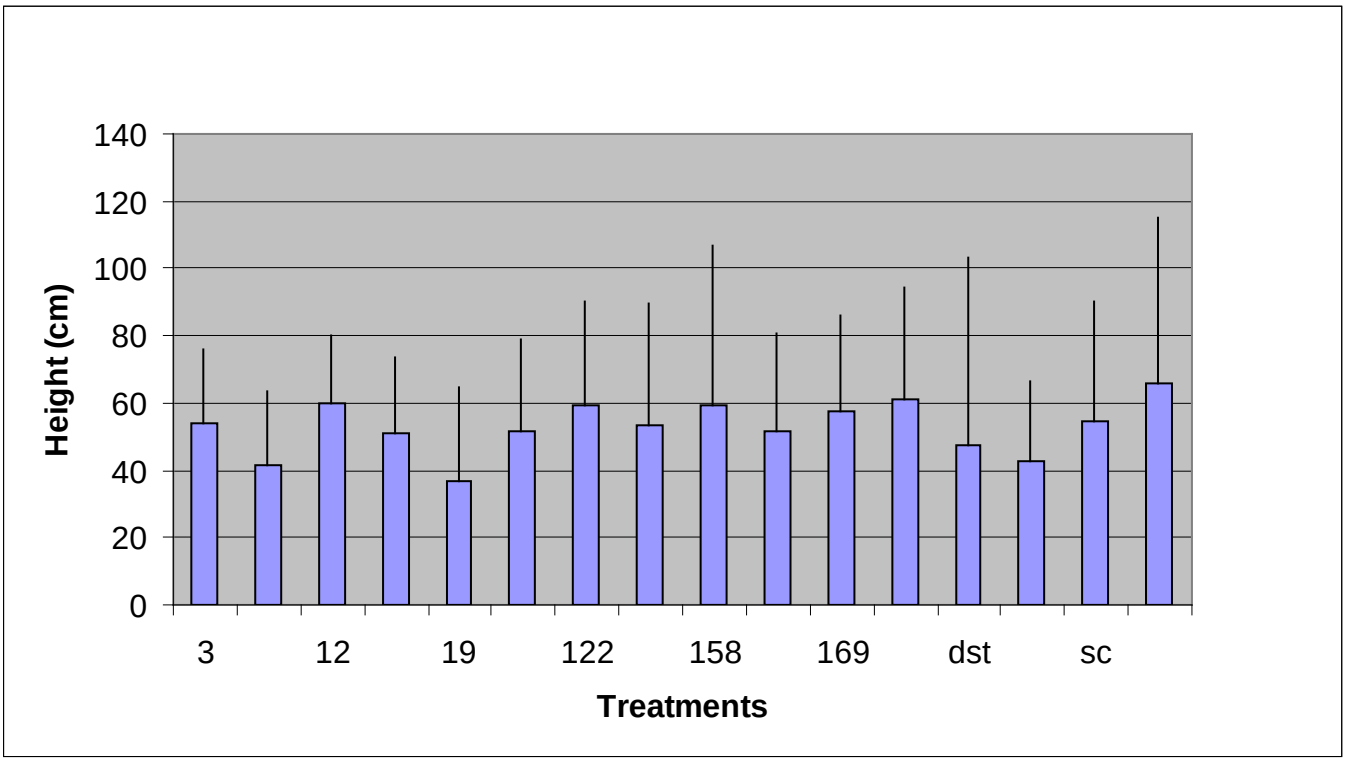
The collection of cuttings began in May 2009, aiming at specific HG numbers. However, it became a case of collecting what was ready and available from May to July, 2009. As the cuttings formed roots and developed, they were transferred to the larger 'forestry tubes' and grown for another three to four months.

Although the weather conditions were favourable during the time of planting (overcast and raining on 13 and 14 January, 2010), the following weeks were dry, with little or no rain. When the first post-planting herbicide was applied on 10 March 2010, a number of Ks plants were dead or lacked vigour. The weeds at this time appeared to be out-competing the cuttings and seedlings of Ks. Weeds were controlled with a grass knock-down herbicide and broad-leaf herbicide, prior to applying fertiliser.

The fertiliser used at planting consisted of 100 g of triple super (NPKS, 0-18-0-2) and Nitrophoska Blue special, (N-12, P-5.2, K-14.1, S-6, Ca-4.3, Mg-1.2, Zn-0.01, B-0.02 and Fe-0.05) applied in two holes adjacent to each seedling. Fertiliser was also applied to individual trees on 16 and 24 March 2010. Rain was recorded in the weeks after fertilising.

Results:

An initial assessment of the trial was undertaken in May 2010. Many plants demonstrated signs of drought stress and grazing damage from wallabies, resulting in some plants having double or multi-leaders. Such plants will not maintain a single straight stem (one of the main traits being selected for) and will be removed at the first thinning at four to five years old or earlier. At the time of assessment, trees with more than one 'leader' constituted 9.2% of the trial trees. In addition, 11.4% of the trees died. Even with these losses, the replicated trial had an 80% survival rate. It is therefore considered worthwhile to continue monitoring to identify trees/families that are best suited to the conditions at this particular site.



Error bars represent standard deviation.

Figure 1. Height of *Khaya senegalensis* clones at DDRF four months after planting

The heights of seedlings four months after planting are shown in Figure 1. Considerable variation existed within and between clones and seedlings at this early stage of the trial. The best performing clones and seedlings at the time of assessment were the 'wildling' cuttings from the HG, followed by the 'Weipa' cuttings and then cuttings from family number 12. The trial will continue.

PROJECT: Establishment of a Second Generation/Progeny Seed Orchard of *Khaya senegalensis* (African Mahogany)

Project Officers: **D. Reilly, D. Marcsik, R. Connelly, P. Bergin, Mike Kahl, with G. Nikles and G. Dickinson of the Queensland Forestry Research Institute**

Location: Darwin, Katherine,

Keyword(s): Mahogany, *Khaya senegalensis*, recurrent selection, clone, seed orchard

Objectives:

To rapidly improve stem straightness and to produce diverse, second-generation progeny.

To begin the establishment of a second-cycle base population that is superior in performance, diversity and numbers of trees compared with the first-cycle base population.

To enable statistical comparisons of the new material and various controls.

To improve the knowledge of Department of Resources staff in genetics and tree breeding.

Background:

The second wave of domestication of *Khaya senegalensis* (Ks), a high value hardwood, began in the Northern Territory (NT) in 2000-01, following the initial introductions and evaluations by the former Forest Bureau in the 1950s. Since 2007, the evolution of Ks towards a commercial plantation species has advanced in a revolutionary way (Nikles et al. 2009). The species is now being planted on a large, industrial scale in the NT and to a lesser extent in northern Western Australia and in Queensland. More than 9000 hectares are now planted in the NT, mostly based on new local African seed obtained annually since about 2005. More than 48 000 hectares have been purchased in the Douglas/Daly region in the past few years by four managed investment scheme companies to plant Ks on a significant proportion of that land. A plantation program of this size warrants a domestication program based on recurrent selection, especially in view of the obvious need for genetic improvement of many characteristics and the large variation in wood quality (Armstrong and Reilly 2004). The concept of recurrent selection is a means of species domestication via generational selection, mating and progeny establishment.

The major source of seed for the establishment of the second generation progeny test/seed orchard was obtained from the clonal seed orchard planted in 2001 at Howard Springs, where 30 clones and 39 ramets (copies of clones derived from asexual propagation) of the original 98 planted in stage 1, yielded seed in 2008-09. In addition, infusions (new or different material) were obtained from African provenances provided by commercial plantations companies and selected 'superior' trees from Queensland and Katherine orchards.

The seedling progeny trial was conducted at Katherine Research Station, where the climate is more typical of the major plantation regions than Darwin (site of the first seed orchard) and where the conditions are more likely to induce early flowering with longer and cooler dry seasons. The seed orchards established in the Darwin region in 2001 at Howard Springs and Berrimah Farm have yielded only minimal amounts of seed up to now, with the

exception of 2008-09 when approximately 40 trees produced seed from the first general heavy flowering. It is hoped plants in the facility established in Katherine will flower early. However, the seed orchard in Katherine is based on open-pollinated seeds, while the earlier trials were established with grafted clones of reproductively mature trees.

Method:

Second generation seed was collected from the established clonal seed orchard at Howard Springs through open pollination of the 98 clones (four ortets - progeny from a single parent- of each) in the dry season of 2008. Unfortunately, the 2008 seed crop included only one of the 'top 15' phenotypes selected for wood properties. As this was the first general seed crop harvested from this facility, it was not possible to apply selection pressure based on parental phenotype or wood property characteristics, which had been previously investigated. A summary of the seed orchard clone numbers, provenance origin, 'bulk' control seed entries and their position in each replicated block is provided in Table 1.

Table 1. Composition of the second generation *Khaya senegalensis* seed orchard

Entry No	Clone No.	Ramets	Provenance	Position in the seed orchard	Healthy seedlings available for planting	No. reps in blocks A&B (and entries used as fills)
1	5	1	Uganda 10053	R18T5	50	6, 5
2	5	1	"	R24 T6	50	"
3	9	1	New Cal 520? /522?	R0 T5	50	"
4	20	1	Uganda 9620	R15 T5	50	"
5	21	1	Nigeria Jos D480	R15 T6	50	"
6	27	1	Nigeria Yola D486	R25 T7	50	"
7	30	1	New Cal D477	R24 T4	50	"
8	31	1	Senegal D417	R25 T11	50	"
9	36	1	Nigeria Jos D480	R19 T4	37	5, 4 (entry 42 in A, entry 45 in B)
10	50	1	Senegal D417	R26 T7	50	6, 5
11	52	1	New Cal D487	R2 T3	50	"
12	61	1	Nigeria Yola D486	R23 T11	50	"
13	62	1	Nigeria Yola D486	R9 T3	50	"
14	63	1	Ivory Coast 10050	R19 T2	50	"
15	63	1	"	R22 T11	47	"
16	63	1	"	R26 T10	50	"
17	65	1	Nigeria Jos D480	R15 T2	50	"
18	66	1	Nigeria Jos D480	R12 T9	50	"
19	67	1	Sudan 9687	R24 T10	50	"
20	77	1	Senegal 9392	R19 T12	50	"
21	78	1	Togo D411	R17 T6	47	"
22	83	1	CAF D391	R4 T6	50	"
23	94	1	Uganda 9620	R12 T7	48	"
24	94	1	"	R23 T7	50	"
25	96	1	Senegal 9392	R22 T9	40	5, 5 (entry 48 in A)
26	102	1	Uganda D407	R26 T11	50	6,5
27	102	1	"	R14 T5	50	"

Entry No	Clone No.	Ramets	Provenance	Position in the seed orchard	Healthy seedlings available for planting	No. reps in blocks A&B (and entries used as fills)
28	104	1	Uganda D407	R10 T6	50	"
29	104	1	"	R30 T10	50	"
30	107	1	CAF D391	R17 T11	21	3, 2 (entries 42 & 45)
31	113	1	Sudan 9368	R17 T1	50	6, 5
32	116	1	Uganda D407	R17 T2	50	"
33	116	1	"	R5 T8	50	"
34	118	1	Unknown	R16 T1	39	5, 4 (entries 42 & 45)
35	119	1	Unknown	R9 T9	50	6, 5
36	119	1	"	R11 T4	50	"
37	121	1	Senegal D417	R5 T1	38	5, 4 (entries 42 & 45)
38	121	1	"	R19 T10	50	6, 5
39	124	1	Uganda 10053	R27 T11	50	"
40	1KS6-020	1	Queensland	NA	20	3, 2 (entries 42, 45 & 48)
41	1KS6-021	1	Queensland	NA	22	3, 2 (entries 42, 45 & 48)
42, 43, 44	Burkino Faso (Bulk)	>10	Tiefora / Burkino Faso	NA	354	6 x 3, 5 x 3
45, 46	Favako	Mali, Favako	Bulk	NA	133	6 x 2, 5 x 2
47, 48	Goldings	Unknown	Bulk ex 3 trees	NA	115	6 x 2, 5 x 2

Forty eight entries were planted in blocks A and B, with six replicates (rep) in block A and five in block B. Each rep consisted of six incomplete blocks, with 32 trees per incomplete block in four rows of eight trees. The reps therefore contain 192 trees each randomly allocated and planted in four-tree line plots. The aim of the trial is to leave only the best one of the four trees per line plot after culling the poorest trees beginning at the age two to three years. The first thinning will bring the population density to 660 stems/ha). Another thinning at five to six years will leave two of the original four and reduce the density to 440 stems/ha. Another thinning at the age of eight to ten years will leave just one of the original four with 220 stems/ha. This will ensure only the best trees remain for progeny testing, which will be the source of second generation selects, seed, scions, and vegetative propagules for further breeding work. These trees will also act as a source of material for future vegetative propagation in clonal seed orchards, tissue culture work and serve as a comparison for first and second generation trees.

Results:

Seedlings were planted in blocks A and B between 16 and 24 February 2010, during very hot weather. The irrigation system, which had been installed during site preparation, was utilised for 'wetting' planting rows to ensure the survival of newly-planted seedlings. At the end of planting, all blocks were assessed for plant survival and initial plant heights were measured. Where plants had died, they were replaced if sufficient extra plants were available. Seed lots with insufficient numbers to complete all replicates were replaced with the bulk entry of Burkina Faso seedlings, which had recently been imported from Africa. The plants in the trial blocks will be further assessed at appropriate intervals in order to select superior phenotypes.

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PROJECT: The Reproductive Biology and Breeding Systems of African Mahogany

Project Officers: D. Marcsik, P. Bergin and D. Reilly

Location: Darwin

Keyword(s): African mahogany, *Khaya senegalensis*, flower biology, breeding systems

Objectives:

To understand the reproductive biology and breeding systems of African mahogany in order to develop protocols for controlled pollination.

To use this information to breed superior African mahogany hybrid seedlings.

Background:

A major impediment to the advancement of the African mahogany (*Khaya senegalensis*) (Ks) tree improvement program has been poor and inconsistent flowering and low seed production of clones in seed orchards. This is of significant concern as seed orchards are the primary source of sufficient genetically- improved seeds.

An understanding of the reproductive biology and breeding systems of Ks is necessary to successfully manage clonal seed orchards for efficient seed production and for breeding superior hybrid seedlings. Very little published information is available on the reproductive biology and breeding systems of Ks and, in general, on the family Meliaceae. Styles (1972) reported that most of the species in Meliaceae were monoecious flowering trees, having separate male and female flowers on the same tree. Recent published work by Gouvea et al. (2007) and Gouvea et al. (2008) describes in detail the floral developmental stages and differentiation of unisexual flowers in the genera *Cedrela*, *Swietenia* and *Toona*.

In 2009, preliminary flowering experiments were conducted to investigate the breeding systems of Ks, such as when female flowers are present on the inflorescence, the proportion of male to female flowers and whether the flowers are self-compatible. Observational notes on the structure of Ks flowers are also provided.

Method:

Two flowering experiments were conducted during the 2009 flowering period for Ks. In experiment 1, a large mature flowering Ks tree located near the Post Entry Quarantine facility at Berrimah Research Farm was chosen as the pilot tree for detailed investigations of Ks inflorescence. Three inflorescences, consisting of immature green buds, were randomly selected on the tree. Each was labelled with a jeweller's tag with the inflorescence number (Figure 1). Each inflorescence was observed from the onset to the termination of anthesis by removing the newly-open flowers at two to three-day intervals. The harvested flowers were placed in vials labelled with the corresponding inflorescence number and taken back to the laboratory for examination. The number of male and female flowers were counted and recorded. In addition, open flowers both male and female, were collected and their floral structure was closely examined using a stereo microscope.

For experiment 2, six clones with more than 50% flowering on the tree were selected from the Howard Springs seed orchard. Six inflorescences per tree were randomly selected at the immature green-bud stage (Figure1), three inflorescences were covered with an insect-proof nylon-mesh bag and the remaining three were left un-bagged or open. Coloured flagging tape was used to label the main branch of the inflorescence with both the tree and inflorescence number. At seven-day intervals, the bags were removed from the inflorescences and the senesced flowers were harvested. The flowers were placed in vials labelled with the corresponding tree and inflorescence number and taken back to the laboratory for examination as described in experiment 1. At the end of the flowering period a fruit count was conducted of both the bagged and un-bagged inflorescences for each clone.

Figure 1. Ks inflorescence at the onset of anthesis

Results:

Flowering experiment 1

The highest frequency of female flowers produced on Ks inflorescences was 17 days after the start of anthesis, with smaller numbers produced during the remaining flowering period (Figure 2). There was an overall predominance of male flowers observed within the Ks inflorescences from the onset to the termination of anthesis.

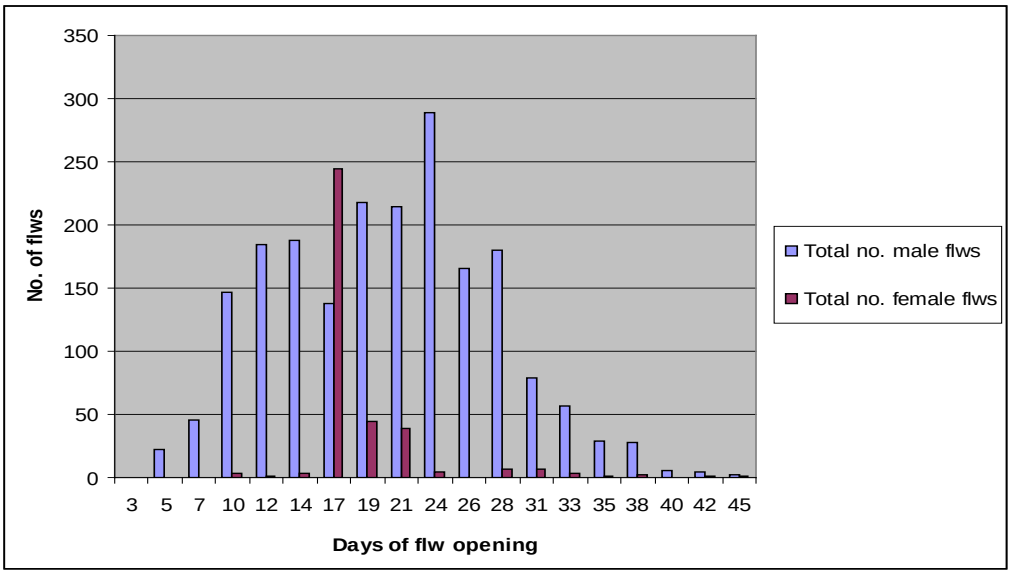

Figure 2. Frequency of male and female flowers within Ks inflorescences observed at two to three-day intervals from the onset to the termination of anthesis

The flowers of Ks are functionally unisexual or perfect. The functional male flower has large, yellow anthers full of pollen compared with the functional female flower, which has smaller brown anthers with no pollen. Furthermore, the pistil of the functional female flower has a swollen round ovary and a short style. This compares with the pistil of the functional male flower, where the ovary is not swollen and the style is longer (Figure 3).

Scale bar = 1 mm

Figure 3. Partially dissected open Ks female flower (left) and Ks male flower (right)

Flowering experiment 2

In general, the un-bagged or open inflorescences had a larger number of pods set compared with bagged Ks inflorescences. The number of pods set per open inflorescence ranged from two to 94, compared with bagged inflorescence that ranged from one to four pods (Table 1). The nylon-meshed bag covering the inflorescence acted as a barrier to pollen transfer by wind and by insects. However, insects such as ants were found inside the bags when removed from the inflorescence to harvest the flowers. The presence of insects suggests they could be transferring pollen and self pollinating the flowers in the bag.

Table 1. The number of pods set on both bagged and un-bagged (open) inflorescences for six different Ks clones

Clone No.	Bag 1	Bag 2	Bag 3	Open 1	Open 2	Open 3
9	0	0	0	0	9	8
10	1	0	0	#	32	0
22	0	0	0	6	10	4
62	2	2	0	27	0	4
88	0	0	4	2	17	#
107	4	2	3	33	0	94

# indicates tag lost, chewed off by ants.

In general, a similar result to flowering in experiment 1 regarding the predominance of male flowers was observed within bagged Ks inflorescences for all six clones (Table 2). The proportion of male to female flowers within the

bagged inflorescences was found to vary between the clones from 1:1 up to 10:1 in Clone 22 and Clone 88, respectively (Table 2).

Table 2. The proportion of male and female flowers observed within bagged Ks inflorescences for six different Ks clones

Clone no.	Total no. of male flowers	Total no. of female flowers	Male : female ratio
9	1610	509	3:1
10	2852	980	3:1
22	573	468	1:1
62	1153	482	2:1
88	1095	106	10:1
107	1523	427	4:1

Discussion:

The preliminary results from the two flowering experiments greatly contributed to the understanding of the reproductive biology and breeding systems of Ks. The observational studies of the inflorescences confirm Ks trees to be monoecious, producing both male and female flowers (Styles 1972). It was observed that both male and female flowers could be distinguished by the naked eye when they were fully open within inflorescences through differences between stamens and pistils. This is of great benefit in controlled pollination experiments as it reduces the difficulty of having to remove the stamens before applying pollen to the very small flowers.

Observations of Ks inflorescences from the onset to the termination of anthesis found the highest frequency of female flowers produced within the inflorescence was at 17 days. This finding is of great benefit when developing protocols for controlled pollination as the frequency of female flowers can now be predicted within the Ks inflorescence. Throughout the period of anthesis, a predominance of male flowers was observed within the Ks inflorescence, similar to that found in other Meliaceae species such as *Swietenia*, *Cedrela* and *Toona* (Styles 1972; Gouvea et al. 2007; Gouvea et al. 2008). Lee (1967) found that whole trees of *Swietenia mahagoni* usually have approximately ten times as many male as female flowers. In the preliminary study, the ratio of male to female flowers in Ks trees varied considerably from 10:1 to 1:1. Further investigations will be conducted next flowering season on the number of male to female flowers to determine if there is a difference from year to year. Also, studies will be conducted to determine the position of male and female flowers as well as the opening pattern of female flowers within the inflorescence.

Preliminary results (Table 1) indicate that both self and cross pollination is possible in Ks, since pod set occurred in both the bagged and un-bagged inflorescences. The un-bagged inflorescences had a larger number of pods set compared with the bagged inflorescences where pollen transferred by wind and by insects was restricted. Pod set within bagged inflorescences could have been the result of self pollination by insects found inside the bag. The difference in the number of pods set between bagged and un-bagged inflorescences suggests that cross pollination may be the dominant reproductive system in Ks. Detailed studies will be conducted next flowering season on the stigma receptivity and pollination mechanism of Ks. In addition, studies will be conducted on pollen viability and pollen storage.

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PROJECT: Demonstrate and Evaluate Mulch Management Techniques in a Legume–Cereal Rotation during the Dry Season

Project Officers: M. Bennett, D. Renfree and M. Kahl

Location: Katherine

Keyword(s): maize, peanuts, crop rotation, cover crop

Objective:

To demonstrate and evaluate mulch management techniques in a legume–cereal rotation.

Background:

A major constraint to growing peanuts continuously is the build-up of diseases and legume weeds (Crosthwaite 1994). One of the better strategies available to ameliorate this problem is to combine crop rotation with no-till sowing methods into cover crop mulch (Peake et al. 1985). Maize grows well in rotation with peanuts, while millet is the ideal cover crop for northern Australia (personal experience).

Peanuts are the principal broad-acre irrigated grain crop in the Northern Territory (NT); 4100 tonnes were produced in 2009 (Department of Resources 2010) and are normally harvested the third or forth week of September, while maize is sown in the first week of November. During this intervening period (September to November) the soil is exposed to wind and water erosion due to the removal of peanut stubble for hay. To minimise erosion and provide a favourable environment for the establishment and growth of maize seedlings, a millet cover crop can be grown. The cover crop provides mulch into which no-till maize can be sown.

In this demonstration trial, a millet cover crop was grown for 29 days and then sprayed with glyphosate to terminate its growth in order to determine the effect of different mulch management strategies on maize establishment, nutrition and yield. The millet mulch was either incorporated into the soil, slashed or left standing (treatments). A maize (*Zea mays)* crop was then sown into the mulch and its development and grain yield were measured.

Method:

The demonstration was conducted at Katherine Research Station (lat. 14°28'S, long. 132°18'E, altitude 108 m) in 2009. The soil is a red earth (Kandosol) (Isbell 1996) of the Fenton series (Lucas et al. 1985).

Design
Demonstration plots were established in the eastern side (A-series) of Putland paddock (north, central and south). Plots were 1.1 to 2.2 ha in size and were superimposed on legume hay pastures (Table 2), including lablab (*Lablab purpureus* cv. Highworth), lucerne (*Medicago sativa* cv. L56 and L90) and Cavalcade (*Centrosema pascuorum* cv. Cavalcade).

Land preparation
The legume pastures in the demonstration site were baled on 18 to 20 March, 2009. Round-up CT (Glyphosate 450 g/L) was applied at 5.0 L/ha to kill pasture regrowth one week prior to sowing the millet (*Pennisetum americanum* cv. Ingrid pearl) cover crop. Ingrid pearl millet was sown using no-till in 0.5-m row spacing at 18 kg/ha on 09/04/2009. The cover crop was then sprayed 29 days after sowing (DAS) with Round-up CT at 2.0 L/ha to terminate its growth.

Three mulch management treatments (plots) were applied randomly to the north of Putland paddock (3.3 ha), re-randomised and applied to the central and south Putland paddock (9.9 ha). Mulch management treatments included mulch cultivated by disc plough (x2) and then harrowed (cultivated), mulch slashed at a height of 9 cm (mulched), and mulch left standing (standing).

Maize (cv. 31G66) was sown in 0.75-m row spacing at 72 000 seeds/ha on 18 to 19 May, 2009. Gesaprim 600 SC (Atrazine 600 g/L) at 2.5 L/ha, Dual Gold (Metolachlor 960 g/L) at 1.5 L/ha and Round-up CT at 2 L/ha were applied to all plots immediately after sowing. Good weed control was achieved throughout the trial.

Seasonal conditions
Katherine has a monsoonal climate with a well-defined hot, wet season from November to March and a dry season for the remainder of the year. Total rainfall for April to September 2009 was 0.0 mm. Average daily maximum temperature during April, August and September was approximately 1°C warmer than the long term average.

Irrigation
Millet and maize plots were irrigated twice a week to maintain a full soil moisture profile. The millet cover crop received 194 mm/ha (9 April – 8 May 2009) and the maize plots received 804 mm/ha (18 May – 28 September 2009).

Fertiliser
No fertiliser was applied to the millet cover crop. Total fertiliser application/ha for the maize plots was 283 kg nitrogen (N), 77 kg phosphorus, 128 kg potassium (K), 51 kg sulphur plus trace elements. Diammonium phosphate was broadcast while the remaining fertilisers were applied through fertigation.

Insect control
Heliothis caterpillars (*Helicoverpa armigera)* were controlled by applying Vivus Max (*Nucleopolyhedrovirus*) at 220 mL/ha through the irrigation water on 9 July (52 DAS), 30 July (73 DAS) and 13 August (87 DAS). Heliothis control was successful.

Measurements
Millet cover crop development
Millet biomass was measured at 29 DAS. Three 1-m^2 quadrats per plot of plants were cut at ground level, dried at 80 °C and their dry weights were recorded.

Maize development

Maize emergence and establishment populations were measured at 7 and 14 DAS, respectively. The number of plants was recorded for ten 2-m rows per plot.

Maize harvest populations and grain yields

At harvest, plants in three quadrats per plot were measured. Each quadrat consisted of two rows 2.2 m long. The number of plants per plot was recorded and all cobs were collected. The cobs were dried at 80 °C for 96 hours, threshed and the dry weights were recorded. The data was converted to kg/ha and rounded to the nearest 100 kg.

Combine harvested grain yields

After hand harvest, a combine harvester was used to measure commercial grain yields from large sample areas. Each sample area was four rows of 50 m. Maize cobs were threshed by the combine and the grain was delivered to a mobile weigh bin. Grain weight and grain moisture content were measured.

Results:

Millet cover crop development

Millet biomass for north Putland and south Putland was 1.3 and 1.2 t/ha, respectively. Millet growth in central Putland was 40% less (0.8 t/ha) due to bird damage. No further data will be presented for central Putland due to bird damage affecting maize establishment in the mulched and standing millet treatments.

Maize development

Mean number of emerging plants/ha were 57 000 and 56 000 for north Putland and south Putland, respectively. The mean number of establishing plants/ha was 57 000 and 60 000 for north Putland and south Putland, respectively. The number of emerging and establishing maize seedlings in central Putland was significantly reduced by bird damage.

Maize plant harvest populations and hand harvest grain yields

Maize plant populations and grain yields were lower in the cultivated treatment compared with the treatments with surface mulch (Table 1). Mean maize plant population for the whole paddock was 64 000 plants/ha. The difference in harvest population between the cultivated and surface mulch treatments was approximately 8000 plants/ha.

Mean maize grain yield for the Putland paddock was 13 400 kg/ha. Grain yields were approximately 1500 kg/ha higher in the surface mulch treatments than in the cultivated treatment. The higher maize grain yields in the lablab plots reflected the initial higher nutrient status (N, K and zinc) of maize plants growing in these plots.

Table 1. The effect of millet mulch on maize plants population and grain yield

Putland paddock (legume)	Mulch treatment	Harvest population (plants/ha)	Hand harvest grain yield[1] (kg/ ha)
North (Lablab)	Cultivated	59 000	12 700
	Mulched	67 000	14 200
	Standing	70 000	14 100
South (Cavalcade)	Cultivated	60 000	12 200
	Mulched	67 000	13 600
	Standing	66 000	13 800

[1] Expressed at 12% grain moisture

Combine harvested grain yields

The mean maize grain yield in the Putland paddock was 11 500 kg/ha. The cultivated treatment in the Cavalcade rotation plots was approximately 800 kg/ha less than in the surface mulch treatments. Maize grain yields were

similar regardless of mulch treatment in the lablab plots (Table 2). Plant population was more critical in combine-harvested yields than the effect of mulch across large areas.

Table 2. The effect of millet mulch on combine-harvested maize grain yield

Putland paddock (legume)	Mulch treatment	Combine-harvested grain yield[1] (kg/ha)
North (Lablab)	Cultivated	11 600
	Mulched	11 600
	Standing	11 800
South (Cavalcade)	Cultivated	10 800
	Mulched	11 700
	Standing	11 600

[1] Expressed at 12% grain moisture

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PROJECT: Enhancing Water Use Efficiency in Table Grape Production in Central Australia

Project Officers: M. Hidalgo, G. Oliver and S. Raghu

Location: Alice Springs

Keyword(s): water use efficiency, table grapes, sustainable horticulture, soil moisture

Objectives:

To assess the effect of irrigation rate on table grape plant performance and yield.

To evaluate the effect of irrigation rate and mulch on water infiltration.

To demonstrate the utility of simple soil moisture monitoring tools in sustainable arid zone plant production.

Background:

A preliminary analysis of irrigation efficiency in 2009 revealed that the application of water through irrigation systems is variable across different growers (Figure 1), with irrigation practices largely managed by trial and error. Theoretical calculations using standardised formulae have shown that current irrigation rates are excessive relative to plant need and evapotranspiration levels. Demonstrating the potential for sustained yield over multiple seasons with reduced irrigation will enable more sustainable irrigation practices in the arid zone. Optimised use of water resources will enable growers to produce more grapes with their current water allocations and provide opportunities for farm diversification in other horticultural products.

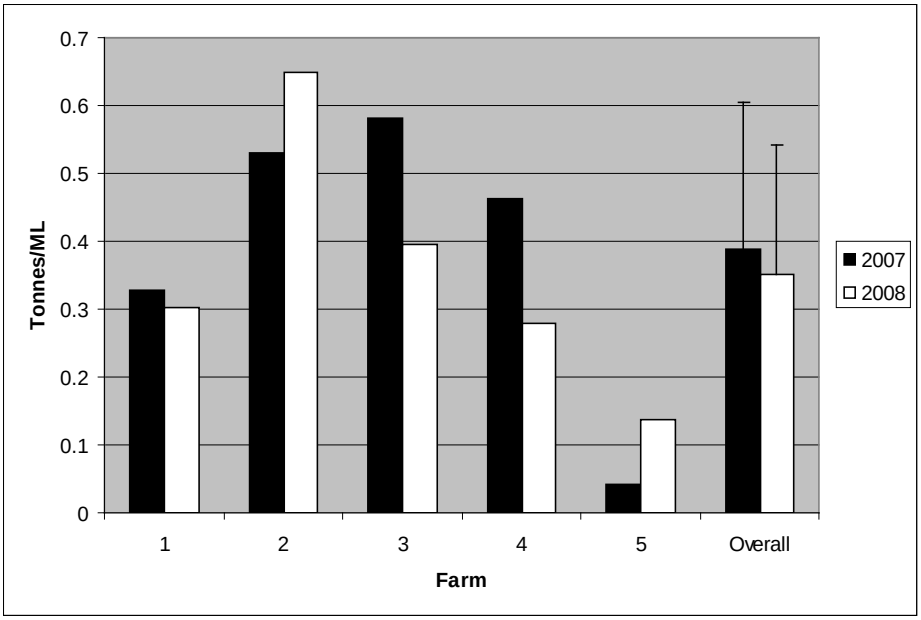


Figure 1. Water-use efficiency (yield/ML) across different table grape farms in Central Australia (2007 and 2008)

A trial to compare irrigation rates in the presence vs. absence of mulch was established on a grower property in Ti Tree in April 2010. This study will document table grape performance (phenology, yield and fruit quality) in response to these treatments. This multi-season project will record the effects of reduced irrigation rates that span growing seasons. The project is expected to continue for at least two growing seasons.

Method:

Using a randomised complete block design, irrigation (two levels: 8L/h [current rate used by grower], 4 L/h) and recycled paper mulch (two levels: present vs. absent) treatments have been established across three rows of table grapes var. Menindee Seedless at Wedgetail Farm at Ti Tree (see Figure 1). Data on plant performance (phenology of pre-defined growth stages of grapes) and yield (quantity of fruit per vine), quality (brix and brix-acid ratio at harvest) will be gathered for each of nine replicate vines. In addition, soil moisture (at 30, 60, 90 and 120 cm depth) at three replicate locations at the interaction of irrigation and mulch treatments will be collected at 2-hourly intervals using G-Bug moisture probes and automated data-logging devices.

The data will be analysed using a mixed-effects analysis of variance with irrigation and mulch as fixed factors and rows as a random blocking factor.

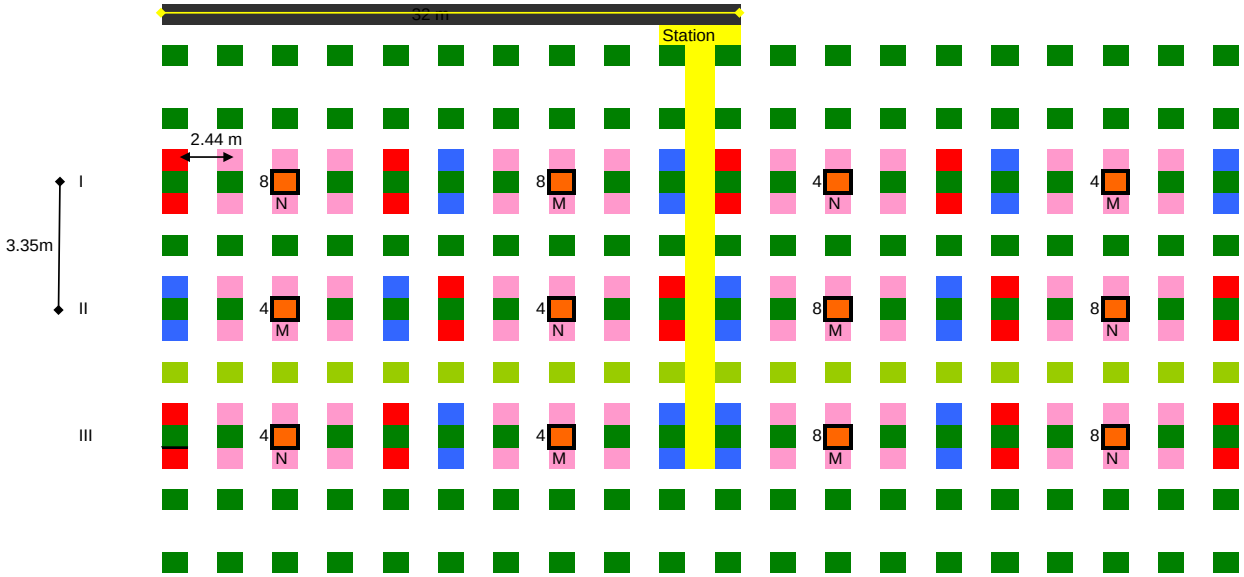


Figure 2. Layout of the field plot

Roman numerals indicate experimental rows, while Arabic numerals indicate irrigation rate (L/h). Lettering: M indicates mulch and N indicates no mulch. Each vine is represented by a single box. Boxes in pink represent vines that will be monitored for phenology and yield. Plants marked with a bold box are locations of G-Bug sensors. Inter-row and inter-vine spaces are also indicated.

Results:

Preliminary data on water infiltration are encouraging and indicate that the reduced irrigation rates are adequate to wet the root zones of table grapes and the presence of mulch enhances retention of soil moisture. Additional data on the effects of reduced irrigation rates on phenology, yield and fruit quality is needed before conclusions can be drawn on ways to improve water use efficiency in irrigated arid zone table grape production in Central Australia.

PROJECT: Cover Crops for Melons - Nitrate Recovery and Alternative Cover Crops

Project Officers: S. Bithell, N. Hartley and C. Martin

Location:	Darwin
Keyword(s):	forage sorghum (*Sorghum bicolour*) cv. Jumbo, ammonium-nitrogen, nitrate-nitrogen, mineralisation, sunn hemp (*Crotalaria juncea*)

Objectives:

To evaluate the recovery of nitrate nitrogen (N) in soil under specific cover crops in the wet season.

To quantify the level of N mineralised under these cover crops.

To evaluate alternative cover crops.

Background:

Melon production is an important horticultural industry in the Northern Territory (NT). Melons are grown in the dry season with relatively high inputs of water and fertiliser. The annual farm gate value of this industry is estimated to be $20-40 million (Anonymous 2010). During the wet season, melon growers establish rotational cover crops, mostly forage sorghum (*Sorghum bicolour*) or pearl millet (*Pennisetum typhoides*), to protect against soil erosion, melon pests and diseases, recover nutrients lost deep in the soil profile during the melon season and to prevent leaching of N mineralised from the organic fraction of the soil during the wet season.

A survey of melon farms in 1989 found that high levels of nutrients were lost through the soil profile during the growing season on some farms (Smith 1991a). At this time, fertilisers were side-dressed and overhead sprinklers were used for irrigation (Smith 1991a), while modern farms use soluble fertilisers applied through fertigation systems as standard practice. Smith (1991b) demonstrated that appropriate practices substantially reduced nitrate losses, resulting in minimal nitrate N remaining after the crop. Blackburn et al. (1987) also established that appropriate irrigation schedules with trickle tape can minimise the loss of water and nutrients through the profile. Even so, there has been no recent evaluation of potential nutrient loss or recovery in, or following, melon crops. NT melon crops are commonly grown on sandy soils having high water infiltration rates and therefore a potential for high nutrient leaching during the wet season unless the nutrients can be intercepted and recovered by a cover crop.

During the high rainfall period of the wet season, most of the conversion of organic N to mineral N in the soil (mineralisation) occurs (Norman 1966). With no cover crop, there is a potential for significant leaching of N to deep layers in the soil.

Identifying sustainable cover crops, including species and management practices, was considered a high research priority by melon growers at an industry research planning meeting in Katherine in November 2009. Limited species comparisons by CSIRO in Katherine have established that millet has a superior root system to sorghum in terms of the depth of rooting and the recovery of nutrients (Wetselaar and Norman 1960). This work, however, was carried out on Tippera soils (loamy sand), which have higher silt content than the Blains and Cockatoo types (sands or sandy loams). There has been no comparison between cover crop species on these sandier soils in the NT.

This project sought to:

1. Compare the ability of the two predominant cover crop species (sorghum and millet) to recover N on a sandy loam soil.

2. Ascertain the level of N mineralised during the wet season on sandy soils.

3. Assess the suitability of a range of alternative cover crops in melon production.

The NT Agricultural Association's '*Caring for Our Country'* project, *"Improve irrigation efficiency, nutrient and land management in Top End intensive farming systems"* supported this project by funding most of the soil analyses.

Method:

Experiment 1: Recovery of N by cover crops and assessment of mineralisation of N on sandy soils

Treatments
A field experiment with four treatments (Table 1) was started on 17 December 2009 on Blain soil at Berrimah Farm, Darwin, in an area adjacent to the Horticulture Research block. The area, which had been in pasture, was sprayed with glyphosate, mowed and residues were removed prior to light surface cultivation. The experiment was a randomised complete block design, with four blocks and each plot measuring 16 x 4 m. Neighbouring plots had a 1-m gap between, followed by a 10-m gap to the next two plots, to allow tractor access from the 16-m side of each plot. Each plot was divided into four, 4 x 4-m sub-plots with a sub-plot randomly allocated to each of four sample times. Seed was drilled with a tractor- mounted drill in rows 15 cm apart with fertiliser banded behind the seed. Fallow treatments were drilled with fertiliser only. Because of poor establishment, the millet plots were re-sown on 24 December, 2009 with different seed. As seed emergence failed both times, the plots were re-classified as 'weed treatment', where existing weeds were allowed to grow.

Table 1. Planting details

Treatments	Fertiliser (kg/ha)	Sowing rate (kg/ha)
Forage sorghum cv. Jumbo (TSW 24.7 g)	53	12.6
Katherine pearl millet (TSW 10.1 g)*	53	14.3
Chemical fallow – fertiliser	Nil	-
Chemical fallow + fertiliser	53	-

* Re-classified as weed treatment

Fertiliser consisted of 13.5% N (ammonium form), 15% P, 12.5% K and 1.2% S

Pre and post-trial sampling in the melon rooting depth zone
Samples were collected on 10/12/2009 (pre-treatment) and 14/04/2010 (post-trial) at depths of 0-150 mm and 150-300 mm using a hand auger (interior diameter 48.9 mm). Eight samples from each depth were collected from each plot and bulked. The samples were placed in a forced air oven (65 °C) on the day of sampling and dried for 48 hours. In the pre-treatment sampling, samples from each treatment were bulked again to provide four samples from each depth. For the post-trial sample, samples from each sub-plot were kept separate, which gave 16 samples from each depth. Samples were analysed by CSBP Laboratories, Lake Pibra, WA for total carbon and carbon to N ratio.

Pre and in-trial sampling to 1.8 m
Samples were collected on four occasions at one month intervals. Each plot was sampled from a randomly allocated subplot. Depth samples were collected using a tractor mounted hydraulic auger (interior diameter 47.7 mm).

At the first sampling, three blocks (12 plots) were completed on 11/12/2009 until heavy rain (347 mm from 11 to 15 December) and wet soil conditions prevented the completion of sampling until the 16/12/2009. This was the pre-trial sampling (the trial plants were planted on 17/12/2009). This sampling was from depths of 0-100, 300-400, 600-700, 900-1000 mm, 1.4-1.5 m, and 1.7-1.8 m from a single sample point in each plot, giving 96 samples.

The three remaining samplings occurred over a two-day period on 18-19/01/2010, 15-16/02/2010 and 22-23/03/2010 (in-trial sampling). The January, February and March samples were taken from depths of 0-300, 300-600, 600-900, 900-1200 mm, 1.2-1.5 m and 1.5-1.8 m from two sample points in each sub-plot and then bulked and dried. After drying, a 300 g sub-sample was sent to CSBP for analysis of nitrate and ammonium N.

Experiment 2: Alternative cover crop species

A demonstration trial was established as a randomised complete block design, which consisted of four blocks; each plot was 4 x 5 m. The demonstration compared four alternative cover species. Table 2 shows the details.

Table 2. Seed and sowing details of species target population based on percentage of viable seed in seed lot with 90% emergence

Species	Weight of 1000 seeds (g)	Sowing rate (kg/ha)	Target population (seedlings/m^2)
White French millet (*Panicum miliaceum*)	4.7	6.9	72
Siberian millet (*Echinochloa frumentacae*)	2.7	3.8	72
Sunn hemp (*Crotalaria juncea*)	33.2	37	20
Unidentified millet species (pers. comm. A. Cameron)	5.5	4.7	66

All crops were sown on 17/12/2009 using the same equipment and fertiliser rate as described for the nitrate trial, except that Sunn hemp was hand-planted.

Results:

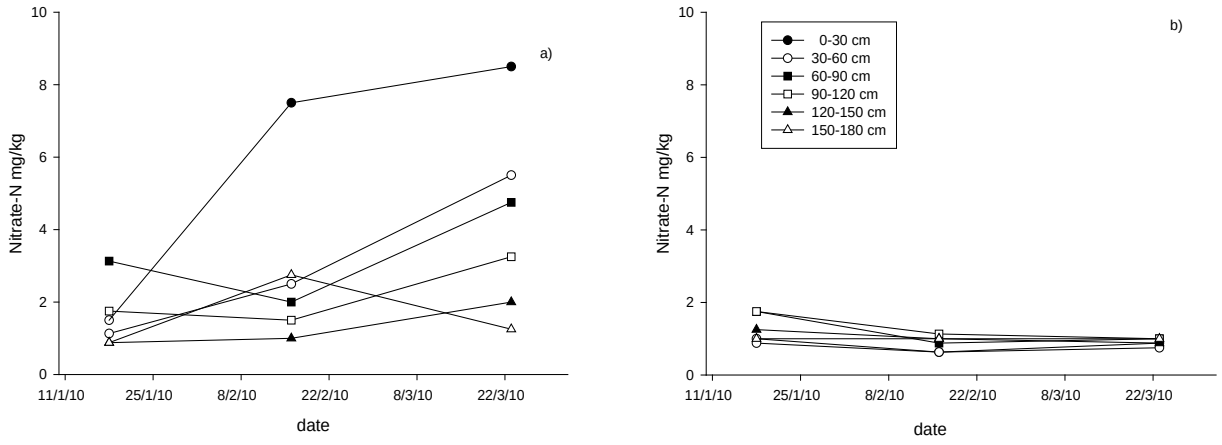
Experiment 1

Pre-trial analysis in the melon rooting zone
Pre-trial samples collected from the melon rooting zone (0-150 mm and 150-300 mm) had total carbon levels of 2.2%; standard error of mean (SE) = 0.11 and 1.4% (SE =.09) and C:N ratios of 19 (SE = 1.2) and 16 (SE = 2.2), respectively.

Pre and in-trial sampling to 1.8 m
Nitrate-N values for depth samples (0-1.8 m) averaged 1.03 mg/kg (SE = 0.164) across the four treatments in the 0-100 mm depth; values at depths below this ranged from 0.50-0.88 mg/kg. There were no treatment differences in December. January nitrate-N levels, averaged over all treatments, were 1.34 mg/kg (SE = 0.256) in the 0-300 mm depth, and were highest at 2.28 mg/kg (SE = 0.391) at 600-900 mm and 2.19 mg/kg (SE = 0.295) at 900-1200 mm.

From January through to the final sample in mid March, there was a trend for nitrate-N levels from 0-1.5 m depth (the top five sample depths) to increase in the fallow treatments and decrease in the sorghum and weed

treatments. Figure 1 shows this trend for the fallow treatment with no fertiliser applied and the forage sorghum treatments. Nitrate-N values were highest at 7.75 mg/kg (SE = 0.773) in the 0-300 mm depth for the average of the two fallow treatments, compared with a mean of 0.88 mg/kg (SE = 0.081) for the combined weed and sorghum treatments. At all depths, nitrate-N levels were almost five times higher in fallow treatments (25.1 mg/kg (SE = 2.39), total of all profiles) than in the sorghum and weed treatments (5.8 mg/kg (SE = 0.55)). This is evidence of mineralisation of N from organic fractions of the soil to nitrate-N and its subsequent use by plants for growth. Ammonium-N levels also increased during the trial, but comparatively less in sorghum and weeds than in the fallow (data not presented).



Each value is a mean of four samples.

Figure 1. Nitrate-N concentrations at six depths for January, February and March samples from (a) fallow plots with no fertiliser applied and (b) plots sown in forage sorghum

Experiment 2

Alternative cover crop species

Since birds damaged the alternative cover crop in the trial, only limited data could be collected. To ensure seed collection, however, Sunn hemp was netted to protect against bird damage. The biomass of Sunn hemp was equivalent to 8.7 t/ha high at early flowering 77 days after sowing (mean density 69 plants m^2) and continued to increase to 22.5 t/ha at 120 days after sowing (mean density 65 plants m^2). Even though Sunn hemp produced flowers, seed-set was low as measured at 120 days.

Leguminous cover crops are not usually suitable in rotation with melons because they are hosts to damaging nematode species, such as *Meloidogyne hapla* (root knot nematode). As Sunn hemp is not susceptible to nematodes such as root-knot (Wang et al. 2007), fixes N, does not produce seed quickly and produces high levels of biomass, the species may provide advantages over forage sorghum and millet for use in melon cover crop systems.

No biomass data was collected for the other species but it was observed that Siberian millet ran to seed quickly, with immature seed present in mid-February.

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PROJECT: Controlling Snake Bean Fusarium Wilt in the NT

Project Officers: B. Condé, M. Traynor, D. Cumberland, M. Hearnden, L. Tran-Nguyen, J. Tilbrook and L. Ulyatt

Location: Darwin

Keyword(s): diseases, snake beans, *Vigna unguiculata* ssp. *sesquipedalis*, Fusarium wilt, *Fusarium oxysporum* f.sp. *tracheiphilum*, disease management, grafting

Objectives:

To develop and promote insect pest management (IPM) controls for soil-borne diseases, specifically Fusarium wilt of snake beans in the Northern Territory (NT).

To characterise strains of snake bean Fusarium wilt by collecting isolates from the field and conducting race differential infection rates, vegetative compatibility groupings and molecular analyses.

Background:

This research was part of Horticulture Australia Limited grant VC01725 on the national “Best-practice IPM Strategies for the Control of Major Soil-borne Diseases of Vegetable Crops” throughout Australia (Vegetable Pathology Sub-program).

Fusarium wilt caused by *Fusarium oxysporum* f.sp. *tracheiphilum* (Fot) was first detected in snake beans (*Vigna unguiculata* s.sp. *sesquipedalis*) in the NT in mid-1999; it has now become a significant disease of snake beans (Condé and Arao-Arao 2001; Gosbee and Bui 2001). The importance of this disease to growers is demonstrated by a 55% reduction in snake bean production between 2005 (690 tonnes, valued at $3.1 million) and 2006 (300 tonnes, valued at $1.4million) due to an increase in the incidence of Fot in snake bean crops (Department of Resources).

Brown discoloration of the vascular tissue near the base of a plant is a characteristic symptom of the disease.

Figure 1. Symptoms of Fusarium wilt caused by the fungus *Fusarium oxysporum* f.sp. *tracheiphilum* in non-grafted snake beans

Previous work (Condé and Arao-Arao 2003; Condé and Arao-Arao 2005) demonstrated that there were at least three strains of Fot differentiated by culture colour on potato dextrose agar (PDA) acidified with two drops of 25% lactic acid (PDAL). Seventy four snake bean lines were screened and showed varying degrees of resistance to Fot. As snake beans and cowpeas (*Vigna unguiculata* s.sp. *unguiculata*) are closely related and both are affected by Fot, various cowpea lines were also screened for resistance. Previous studies overseas have also examined the reaction of cowpeas to Fusarium wilt (Armstrong and Armstrong 1980; Smith et al. 1999; Ehlers et al. 2009). Four cowpea types were found to be resistant to all three strains of Fot. Cowpea variety "Iron" was chosen as a rootstock for grafting snake beans because of its excellent resistance, strong root system and resistance to root knot nematode caused by *Meloidogyne incognita* and *M. javanica* (Conde et al. 2002, 2010). The research detailed in this report is a continuation of that earlier work.

Method:

In 2009, grafted vs. seeling trials were conducted on the commercial snake bean Kaohsiung Green Pod variety at three farms which had extremely high, moderate and low levels of Fusarium wilt. Site 1 (at Berry Springs) was planted on 11/05/2009 (high level of Fusarium wilt) in four alternate rows of 42 grafted and 42 seedling plants (total of 84 grafted and 84 seedling plants). Site 2 (at Acacia Hills) was planted on 04/05/2009 (medium level of Fusarium wilt) in four alternate rows of 75 grafted and 75 seedling plants (total of 150 grafted and 150 seedling plants). Site 3 (at Buckley Road, Humpty Doo) was planted on 12/05/2009 (low level of Fusarium wilt) in four alternate rows of 53 grafted and 53 seedling plants (total of 106 grafted and 150 seedling plants).

Plants at the three trial sites were assessed weekly for external and internal symptoms of Fusarium wilt. Samples were collected to confirm Fusarium wilt, to indicate the strains of Fot at the trial sites and for later characterisation of the strains. Isolations were made onto PDAL. Single spore (SS) isolates were obtained by streaking a suspension of spores on to water agar. The SS isolates were stored as filter paper cultures (Correll et al. 1986) at 4 ºC. Field days were arranged for farmers to demonstrate the effectiveness of grafting as a technique for controlling snake bean Fusarium wilt. A grafting workshop was held in 2010 prior to the 2010 on-farm grafting demonstration plantings. Field days will be held towards the end of the 2010 season together with a soil health workshop.

Data analysis

The differences in average days to infection and average days to death in plants were assessed to compare sites using Cox's proportional hazards models for censored data (Link 1984).

Results:

Plants grew well with few problems except for the Fusarium wilt, Cercospora leaf spot, caused by *Pseudocercospora cruenta* (Sacc.) Deighton and bean flies (*Ophiomyia phaseoli*). Bean flies had to be controlled at seedling emergence to prevent plant mortality. A fourth trial site was abandoned due to high levels of Pythium base rot caused by the fungus *Pythium myriotylum* Drechsler. Identification to species was done by Len Tesoriero and Leanne Forsyth, NSW Agriculture at the Agricultural Institute, Menangle NSW. About 22.9% of plants were affected by Pythium base rot in the 12 rows at the fourth site.

Figure 2. Early dying" of snake beans caused by the fungus *Fusarium oxysporum* f.sp. *tracheiphilum* seen here as two alternating rows of dead non-grafted plants and two healthy grafted plants at trial site 2

Differing rates of seedling infection and death over time (Figure 3) can be attributed to the different levels of Fusarium present in the soils across the three sites. The average number of days to infection for plants was significantly different between the sites (95% confidence limits for Site 1, 44 to 51 days < Site 2, 79 to 86 days < Site 3, 120 to 134 days). Similarly, the average number of days to plant mortality followed the same pattern (95% confidence limits for Site 1, 58 days < Site 2, 86 to 93 days < Site 3, 127 to 134 days). The seedlings at Site 1 showed signs of infection as early as 30 days after planting (DAP). Infection and death increased rapidly until all seedlings were dead by 78 DAP prior to any possible harvest. Seedling infection at Site 2 commenced fairly early (44 DAP) but was slower to develop compared with Site 1. No infected seedlings were recorded at Site 3 until 71 DAP and the rate of infection was slow compared with the other sites. The grafted plants remained uninfected at all three sites for the duration of the trials.

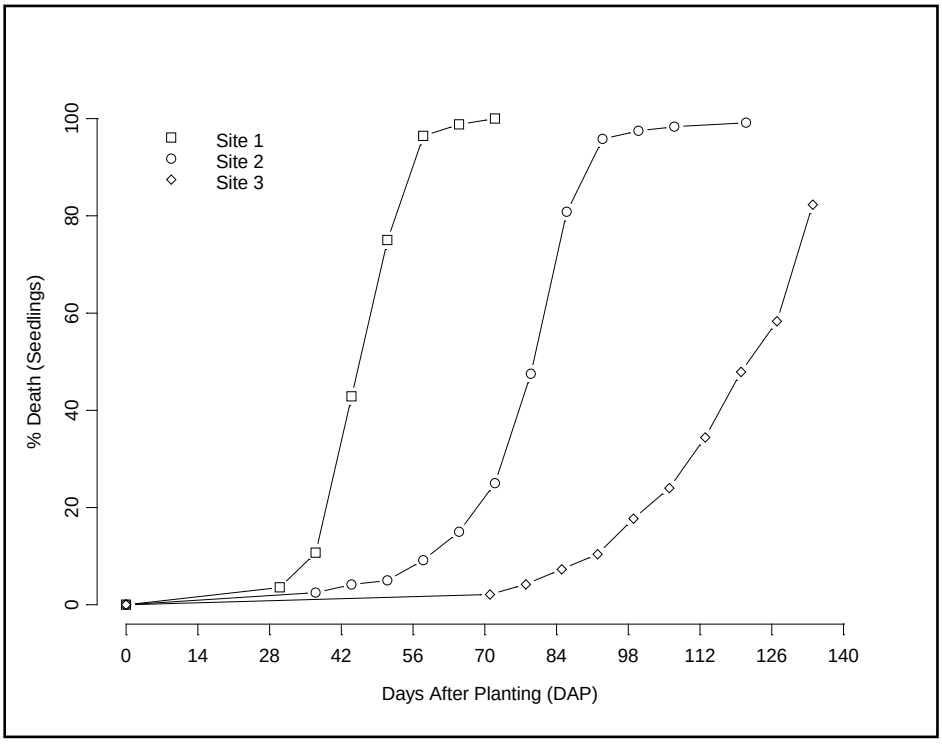

Figure 3. Cumulative death (%) of seedlings on days after planting for Sites 1 to 3

Yield data showed that the benefit of grafted plants depended on how quickly the seedling plants became infected and stopped production (Table 1; Figure 4). There was a 114% increase in the average comparative yield per plant due to grafting at Site 2. In contrast, there was only a 15% increase due to grafting at Site 3, due to a delayed and reduced infection rate. Plant yields over time (Figure 4) for both sites show that seedling and grafted yields are comparable until such point when seedling yields become increasingly affected by mortality from Fot, at which point yield drops significantly and only grafted plants remain.

Table 1. Average seedling yield per plant, average grafted yield per plant and increase in yield in grafted plants at Sites 2 and 3

	Seedling yield/plant (kg)	**Grafted yield/plant (kg)**	**Increase in yield (%)**
Site 2	2.1	4.5	114
Site 3	3.3	3.8	15

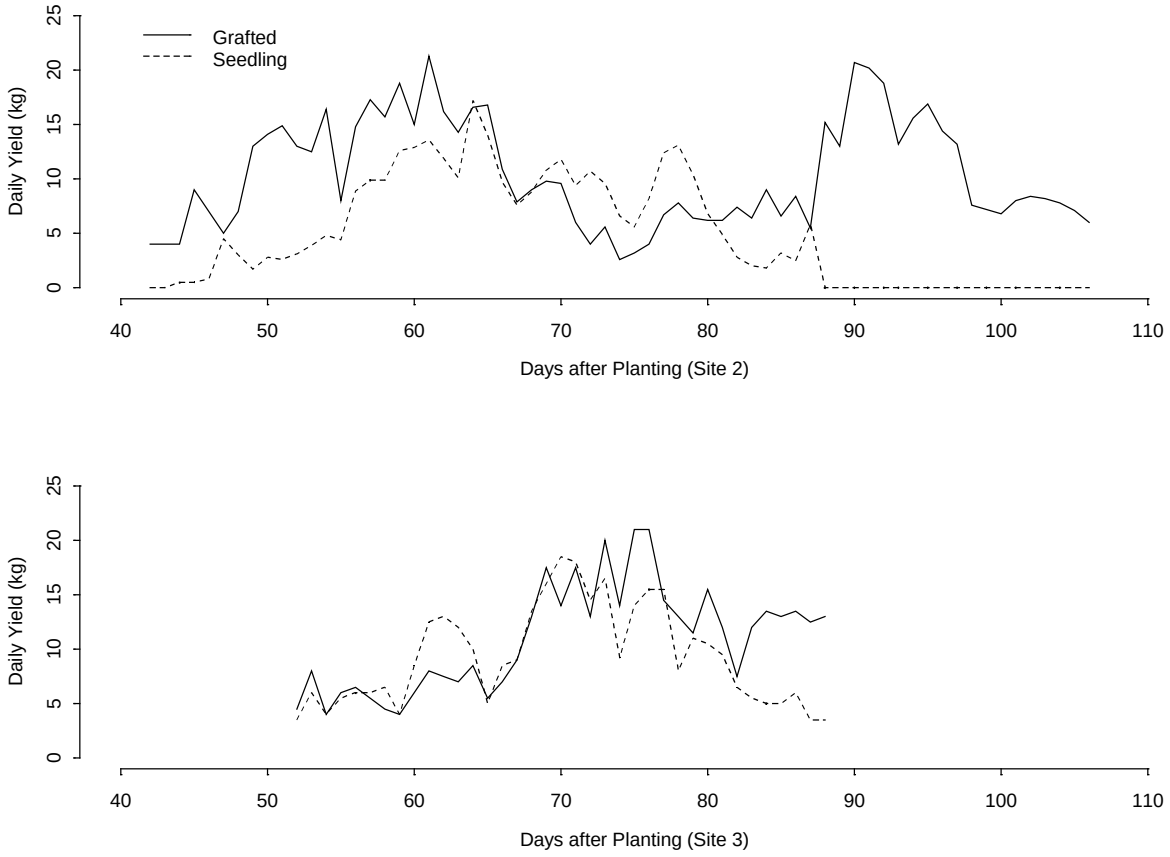

Figure 4. Yield comparison (kg) over time (DAP) for seedlings and grafted plants at Sites 2 and 3

During 2009,127 Fot isolates were collected from on-farm trial sites and other farms. The majority of isolates collected since 1999 were classified as the “Pink strain”, with fewer numbers of the “White strain” and “Plum strain”. During a field day held in conjunction with the on-farm field trials on 15 July 2009 on Site 1 and another on 5 August 2009 at Site 2, the benefits of grafting were clearly demonstrated. A grafting workshop was held on 14 May, 2010 at Berrimah Farm. Grafted and seedling plants of the commercial Kaohsiung Green Pod variety were given to farmers to plant for the 2010 grafted vs. seedling demonstrations.

A total of 38 Fot isolates comprised of 22 isolates from the 2009 field trials together with 16 isolates collected and stored as filter paper cultures prior to 2009 were grown as fungal lawns on PDA. DNA was extracted from these lawns for molecular characterisation. Fot Races 1 and 2 were imported as cultures. DNA from Races 3 and 4 were imported from the University of California, Riverside and DNA from two single spore Fot isolates was imported from Taiwan for comparison with the NT Fot isolates. Work on the molecular and race characterisation is continuing.

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Acknowledgement:

We acknowledge the cooperation and assistance of the three farmers involved in the three trial sites.

PROJECT: Evaluation of Rice Cultivars Grown under Aerobic Conditions in the Wet Season at Katherine Research Station

Project Officer: D. Hussie

Location: Katherine

Keyword(s): aerobic rice, rice cultivars, yield evaluation

Objectives:

To observe how various rice cultivars performed in a semi-arid tropical environment when grown during the wet season with supplementary irrigation.

To assess yield potential and gain a basic understanding of the agronomic principles associated with growing aerobic rice.

Background:

About 79 million hectares of irrigated lowlands provide 75% of the total rice produced globally (Bouman et al. 2007). Lowland rice is traditionally grown in bunded fields (paddies) that are continuously flooded from crop establishment until crop maturity. It is estimated that rice grown in this manner receives some 34% to 43% of the

world's total irrigation water, or 24% to 30% of the world's total freshwater withdrawals (Bouman et al. 2007). However, the declining availability and increasing cost of water threaten traditional methods of irrigated rice production. A fundamentally different approach is to grow rice like an upland crop, in an irrigated aerobic system, which can potentially lead to water savings and increased water use efficiency. Rice grown in non-flooded and non-saturated soil with supplemental irrigation is suitable for water-scarce environments, yet it can withstand periodic flooding, making it a suitable crop for the semi-arid tropics (Yang et al. 2005).

Rice research in the Northern Territory (NT) was only conducted in anaerobic conditions. That work first began in the mid 1950s simultaneously at the Coastal Plains Research Station by CSIRO and at the Upper Adelaide River Experimental Station by the NT Government. Research at those facilities focused primarily on cultivar assessment, establishment techniques, soil chemistry, fertiliser use, and weed and insect pests and diseases. As the research was conducted in flooded paddy systems (anaerobic) various limitations to productivity were encountered, such as uncontrolled flooding, rising salinity, difficulty of personnel movement, unsuitable cultivars, lack of suitable rotation crops and pest incursions. Consequently, the work was discontinued in the mid 1980s due to low economic viability.

Method:

In this assessment, 10 cultivars were from SunRice, 20 from the Biloela Genetic Resource Centre and 26 from the International Rice Research Institute (IRRI).

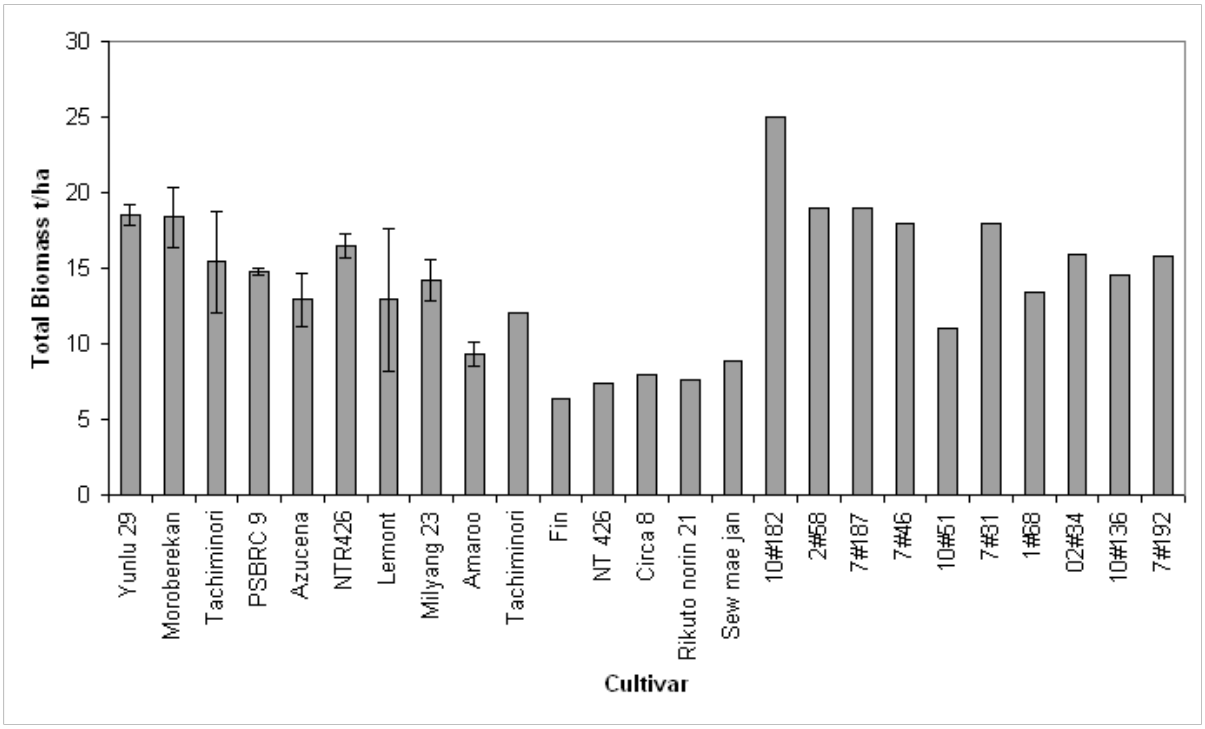
SunRice provided sufficient seed for a replicated experiment (randomised complete block with three replicates of ten cultivars). Plot size was 6 rows by 7.1 m long with row spacing of 0.18 m. Since Biloela and IRRI supplied only a small amount of seed, it was possible to make only un-replicated observation plots for that seed. Depending on seed quantity, plot size varied between one and two rows. All rows were 7.1 m long.

Crop establishment data, date of flowering, nutritional status, crop biomass and grain yield were recorded. Biomass and yield are reported here. Total biomass at harvest was determined by cutting all plants at ground level along 1.3 m of a single row. Samples were dried at 60 °C for three days and dry weights were recorded at 0% moisture. To determine grain yields, one quadrate per plot was harvested (one row by 1.3 m long) by cutting all plants at ground level. Harvesting of the cultivars commenced on 30 March, 2010 and was completed on 23 April, 2010. The plants were then dried at 50 °C for 24 hours, threshed and the dry weights of the grain were recorded. Grain yields are expressed at 14% moisture.

Results:

The aim of this trial was to assess several rice cultivars to see how they performed and to estimate their yield potential. The cultivars from SunRice, Biloela and IRRI were not all specifically suitable for a semi-arid tropical environment since some of the lines did not perform well. Results are therefore presented only for ten SunRice cultivars, six Biloela cultivars that recorded values for biomass and ten of the best performing IRRI cultivars, based on their grain yield.

Total biomass at harvest varied across all cultivars (Figure 1), with a mean of 14.1 t/ha. The IRRI cultivar 10#182 recorded the highest total biomass of 25 t/ha and also produced the highest grain yield of more than 5.8 t/ha. Of the SunRice cultivars, Yunlu 29 and Moroberekan produced the highest total biomass of 18.5 and 18.4 t/ha, respectively. Doongara was the only Sunrice cultivar that failed to produce any biomass. The Biloela cultivars performed poorly compared with SunRice and IRRI cultivars, with a total biomass average of 8.3 t/ha. Cultivar Tachiminori, however, produced both a higher biomass (12 t/ha) and harvestable grain yield.

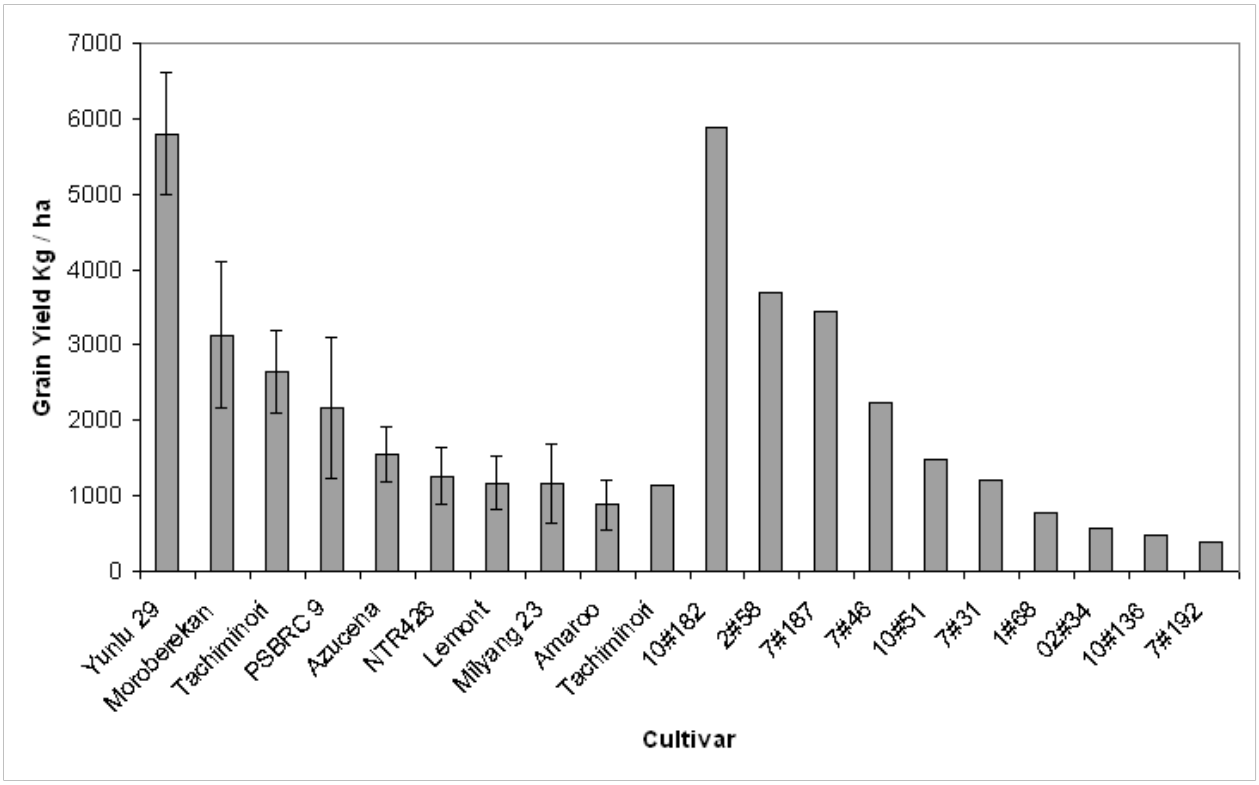


95% confidence intervals are presented for the mean biomass of SunRice cultivars (replicated treatments).

Figure 1. Total biomass (t/ha at 0% moisture) at harvest in 25 of the cultivars assessed

Only 20 of the 26 cultivars assessed for biomass produced harvestable grain yield (Figure 2). The median grain yield of the 20 cultivars was 1370 kg/ha, while the average was higher (2050 kg/ha), affected by the high grain yield of Yunlu 29 and 10#182. The highest mean grain yield recorded for a SunRice cultivar was for Yunlu 29, at 5799 kg/ha; one of the replicates of this cultivar recorded a grain yield of 7017.3 kg/ha. Tachiminori was the only cultivar in the Biloela observation plots that produced a grain yield of 1138.8 kg/ha; this was lower than that observed in the replicated plots for this cultivar (2641.5 kg/ha).

The lowest yielding IRRI cultivar was 7#192, which produced 398.1 kg/ha, while Amaroo recorded the lowest grain yield in SunRice cultivars (880 kg/ha). The other six cultivars were harvested only for biomass due to very low grain yields.



95% confidence intervals are presented for the mean grain yields of SunRice cultivars (replicated treatments).

Figure 2. Grain yield (kg/ha at 14% moisture) in 20 of the cultivars assessed

A number of obstacles were encountered that may have affected final grain yields, including high locust pressure throughout the growing season, reduced solar radiation (a characteristic of the wet season) and an observed lack of root proliferation. Perhaps in a more favourable season, rice yield may have been higher in some or all of the varieties.

As a result of this trial, a number of key areas have been identified for further investigation, including herbicide management, fertiliser management, time of sowing, machinery options and baseline data for rice diseases and pests.

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PROJECT: Boosting Rambutan Production by Improving Fruit-set

Project Officers: M. Traynor and J. Drinnan (DEEDI)

Location: Darwin

Keyword(s): rambutan, flowering, fruit-set, synthetic auxin NAA

Objectives:

To determine the ideal rates and timing of application of the synthetic auxin NAA to promote the production of male flowers to improve pollination, fruit-set and yields.

To use the efficacy and residue data collected in this project to apply to register NAA for use in rambutan production.

Background:

Rambutan (*Nephelium lappaceum*) is native to the humid, tropical regions of Malaysia and Indonesia. In those areas, where the climate is reasonably predictable, production is also reasonably reliable. Poor fruit-set and low yield have been a problem in Australian rambutan orchards, especially in seasons when flowering is early. Observations have indicated that very few male flowers are produced in these early flowering panicles, which leads to poor pollination due to the absence of pollen. Similar observations have also been made in Hawaii and parts of Thailand (Kawabata et al. 2004; Salakpetch 2000). Rambutan trees are known to naturally have a very low percentage of male flowers, often below 1% and sometimes below 0.05% (Valmayor et al. 1970). When rambutan trees are cultivated in areas outside their natural environment, with cooler than ideal temperatures, this problem is further exacerbated. When pollination is poor, fruit-set is poor, which can lead to the development of deformed parthenocarpic fruit, which fails to develop properly, reducing yields. Rambutan pollen is known to have good viability, remaining viable for a long time and the stigma on female flowers remains receptive for up to 48 hours, so when pollen is available, pollination is usually successful.

Rambutan trees produce three types of flowers – male, hermaphrodite female and hermaphrodite male. Pollen is produced by the male and hermaphrodite male flowers and fruit is only set in hermaphrodite female flowers.

Naphthalene acetic acid (NAA) has been used in Thailand and Hawaii to increase male flower development to aid pollination (Kawabata et al. 2004; Salakpetch 2000). The application of NAA to developing panicles during early anthesis induced the development of hermaphrodite male flowers. Experiments in Hawaii (Kawabata et al. 2004), Thailand (Salakpetch 2000) and preliminary trial work here in Australia (Diczbalis and Drinnan 2007) have shown that NAA applied at 40-90 ppm as a foliar spray is able to stimulate the development of male flowers. With the increase in available pollen, it is assumed pollination should be better and yields should increase; however, yield benefits from these treatments have not been determined.

Method:

During 2009, research trials were conducted at rambutan orchards in the Northern Territory around Darwin (12°S). The commercially-grown varieties used were R134, Binjai and Jitlee. In all the trials, early flowering trees (mid to late June) were identified, marked and divided into two groups – those to be treated with NAA and those left as untreated (control). All trees within individual trials were of equal size and had uniform amounts of flowering. The two groups of trees were separated within the orchard as much as possible to reduce the chance of pollen moving from treated to untreated trees by wind or insects. The trees were mostly separated by at least 50 m and/or by several rows of trees. Four to five panicles were tagged on treated trees and sprayed with 40 ppm (2 mL/L) NAA

plus 0.1% Kendral 600 wetter (manufactured by Kendon Chemicals Pty Ltd). The orchards were protected from birds and bats with either permanent or temporary netting. All orchards were irrigated, fertilised and the trees were kept free of pests and diseases, in a healthy productive state.

The rate of 40 ppm was chosen after referring to preliminary work conducted by Diczbalis and Drinnan (2007) in Australia, Salakpetch (2000) in Thailand and Kawabata et al. (2004) in Hawaii. In these studies, it was concluded that rates above 90 ppm can damage (burn) the flowers and rates below 20 ppm were less effective. By using the low rate of 40 ppm it was hoped that the flowers which did not respond to NAA on the first application, would not be damaged and could be used in subsequent applications.

Panicles were sprayed with NAA using a hand sprayer until runoff. Spraying occurred late in the afternoon to reduce the chance of evaporative losses and improve the absorption of NAA. Panicles were first sprayed at the start of anthesis or when the pistil could be seen protruding from the closed florets (Figure 1). After seven days, the same panicles were sprayed again if there were still unopened flowers at the correct stage of development. If there were no flowers left to open, new panicles were tagged and sprayed in subsequent applications. This continued until all the flowering on the trees had finished – usually three to four applications (three to four weeks). In deciding the number of panicles to treat per tree, about one in 10-15 panicles or one per 2 m² of the flowering canopy was treated. Panicles were chosen in the upper part of the canopy and evenly spaced amongst the other non-treated panicles to aid pollination by wind or insects. Flowering and fruit-set were observed in the trees at weekly intervals and yield data was collected from individual trees. A sample of fruit was collected from some of the trials for fruit quality assessment (size, weight and seed development) and residue testing (Agrisearch Analytical Pty Ltd).

Figure 1. The ideal stage to apply NAA is when flowers have just started to open or when the pistil is seen just protruding from the closed florets (inset)

Results:

Flowering and fruit-set

In all trials and on all varieties studied, NAA applications stimulated the production of male and hermaphrodite male flowers on treated panicles within seven to 10 days of application (Figure 2). Closed florets with the pistil protruding developed into hermaphrodite male flowers. Flowers slightly less developed opened as male flowers. The flowers already open at the time of application remained female and were generally damaged or burnt by NAA sprays. Observations of the flowers throughout the orchard on panicles not sprayed with NAA indicated that there were either no male flowers present up until September, or the numbers were so small that they were not detected.

In all the trees treated with NAA, a significant increase in the number of fruit-set within the first few weeks of male flowers being present was evident, especially on the panicles in close proximity to treated panicles. Presumably this was due to better pollination. In all cases this led to a significant increase in yield. No fruit- set was observed on treated panicles. This indicates that when using NAA, choosing the correct number of panicles to treat will be a compromise between getting a good number of male flowers and hence pollen supply evenly distributed throughout the trees and orchard to ensure the highest likelihood of pollination, and not treating too many panicles that will result in reduced yields due to the limited number of panicles that are able to crop. Results from these trials suggest that treating one in every 10-15 panicles or one panicle per 2 m² of the flowering canopy is a good starting point.

The location of treated panicles within the trees or the orchard is also a key consideration. Ideally, they should be chosen amongst other panicles and higher in the canopy as they will be more exposed to wind and insects for disseminating the pollen. From higher in the canopy pollen can also fall through the tree onto other panicles.

Figure 2. Male flowers are seen on panicles 7-10 days after spraying with NAA

Yield

The results from the trials are shown in Tables 1 and 2. In all four trials, yields significantly increased in treated trees compared with control trees, with the treated trees recording up to ten times (15 kg/tree) the yield of control trees. These significant yield increases again demonstrate the dramatic effect of low numbers of male flowers (pollen supply) on limiting the potential yield capacity of early-flowering rambutan trees growing in regions outside their natural environment.

Table 1. The effect of NAA panicle sprays on tree yields at site 1, Berry Springs

Variety	Control	NAA
	Yield (kg/tree)	
Binjai	2.6	5.1

The trees were five years old, measuring 2 m wide × 3 m high. Data is the average of five trees. Yield between the two groups was significantly different (*P*=0.05).

Table 2. The effect of NAA panicle sprays on tree yields at site 2, Humpty Doo

Variety	Control	NAA
	Yield (kg/tree)	
Binjai	6.9	17.0
Jitlee	1.8	17.6
R134	1.7	14.3

The trees were five years old, measuring 2 m wide × 3 m high. Data is the average of 7-8 trees. In each variety, the yield in treated trees was significantly higher than in control trees (*P*=0.05).

The response in the different varieties studied in these and previous trials indicates that NAA is likely to have a significant positive effect on improving yields in all varieties. However, significant differences in the size of the response were observed between the varieties studied. The yield increase in the variety Binjai was very consistent at around 2-2½ times the yield in control trees. For R134 and Jitlee, the increase in yield was much larger at around eight and ten times, respectively. This indicates that the use of NAA for improving pollination may be even more critical in varieties R134 and Jitlee, compared with variety Binjai.

Nagao (2004) and Kawabata et al. (2004) both found that some varieties produced a lot of male flowers and others much fewer when treated with NAA. They found that Jitlee produced the largest number of male flowers followed by R134 and then Binjai. Diczbalis and Drinnan (2007) also found variety R134 very responsive to NAA, producing hundreds of male flowers per panicle. The responsiveness of these varieties to NAA reported by these researchers is similar to the results of this study.

Fruit maturity, quality and residues

Flowering at both trial sites commenced during the last week of June and harvesting commenced at the end of October (four months).The pattern of fruit ripening across all the trials indicated that fruit generally matured at about the same time in the NAA-treated trees as in control trees.

Fruit quality assessments indicated that there was no significant difference in size, weight or seed development between fruit from NAA-treated trees and control trees. Comparative fruit weights are shown in Table 3.

Table 3. The effect of NAA panicle sprays on fruit weight in trials at site 2, Humpty Doo

Variety	Control (g)	NAA (g)
Binjai	32.8	31.6
Jitlee	26.4	27.6
R134	33.6	34.0

The data is the average of 50 fruits.

Residue analysis of fruit collected from treated and control trees did not detect any NAA residues (Table 4). This was expected as NAA was only applied to a very small proportion of the tree and the treated panicles did not set any fruit.

Table 4. NAA residues in fruit from treated and untreated trees

Sample	Residue in skin and flesh
NAA treatment, three weeks before harvest	< LOD
NAA treatment at harvest	< LOD
Control	< LOD

LOD = Level of detection: 0.02 mg/kg (Agrisearch Analytical Pty Ltd).

Recommendations:

The project has developed a management strategy to overcome a major constraint on production in local rambutan orchards. The use of NAA to ensure the production of male flowers has the potential to markedly improve the productivity of rambutan trees, especially in years when seasonal and climatic conditions are conducive for early flowering. Given that the number of male flowers produced is always very low in rambutan trees, it is likely that using NAA during flowering at any time of the year will be useful in improving pollination, fruit-set and yield.

NAA treatments did not affect the pattern of maturity or fruit quality and no residues were detected. An application for registration has been submitted to APVMA. Once NAA is registered, it is recommended that growers spot-spray evenly around trees and the orchard one in 15, or 1 per 2 m² of flowering panicles just prior to flower opening, with 40 ppm NAA at seven to10-day intervals during the flowering period. The increase in early production by using NAA would allow growers to capitalise on high, early prices, thereby raising income, which would exceed by far the estimated cost of applying NAA ($100/ha).

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Acknowledgment:

We acknowledge the assistance and cooperation of participating rambutan growers in these on-farm trials; we also thank the Rural Industries Research and Development Corporation for funding this research.

PROJECT: The Influence of Pests and Diseases on Rambutan Fruit Quality in the NT

Project Officers: D. Chin, R. Meldrum, A. Daly, B. Conde and B. Thistleton

Location:	Darwin
Keyword(s):	caterpillars, Coccidae, Coccinellidae, Coccus, *Cryptolaemus montrouzeri*, diseases, false-spider mites, *Ferrisia virgata*, fluted scales, *Icerya aegyptiaca*, *Icerya seychellarum*, insects, *Lasiodiplodia theobromae*, Margarodidae, mealybug ladybird, mealybugs, pathogens, *Pestalotiopsis*, planthoppers, Pseudococcidae, Pulvinaria, Pyralidae, rambutan, red-banded thrips, soft scales, Tenuilpalpidae

Objective:

To determine the effect of pests and diseases on rambutan fruit quality in the Northern Territory (NT).

Background:

The production of rambutan in Australia is restricted to the tropical areas of north Queensland and the Darwin region in the NT. Due to bird, fruit bat and other vertebrate pests, commercial rambutan crops are grown either in permanently-netted orchards or in seasonally-netted orchards, during the fruiting period. The harvest period in the Darwin region is from December through to February, which complements that of north Queensland, which happens from February to May.

Lim and Diczbalis (1998) described the attributes of a 'good' commercially acceptable rambutan variety as having fruit weight of over 40 g (with flesh recovery ratio over 50 %), yellow coloured skin, with fewer than 5.5 spinterns/cm² on the surface and a spintern length of under 12 mm to reduce moisture loss. About 15 commercial rambutan varieties are grown in Australia, of which Jitlee, R156, R167 and R134 are the most popular (McMahon 2003).

Earlier work by the Department of Resources in 2002-03 as part of a Rural Industries Research and Development Corporation project with Queensland involved integrated pest management and monitoring of rambutan orchards in the NT and Queensland. The results were based mainly on pre-harvest sampling and monitoring (Chin et al. 2003; Astridge 2006). A brief assessment of diseases of rambutan was documented by Lim and Diczbalis (2005).

Method:

The trial was conducted between December 2009 and January 2010, using three commercial varieties (Jitlee, Binjai and R134). Fruit was supplied by four properties within 60 km south of Darwin. Each of the four growers supplied 200 fruits. Of those, 100 were unwashed and 100 were washed, using each grower's standard conventional practice. A total of 800 fruits were examined. Each batch of fruit was freshly-harvested and delivered to the Entomology laboratory at Berrimah Farm on the day of harvest. The fruits were initially scored on the day of harvest for insects and mites (or their damage) as well as for pathogens and were then stored in an air-conditioned laboratory at 23 °C and 50-60% relative humidity for up to eight days for further observation.

Fruit quality

Fruits were recorded for individual weight and length; then they were allocated to three classes: class 1, class 2 and reject. Other disorders that were recorded included immature/green, flat fruit, over-ripe, black spinterns, desiccated spinterns and skin abrasion. These characteristics were selected from the Rambutan Quality Standards developed by Lim et al. (1996).

Insects and mites

Each fruit was examined under a stereo microscope for pest damage and individual numbers of insects and mites were recorded. All insects and mites were identified to family and/or species.

Microorganisms associated with fruit rot

Fruit was assessed visually for the presence of diseased tissue, which was generally represented by topical necrotic lesions. Potential pathogens were isolated from suspect fruit lesions by surface-sterilising the fruit material in 70% ethanol, rinsing in sterile distilled water, then plating onto potato dextrose agar amended with lactic acid. Growth was observed and the causal pathogen was identified by cultural characteristics.

Results:

Fruit quality

Table 1. Fruit quality assessment of unwashed fruit - proportion of affected fruit per property (%)

Grower property	Mean fruit weight (g)	Mean fruit length (mm)	Class 1	Class 2	Reject	Immature	Sunburn	Flat fruit	Over-ripe	Black spinterns	Desiccated spinterns	Skin abrasion
A	40.38	24.50	97.0	3.0	0	0	0	0	0	0	5	0
B	24.58	40.9	18	63.0	19.0	31.0	0	0	0	0	15.0	0
C	29.82	44.9	33.0	54.0	13.0	0	0	0	0	0	0	0
D	34.01	45.2	81.0	19.0	0	0	0	0	9.0	2.0	0	0

Table 2. Fruit quality assessment of washed fruit - proportion of affected fruit per property (%)

Grower property	Mean fruit weight (g)	Mean fruit length (mm)	Class 1	Class 2	Reject	Immature	Sunburn	Flat fruit	Over-ripe	Black spinterns	Desiccated spinterns	Skin abrasion
A	31.99	46.3	100	0	0	0	0	0	0	0	49.0	10.0
B	16.90	36.6	11.0	89.0	0	25.0	0	0	0	0	0	0
C	33.8	48.0	36.0	59.0	5.0	0	0	0	0	0	0	0
D	37.97	47.0	81.0	19.0	0	0	0	0	6.0	10.0	0	0

There was a large variation in the standard of fruit from all four properties, reflecting the quality of fruit at the time of the trial. The fruit varieties that were supplied by each property were: A: R134 (200 fruits), B: Jitlee (200 fruits), C: Binjai (100 fruits), Jitlee (100 fruits) and D: Jitlee (200 fruits). Only property A was assessed to have the ideal mean fruit weight of 40 g or over. That property was also the only one with a high percentage of class 1 fruit (Tables 1 and 2) (97% in unwashed and 100% in washed) followed by property D (81% in unwashed and 81% in washed) then property C (33% in unwashed and 36% in washed) and then B with the lowest (18% in unwashed and 11% in washed). Property D was the only property that did not use brushes for cleaning the fruit, only using jets of water. Observations under the microscope indicated that fruit from that property had fewer skin abrasions. It was also noted that cleaning fruit with brushes and/or water had a tendency to enhance the appearance of insect or mite damage on the skin, especially sucking and rasping damage, such as that caused by mites. That damage was not noticeable until a few days after harvest. Properties A, B and C all used brushes either on the production line or manually cleaned the fruit with small hand-held brushes (such as tooth brushes). Although property A had a high percentage of Class 1 fruit, a high percentage of fruit had desiccated spinterns (49%), which were largely caused by the meticulous cleaning process carried out with small hand held brushes by packers.

Insects and mites

Table 3. Insects and mites recorded on unwashed fruit - proportion of fruit affected per property (%)

Grower property	Caterpillars	Red-banded thrips	False-spider mites	Soft scales	Mealybugs	Fluted scales	Planthoppers	Mealybug ladybird
A	18.0	0	9.0	31.0	33.0	8.0	4.0	1.0
B	6.0	0	7.0	91.0	4.0	0	0	52.0
C	13.0	1.0	7.0	1.0	17.0	3.0	0	3.0
D	4.0	1.0	4.0	15.0	15.0	1.0	2.0	7.0

Table 4. Insects and mites recorded on washed fruit - proportion of fruit affected per property (%)

Grower property	Caterpillars	Red-banded thrips	False-spider mites	Soft scales	Mealybugs	Fluted scales	Plant hoppers	Mealybug ladybird
A	2.0	0	0	1.0	12.0	0	0	0
B	2.0	0	0	18.0	19.0	2.0	0	2.0
C	1.0	0	0	3.0	1.0	0	0	0
D	1.0	1.0	2.0	10.0	16.0	12.0	1.0	2.0

The main insects and mites recorded on harvested fruit are shown in Tables 3 and 4. The insect and mite pests recorded in higher densities on fruit included soft scales (various species in the family Coccidae), including *Pulvinaria* sp., *Coccus* sp.), mealybugs (Pseudococcidae) including *Ferrisia virgata* Cockerell, caterpillars (various species in the family Pyralidae), fluted scales, (Margarodidae), *Icerya aegyptiaca* (Douglas) and *Icerya seychellarum* (Westwood) and mites (family Tenuilpalpidae). The insects and mites were either attached to the fruit surface, or were protected by the spinterns (including crevices between spinterns), or were protected within the fruit or frass (e.g. caterpillars).

Various other more mobile invertebrates become disturbed and move or fall off the fruit more easily during harvest and they are found in lower numbers, such as plant hoppers. Red-banded thrips were also found in low numbers; although the harvesting process may disturb them on the fruit, they are generally in low numbers during the harvest period, which is December to February.

Mealybug ladybirds (*Cryptolaemus montrouzeri)* (Mulsant) (Coccinellidae) were recorded on all properties and are the dominant predator (occurring naturally) feeding on mealybugs, fluted scales and soft scales.

Microorganisms associated with fruit rot

Table 5. Microorganisms recovery from unwashed fruit (% of fruit affected)

Trial site	*Pestalotiopsis* sp.	*Lasiodiplodia theobromae*	Bacteria
A	4	0	0
B	13	7	2
C	1	1	1
D	2	0	0
Mean	5	2	1

Table 6. Microorganism recovery from washed fruit (% of fruit affected)

Trial site	*Pestalotiopsis* sp.	*Lasiodiplodia theobromae*	Bacteria
A	2	0	3
B	1	1	1
C	6	0	0
D	12	9	0
Mean	5	3	1

Table 7. Microorganism recovery from total fruit (% of fruit affected)

	Pestalotiopsis sp.	*Lasiodiplodia theobromae*	Bacteria
Mean	5	2	1

Two main fungal organisms were identified from the rambutan fruit sampled: *Pestalotiopsis* sp. and *Lasiodiplodia theobromae*. Both of these pathogens are associated with post-harvest diseases in rambutan fruit. *Pestalotiopsis,* which was found in 5% of the fruit sampled, can cause rambutan fruit rot (Coates et al. 2003). *L. theobromae,* which was found in 2% of the fruit, can cause stem-end rot in rambutan fruit (Coates et al. 2003).

While bacteria were isolated from the fruit lesions, less than 1% of the sampled fruit were affected; the presence of bacteria was associated with insect wounds. In this study, there was no significant difference in the presence of pathogens between the washed and the unwashed fruit (Tables 5 to 7).

Possible future research could determine the degree to which insects facilitate post-harvest fungal and possibly bacterial infections, and investigate the role of integrated pest management as a means to control pathogens.

Further studies

Proposed studies for 2010 involve a larger fruit sample size, with research restricted to one or two commercial varieties. Due to a large variation in the results, due possibly to selective harvesting by the grower, it is suggested that the fruit used in future trials should be harvested by Departmental staff using statistically valid techniques.

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PROJECT: Comparative Performance of Date-palm Scale on Different Date Cultivars in Central Australia

Project Officers: S. Raghu, J. Robson, G. Walter, G. Oliver and M. Hidalgo

Location:	Alice Springs
Keyword(s):	*Parlatoria blanchardii*, date-palm, scale, biological control, integrated pest management, risk analysis, nutrients, irrigation rates

Objectives:

To diagnose pest species and any parasitoid and predator species present according to the most up-to-date taxonomy/systematics.

To evaluate pest pressure imposed by scale insects across the different date-palm cultivars related to fertiliser and irrigation use.

To investigate the parasitism and predation rate of date-palm scale insects across the different date-palm cultivars and localities.

Background:

Date-palms have a significant potential in Central Australia. In the early 1990s, the Northern Territory (NT) Government invested significantly in acquiring a date-palm collection from major date-producing regions and germplasm collections. A trial was established in Alice Springs with over a dozen cultivars. This vital resource has helped establish a fledgling date-palm industry in Central Australia. A significant obstacle in the transformation of this niche industry into mainstream plant production is the deleterious date-palm scale insect (*Parlatoria blanchardii*). Its feeding activity reduces the productivity of date palms, while its control with synthetic organic pesticides is costly. The presence of this pest also imposes significant quarantine barriers to the movement of productive cultivars and the transfer of new germplasm acquisitions from nurseries in date-palm scale-infested areas to farms.

Scale insect pests can be effectively managed through the ecological manipulation of parasitoid wasps (highly host-specific) and arthropod predators that feed on them and kill them. The development of a biological control

program for the date-palm scale would benefit growers significantly by increasing access to germ-plasm collections, reducing production costs associated with controlling scale insects with chemicals, developing environmentally safe and sustainable management practices, and improving market-access to locally-produced dates nationally and internationally.

Method:

Sampling program

A structured sampling program has been designed to document the abundance of scales in relation to (1) the different date palm cultivars (Barhee, Mehdjool, Deglet Noor, Fard, Zahidi), (2) timing of the major infestation periods (January to September) (3) fertiliser application (two levels) and (4) irrigation rate (two rates). Scale insects are being collected on infested plant parts. Ten randomly selected leaflets are harvested at each sampling time from each palm and counts of dead and living scales are made to quantify relative infestation rates in relation to the above factors. Sampling was completed in February, April and June, with two more sampling events in August and October. Arthropod predators and parasitoids that are present will be removed, counted and stored for identification.

The identification of scale insects, parasitoids and predators

All insects will be submitted to specialists for identification because reliable keys to Australian insects in these groups are not available and their correct identification depends on the interpretation of subtle morphological characters (scale insects: USA; predators: Australia or the UK; and parasitoids: Germany, South Africa or the UK, depending on which genus they represent). At least 28 parasitoid and predator species are known to be associated with date palms globally. We will determine which of these, if any, are present in Australia and what local species attack them. This information, together with the sample data, will provide the basis for developing an approach to pest management suitable for Central Australian conditions.

The data will be analysed using mixed-effects models with the cultivar, fertiliser treatment and irrigation rates as fixed effects and time as a random effect. Response variables will include abundance of scales and rates of natural enemy attack.

Results:

This project commenced in January 2010 and will be completed in December 2010. A preliminary analysis of the data collected to date indicates that cultivars Barhee and Mehdjool support a higher population of live scales than Deglet Noor, with Fard and Zahidi supporting an intermediate abundance of scales. Whether this trend holds up across multiple sampling periods will be determined once all the data is collected and analysed. Natural enemies collected thus far have included predatory mites and beetles, and parasitoid wasps. These will be accurately diagnosed to assess predation/parasitism pressure relative to cultivars and experimental treatments.

Acknowledgement:

This work is being funded through an NT Research and Innovation Board grant and is part of the research project of Jonathan Robson (University of Queensland).

PROJECT: Challenges and Opportunities for Commercial-scale Bush Tomato Production

Project Officers: G. Ellis, G. Oliver and S. Raghu

Location:	Alice Springs
Keyword(s):	*Solanum centrale*, irrigation rates, bush foods, weed management, Indigenous economic development

Objective:

To assess the potential for commercial scale production of varieties of Solanum centrale (bush tomatoes) in relation to irrigation rates and weed competition.

Background:

The wild harvest of bush tomatoes in 2000-01 was estimated to be 800 to1000 kg, fetching between $15 to $20/kg (Robins and Ryder 2004). The ongoing drought reduced wild harvest supplies, resulting in an increase in the price of bush tomatoes to $35-$50/kg in 2007-08. The total volume of fruit from both wild harvest and cultivated supply in this period was estimated to be ~600 kg (Juleigh Robins, Director – Robins Foods pers. comm.). An increase in awareness of bush tomatoes has elevated their demand and price. Collectively, these factors have resulted in increased interest by various groups to establish commercial-scale production of bush tomatoes and other bush foods (Miers 2004; Robins and Ryder 2004). The demand for bush tomatoes and the variability in wild harvest have led to limited exploratory cultivation under irrigation in several areas outside Central Australia, notably in South Australia in the Yorke Peninsula to Riverland regions, Murray Bridge and Ceduna. Production has also been tried in the cooler areas of South Australia with little success (Anon. 2008). However, since bush tomatoes require dry, sunny and warm conditions for optimal plant health and to ripen and dry the fruit, it is unlikely to be suitable for cooler or wetter areas, unless adapted varieties become available (Hele 2006).

Initial research conducted by the Desert Knowledge Cooperative Research Centre (DKCRC) resulted in the establishment of a number of small-scale plots (<100 m²) to explore basic aspects of plant biology and potential yield. Transitioning from small research plots to large-scale horticulture is required for the commercial bush tomato industry to establish and grow. However, it is a substantial step to take a plant from the wild and use it in horticulture in a few generations. While it is easy in the short term to establish any wild plant on a reliable water supply, many attributes of its phenology (flowering and fruiting cycles) may be unfavourable for large-scale production as its behaviour under these conditions may change in unexpected ways from that observed in the wild.

In order to gain a preliminary understanding of the horticultural potential of bush tomatoes in Central Australia, we investigated the effect of (a) irrigation rates, (b) origin of plant material (variety hereafter) and (c) weed competition on yield.

Method:

The study site was situated on the Frank McEllister Horticulture Block at the Arid Zone Research Institute (AZRI) (23º 46'07S 133º 53'21"E) located south of Alice Springs. Approximately 11 000 seedlings were germinated from seed from three geographical sources (varieties): Napperby (22.50888^{o}S, 132.7529^{o}E), Ambalindum (23.38415^{o}S, 134.6839^{o}E) and Utopia (22.23271^{o}S, 134.5627^{o}E). Plants exhibited variation in morphology both within and between varieties. Commercially available irrigation lines (Netafim Australia, Victoria) with a drip spacing of 0.3 m were used to deliver three different irrigation treatments: 1, 2, and 3 L/h. The plants were watered for 2 hours per

week during April-May and August-September; and twice weekly for 2 hours at a time during October-March. The plants were not irrigated during the cooler parts of the year (June-July). Fruit was hand-harvested from November 2007 to April 2008 and both hand and machine-harvested from May to October 2008. The fruit was sun-dried and weighed to quantify yield.

Trials

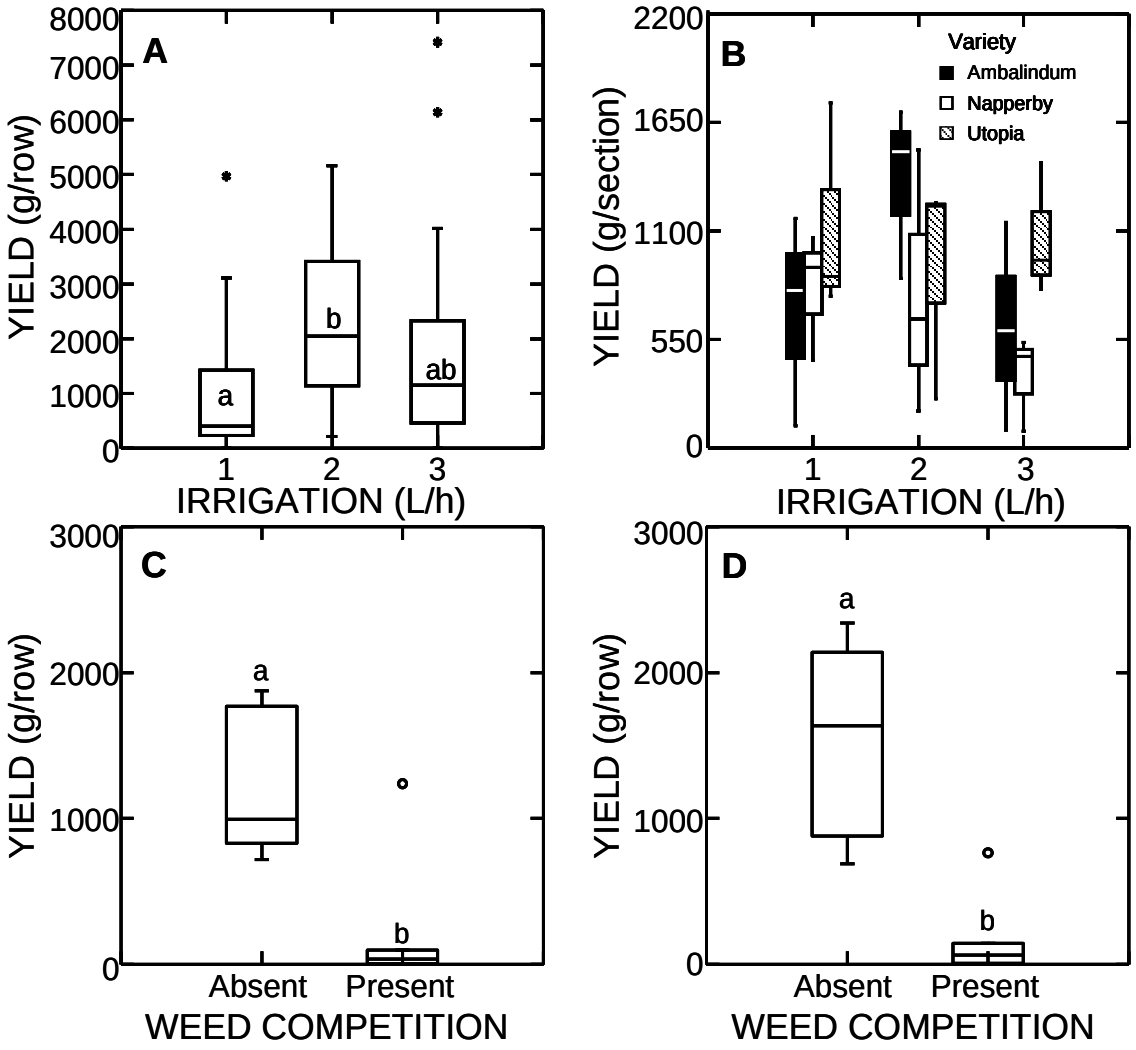
In Trial 1 we investigated the effects of three levels of irrigation on yield of the Napperby variety of bush tomatoes. Trial 2 was a comparative study of the effects of the same three levels of irrigation on the yield of the three varieties of bush tomatoes. In Trial 3 we compared the performance of two varieties of bush tomatoes (Ambalindum and Napperby) in the presence and absence of weeds, and varying irrigation rates. Weeding was regularly done by hand.

Data from Trial 1 was analysed as a mixed-model ANOVA (randomised complete block design) with block as a random factor and irrigation as a fixed factor. Trial 2 was analysed as 2-way fixed factorial ANOVA with variety and irrigation as factors. For Trial 3, the data was analysed as a 3-way fixed factorial ANOVA with variety, irrigation and weed competition as factors. Where significant treatment effects were detected, Tukey's HSD test was used to carry out post-hoc pair-wise comparisons of means.

Results:

There was a significant effect of block on yield of bush tomato ($F_{4,38}$=7.82, P<0.001) in Trial 1, but the differences in yield in relation to irrigation were consistent between blocks as evidenced by the absence of an interaction effect. Yield varied significantly in relation to irrigation level ($F_{2,38}$=3.22, P=0.05); yield was significantly higher in the 2 L/h treatment (2359.00 ± 392.43 g; mean ± SE) than in the 1 L/h treatment (1121.13 ± 350.98 g) (Figure 1A). The yield from 3 L/h treatment was 1955.07 ± 578.23 g and was not significantly different from either the 1 L/h or the 2 L/h treatments. The yield of the three bush tomato varieties (Ambalindum, Napperby, Utopia) in Trial 2 did not differ from each other under any of the irrigation rates (Figure 1B). The results indicate that at AZRI, 2 L/h was the optimal rate of irrigation in terms of bush tomato yield. The yield of bush tomatoes was significantly higher in the 2 L/h treatment than in the 1 L/h treatment. However, yield at 3 L/h irrigation was not significantly different from that at 2 L/h irrigation. This suggests that there was a threshold level above which irrigation had no further effect on yield. The irrigation rate did not significantly affect yield across varieties. The absence of an effect of irrigation on yield across varieties may be due to an inadequate definition of variety in terms of the underlying genotypes of bush tomatoes. It was expected that morphological characteristics would be similar in plants sampled from a location, but this was not observed to be the case. Differences were observed in leaf shape and colour, abundance of spines, growth habit, flower colour, and the shape, size and yield of fruit both across and within varieties. Careful characterisation of different genotypes of bush tomatoes and the implications of genotype-by-environment interactions (i.e. phenotypic plasticity) on plant traits and associated yield is required.

The presence of weeds significantly affected yield in both Amerlindum and Napperby bush tomato varieties consistently across all irrigation rates in Trial 3 ($F_{1,12}$=44.19; P<0.001). The yield of plants growing in competition with weeds (206.25 ± 112.19 g) (mean ± SE) was less than a fifth of those growing without weed competition (1375.42 ± 170.59 g) (Figures 1C and 1D).



The box represents the inter-quartile range and the line within the box indicates the median. Whiskers represent the range and symbols mark the outliers. Boxes with the same letter are not significantly different at *P*=0.05.

Figure 1. Box-plots depicting the effect of irrigation (L/h) on bush tomato yield (dry weight) in (A) Trial 1, and (B) Trial 2, and the effect of weed competition (Trial 3) on yield (dry weight) of (C) Ambalindum and (D) Napperby bush tomato varieties

From a production system perspective, future studies need to consider changes in yield of bush tomatoes with age to determine if treating this species as an 'annual' in a production sense would be more profitable than harvesting from the same individuals across multiple seasons. The efficiency of the harvesting method is another aspect that needs careful consideration, paying particular attention to optimising harvest machinery for bush tomato harvest. Future research in horticultural production systems needs to investigate the compatibility of commercial-scale activities (e.g. automated irrigation systems and mechanical harvesting) with the wild harvest industry and the developmental aspirations of Aboriginal participants to ensure benefits obtained from commercial production of bush tomatoes flow to Aboriginal people living in arid zones of Australia.

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Note: A comprehensive report on this project (including the above results) has been published as a Working Paper of the DKCRC. Citation details are:

Ellis, G., Oliver, G., Vincent, A. and Raghu, S. (2010). The effect of irrigation and weed competition on yield of a commercial-scale bush tomato (*Solanum centrale)* production system in arid Central Australia. DKCRC Working Paper 74, Desert Knowledge CRC, Alice Springs.

PROJECT: Environmental Impacts of Plant Industries on the Soil in the Northern Territory – a Literature Review

Project Officers: S. Smith and J. Hill (NRETAS)

Location: NT

Keyword(s): environment, soil, plant industries

Objective:

To review current literature on the impact of plant industries on soil resources in the Northern Territory (NT).

Background:

The Department of Resources (DoR) is focusing on understanding the environmental impacts of plant industries on soil and water in the NT. A review of what is already known helps to highlight knowledge gaps for future investigation.

The review will benefit DoR and/or stakeholders by:

- Recording what has already been achieved so that duplication of effort is avoided.
- Identifying issues of concern and priority to the government/community on which to concentrate future effort.

- Facilitating the development of new projects that focus on priority issues in order to seek internal and external funding.
- Facilitating the development of policy based on clear information.

Method:

On line databases (Department of Natural Resources, Environment, the Arts and Sport Technical Publications, Australian Agriculture and Natural Resources Online, and Agriculture and Natural Resources Compilation) were searched. Reference lists at the back of relevant publications were also used to find leads to new articles. DoR staff also contributed material.

Relevant articles were catalogued using Refworks and all references were transferred to End Note.

The material was critically reviewed, resulting in two papers; one has been already published and the other will be published later in 2010 (see below).

Results:

Technical Bulletin 330, titled '*The Environmental Impact of Plant Industries on Inland Water'* was published in 2009. The main findings were:

- The impact of plant industries in the NT on water resources is currently small because of the small size of the industries in comparison with the large size of the NT.
- The climate of the NT is mainly arid, with a distinct wet season in the Top End. Plant industries rely mainly on ground water stored in aquifers for irrigation in the dry season and little surface water is used.
- Plant industry activities, which include clearing land and planting commercial crops, and extracting water from aquifers, change hydrological systems, which in turn affect erosion risk and water balance between runoff, groundwater recharge, soil storage and evaporation. Ongoing hydrological research is needed to fully understand these impacts.
- The use of pesticides and mineral fertiliser may lead to off-target effects with water as the carrier. There are few studies or documented evidence indicating the occurrence of this in the NT.
- The use of saline ground water, especially in Central Australia, is a risk to surface soils. However, such risk can be reduced by judicious monitoring. There is little risk from surface soil salinity occurring due to rising groundwater tables in the NT.

Another publication on the impact of plant industry activities on the soil will be published later in 2010. Preliminary findings from that paper show that:

- Plant industry practices in the NT have little overall impact at present on the condition of the soil, although there are significant impacts in localised areas. The characteristics of the climate and the soils commonly used by plant industries increase the risk of erosion, loss of carbon, surface crusting and sealing, soil acidification and off-target movement of soil nutrients. They, however, reduce the risk of increased salinity.
- Soil erosion is locally significant in the NT. There is considerable risk of soil erosion caused by water in Top End uncovered soils due to the intensity of rainfall and poorly structured soils. Wind erosion is less significant but is still important. Current plant industry practices contribute little to the acceleration of wind erosion. Much work has been done on the mechanisms and processes of soil erosion and its control in the NT. Further work is needed in this area, particularly in extension, to ensure adoption of existing technology to control both wind and water erosion.

- Soil acidification has been detected in the NT. It is recognised as a risk in the semi-arid tropics, but little work has been done on it. Acidification can occur under specific plant species, especially leguminous crops. Fertiliser use may also lead to acidification. More work is needed to determine the extent of the problem and to develop ways to tackle it, if it is significant.
- NT soils are low in organic carbon and have been shown to rapidly lose it when cultivated. There is, however, little information on the effect of plant industry practices on carbon levels in the soil, which requires further investigation.
- Crusting or surface sealing of NT soils is common in the Top End and relates to the breakdown of the already limited structure of surface soils in the region after the removal of vegetation and/or cultivation. Crusting can be very detrimental to emerging seedlings in cropping operations. It can be mitigated by frequent shallow cultivations or, more preferably, by using stubble retention and minimum-tillage techniques.
- Off-target nutrient movement, especially downward movement of nutrients beyond the root zone of desirable plants, is probable in the NT due to the use of light and well-drained soils in many plant industry operations. A limited amount of work has been completed on measuring off-target nutrient movement. However, more research is needed, particularly in horticultural crops. Methods to reduce the problem, including the use of deep-rooted cover crops to harvest nutrients deep in soil need further investigation.

PROJECT: The Relationship between Mango Yield and the Number of Days with Temperatures Higher than 32 ºC in July in the Darwin Area and the Possible Consequences of Climate Change

Project Officers: P. Stephens and N. Hartley

Location: Darwin

Keyword(s): mango, temperature, climate change, yield, overhead irrigation

Objective:

To determine the influence of increased temperature due to climate change on mango yield in the Northern Territory (NT).

Background:

The 2007 Intergovernmental Panel on Climate Change (www.climatechangeinAustralia.gov.au) provides the most recent climate change modelling data for Australia. It looks at three scenarios of low, medium and high continued greenhouse emissions, given that future climate change is likely to depend on the amount of emissions. The predicted temperature and rainfall changes in the major mango producing areas of Australia are detailed below. The data represents the mid-point of the various model results. M/B/B/A/B represent Mareeba/Burdekin/Bowen/Ayr/Bundaberg.

Temperature

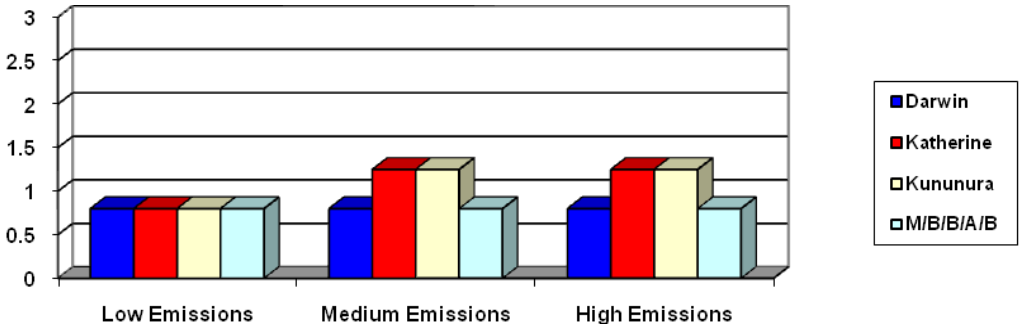

Figure 1. Predicted change in temperature (ºC) by 2030

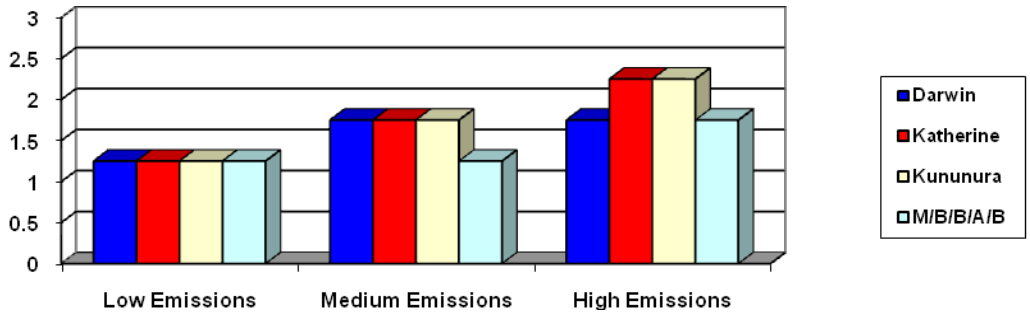

Figure 2. Predicted change in temperature (ºC) by 2050

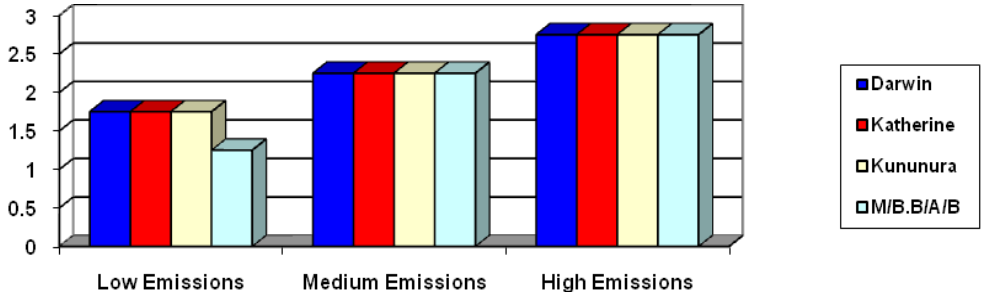

Figure 3. Predicted change in temperature (ºC) by 2070

Rainfall

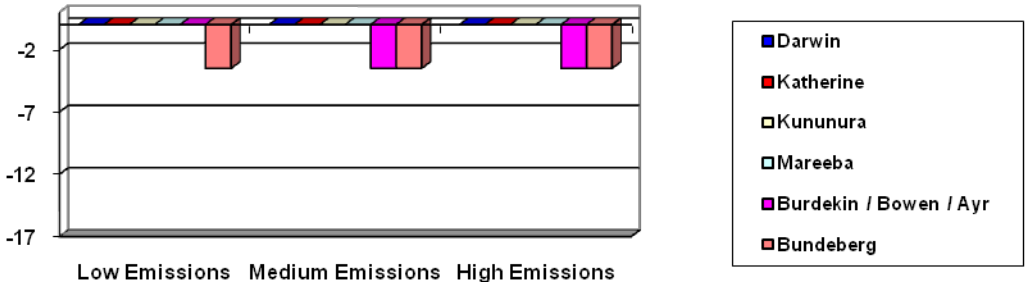

Figure 4. Predicted change in rainfall (%) by 2030

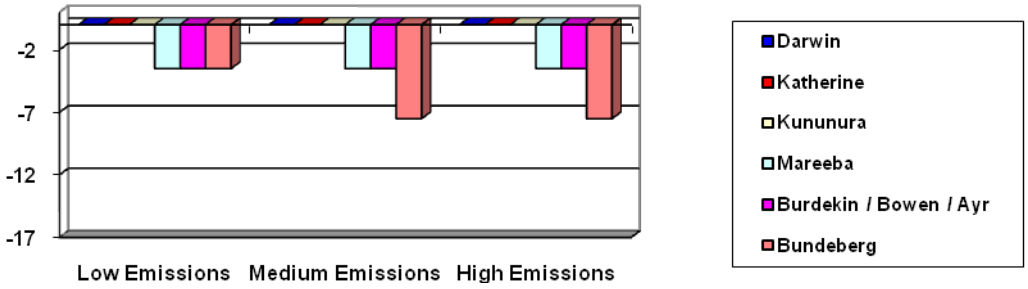

Figure 5. Predicted change in rainfall (%) by 2050

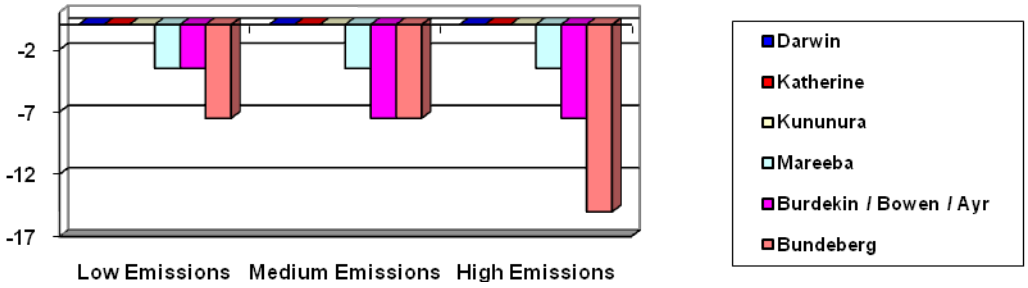

Figure 6. Predicted change in rainfall (%) by 2070

Each major mango growing area in Australia may be subject to a 2-2.5 ^{o}C rise in temperature by 2070, should medium greenhouse emissions continue (the most likely scenario). In contrast, the predicted changes in rainfall due to climate change under each of the emission scenarios in Darwin, Katherine or Kununurra are minimal. Mareeba can expect a -2 to -5% reduction in rainfall, while Burdekin, Bowen, Ayr and Bundaberg can expect a -5 to -10% reduction in rainfall by 2070 using the same medium emissions scenario. Given these predictions, research has been initiated to evaluate the possible impact of temperature increases on mango production in the NT.

Method:

To assess the possible impact of temperature on mango production in the NT, yield data for Kensington Pride (KP) trees in outer Darwin (Berry Springs) was correlated over a period of eight years with a variety of temperature records (Bureau of Meteorology, Middle Point) before, during and after flowering. This included minimum and maximum daily temperatures between March and August. The trees, which were planted in 1992, were uniform and were maintained under a relatively constant management, with flowering typically occurring between late May and early July. Yields between 2002 and 2009 varied between four and 13 trays per tree, which was below the regional average.

Results:

There was a significant negative correlation (*P*<0.01) between mango yield and the number of days with more than 32 ºC in July (Figure 7). Using linear regression models, this predictor explained 88% of the variation in mango yield over the eight year trial period. The results of the model suggest that a further increase of 1 ºC in average daily temperature may result in an 18 to 28% reduction in mango yield in the highest yielding seasons.

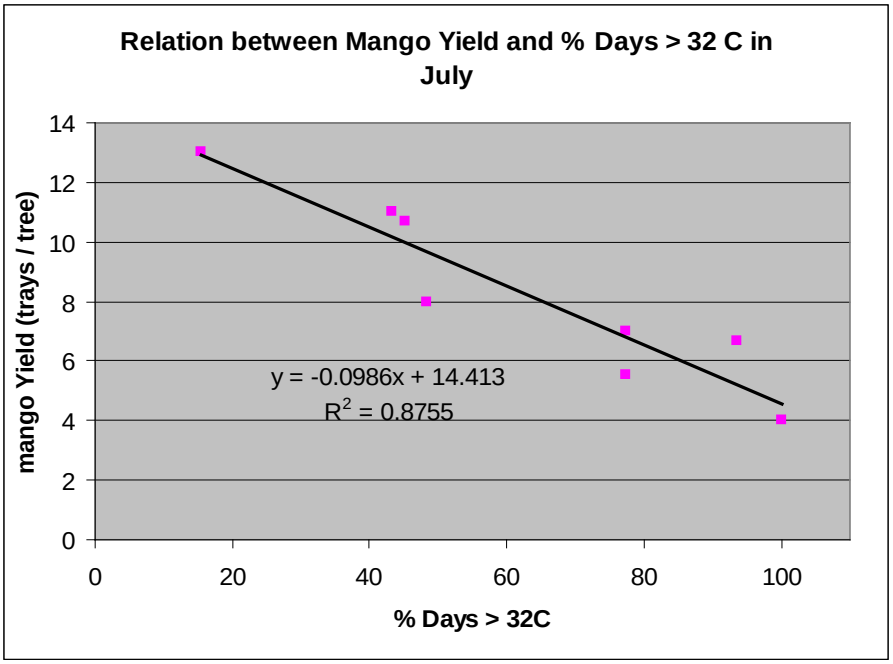

Figure 7. The relationship between mango yield (2002-09) and the number of days with over 32 ºC in July in an orchard in outer Darwin

For the correlation between mango yield and temperatures in July, the mean yield data per tree was estimated by dividing the yield from the block by the number of trees. Obtaining the mean in this manner (rather than from individual trees) may have exaggerated the r-squared value. While these limited results suggest that mango yield in outer Darwin may be significantly reduced by even a small increase in temperature due to climate change, long-term yield data is being sought from other suitable and more productive orchards containing KP and other varieties to help validate this hypothesis. In 2010, a trial will evaluate the effect of reducing tree temperature (by using evaporative cooling with overhead irrigation) on mango fruit number and yield.

Acknowledgement:

Thanks to Murray Linton and Ian Baker for helping to conduct this trial.

PROJECT: Comparison of Relative Efficacy of Different Sub-label Rates of Two Post-emergent Herbicides for the Control of Common Weeds under Irrigated Production Systems in Central Australia

Project Officers: M. Hidalgo, G. Oliver and S. Raghu

Location: Alice Springs

Keyword(s): integrated weed management, herbicide, sub-label rates, date-palm, irrigated production

Objective:

To investigate if herbicide use can be reduced in arid zone irrigated production systems by applying below label recommended rates.

Background:

Increasing attention is being paid to the sustainable use of resources in primary industries. A central component of sustainability is to reduce external inputs, such as agrichemicals, in the production of food and fibre.

Agriculture in arid Central Australia relies on irrigation using ground water. One of the consequences of localised irrigation of high frequency is an increase in weed density along drip lines, resulting in increased weed-crop competition. As elsewhere, Central Australian growers rely on herbicides for managing weeds on their farms. Such management adds significantly to production costs and also poses health risks to farm workers and the environment.

Numerous studies have shown that optimal rates of application of herbicides vary in relation to the environmental context (Boström and Fogelfors 2002) and that in many instances the required dose/rate of application of such chemicals is below the dose/rate recommended by the manufacturers of the chemicals (Zhang et al. 2000)

To assist growers to reduce their herbicide inputs, we investigated the feasibility of reducing herbicide in weed management in irrigated production systems in Central Australia.

Method:

The experiment was conducted at the Arid Zone Research Institute between June and December, 2009. The area of study contains 60 date palms comprising 18 cultivars, under drip irrigation. Prior to commencement of the trial, all date palms were pruned, and the weeds in the area of study were slashed. The canopy area of each date palm was projected and marked on the ground, becoming the sampling unit, where treatments were applied using a Solo Backpack sprayer once a month for four months.

We selected two commonly-used post-emergent herbicides in Central Australia (Basta and Amitrole T) and evaluated whether sub-label rate applications of these herbicides (applied individually) would deliver the same level of weed suppression as the recommended label rate. We compared the effects of label rate (LR) = Dose 1 or (D1) and three sub-label application rates (85% LR (D2), 75% LR (D3) and 65% LR (D4)) of these herbicides with positive (Roundup) and negative (water) control treatments to assess their relative efficacies.

The living/green weed cover was estimated as a proportion (%) of the projected canopy area of each of the date palms prior to each herbicide application. An economic benefit-cost ratio (EcBCR) and an environmental benefit-

cost ratio (EnBCR) were calculated. EcBCR was calculated at each sampling time as the percent reduction in cover (relative to initial weed cover) per unit cost ($) of active ingredient (ai) used in the treatment, while EnBCR was calculated at each sampling time as the percent reduction in cover per unit volume (mL) of ai used in the treatment. EnBCR assumes that reducing the volume of ai released in herbicides is beneficial to grower health and the environment.

The data was analysed using a repeated measures ANOVA with treatment as a between subjects factor and time as a within subjects factor. Where significant treatment effects ($P<0.05$) were detected, protected posthoc pairwise comparisons were made using Tukey's HSD test.

Results:

The data revealed that a reduction in the dose of Amitrole T and Basta by 35% (D4) and 25% (D3) respectively, resulted in the same reduction in weed cover (%) over the trial duration as the recommended label rate (D1) of Roundup. This translates to a potential cost savings of 35%. Roundup at the recommended label rate was the most economically-efficient treatment in terms of reduction in weed cover per dollar of ai. Amitrole T at 65% of the label rate was the most benign environmental treatment in terms of reduction in weed cover per unit volume of ai, suggesting its use is likely to be least deleterious to the environment and the health of farm personnel. The trends in rates of weed control over time are being analysed to evaluate which herbicide x rate combinations perform best.

Our results also suggest that the use of a combination of herbicides with different modes of action can be used in an integrated manner to minimise the risk of resistance developing in weeds. This also provides an opportunity for the grower to integrate herbicide x rate combinations that have optimal EcBCRs and optimal EnBCRs. While our results are indicative of the potential for more sustainable use of chemical weed control, we stress that they are of a preliminary nature. Further evaluations are needed in this regard to gain a better understanding of the risks of herbicide resistance from such sub-lethal use.

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PROJECT: Diagnostic Protocol for Sugarcane White Leaf Phytoplasma

Project Officer: L. Tran-Nguyen

Location: Darwin

Keyword(s): sugarcane disease, biosecurity, phytoplasma, PCR, leaf hopper, Thailand

Objectives:

To conduct extensive training to diagnose sugarcane white leaf phytoplasma.

To document field symptoms of sugarcane white leaf disease and compile a national diagnostic protocol for sugarcane white leaf phytoplasma.

Background:

White leaf diseases of sugarcane (*Saccharum* spp.) and *Saccharum* hybrids are associated with the sugarcane white leaf (SCWL) phytoplasma (Nakashima et al. 1994; Wongkaew et al. 1997). In many parts of Asia, SCWL and sugarcane grassy shoot disease can severely reduce yields (Blanche et al. 2003). It is the most destructive sugarcane disease in Thailand (Nakashima et al. 2001). To date, there have been no published reports of white leaf phytoplasma diseases in Australian sugarcane (Chen and Kusalwong 2000; Tran-Nguyen et al. 2000). However, there are two phytoplasmas closely related to SCWL and members of its group ("*Candidatus* Phytoplasma cynodonitis") which are found in northern Australian grasses. These closely related phytoplasmas are cynodon white leaf found in *Cynodon dactylon* and sorghum grassy shoot found in *Sorghum stipoideum* and *Whiteochloa cymbiformis* (Schneider et al. 1999). The presence of these two phytoplasmas in native and introduced grasses could pose a threat to the Australian sugarcane industry, valued at between $850 and $1150 million (SRDC 2007/2008 Annual Report; http://www.srdc.gov.au/pages.aspx?id=35).

SCWL disease is characterised by the proliferation of tillers and leaf chlorosis (Wongkaew et al. 1997) (Figure 1). Other symptoms include stunted stalks, abnormal tillering and the absence of side shoots on the upper part of infected stalks (Ling 1962; Liberato et al. 2007). The disease is transmitted by leafhoppers (*Matsumuratettix hiroglyphicus)* (Wongkaew et al. 1997) and *Yamatotettix flavovittatus* (Ritthinson 2004) which are not found in Australia.

Figure 1. Symptoms of white leaf disease in sugarcane

Method:

The training was conducted in Thailand where SCWL disease is prevalent. Field surveys were conducted in Khon Kaen in north-east Thailand. Sugarcane samples showing classic white leaf symptoms were collected in addition to symptom less sugarcane samples. The laboratory component was conducted at the Prince of Songkla University in Hat Yai.

Since phytoplasmas cannot be cultured, their detection is based on polymerase chain reaction (PCR) techniques (Wongkaew et al. 1997). Total DNA extractions were performed using a 'phytoplasma enrichment' method using midribs of sugarcane with white leaf symptoms. Phytoplasma DNA is amplified using universal primers fP1 (Deng and Hiruki 1991) and rP7 (Schneider et al. 1999). The target region includes the 16S ribosomal (rRNA) gene, the intergenic spacer region and the start of the 23S rRNA gene.

PCR reactions were set up as described in Table 1 with the addition of 2 µL DNA template, cycling conditions as described in Table 2.

Table 1. The universal phytoplasma PCR test

Reagent	Volume for master mix of 10 reactions (µL)
PCR water	320
10 X PCR buffer	50
25 mM $MgCl_2$	30
P1/P7 (10 µM)	25
dNTPs (2.5 mM)	50
I-*Taq*	5

Table 2. PCR cycling conditions for universal phytoplasma primers (P1/P7)

Steps	Temperature (ºC)	Time (min)	No. of cycles
Initial denaturation	94	5	1
Denaturation	95	1	
Annealing	55	1	35
Elongation	72	1.5	
Final elongation	72	10	1

PCR products were separated on a 1% agarose gel and electrophoresis was conducted at 80 V for 40 minutes. Genetic relatedness can be conducted using restriction fragment length polymorphism (RFLP) and/or DNA sequencing. RFLP or DNA fingerprinting using the restriction enzyme, *Mse* I, can clearly distinguish grass phytoplasmas. Restriction digestion was set up as described in Table 3 using the P1/P7 PCR product.

Table 3. Restriction digestion of P1/P7 PCR products

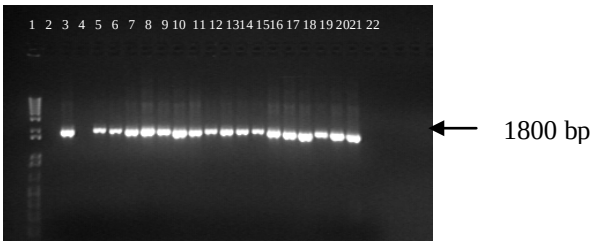
Reagents	Volume per reaction (µL)
Buffer	2
Mse I	1
Bovine serum albumin	0.2
Sterile distilled water	9.8

A 7 µL volume of P1/P7 PCR product were added to the mixture and allowed to digest for 2 hours at 37 ºC. The DNA bands were separated on an 8% polyacrylamide gel at 150 V for 5 hours.

P1/P7 PCR products were purified using the Qiagen PCR purification commercial kit according to the manufacturer's instructions. Sequencing was conducted at Bioscience North Australia, Charles Darwin University using the Big Dye Terminator mix. Bioinformatic analyses were conducted using GenBank (http://www.ncbi.nlm.nih.gov/) BLAST program and multiple nucleotide alignments using Geneious software.

Results:

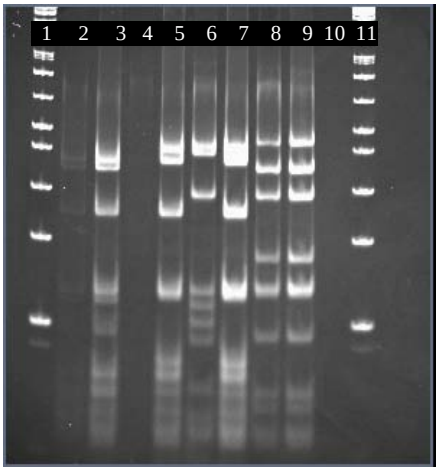
The universal phytoplasma PCR test using fP1/rP7 primers amplified a fragment 1800 bp in size (Figure 2).



Lane 1, 1 Kb DNA marker; lane 2, healthy tissue culture sugarcane; lane 3, – 20 sugarcane with white leaf disease; lane 21, Periwinkle Phyllody phytoplasma and lane 22, Sterile distilled water.

Figure 2. P1/P7 Phytoplasma PCR test

Phytoplasma DNA was detected in all but one sample with SCWL symptoms; no phytoplasma DNA was detected in symptom less samples (data not shown). RFLP analysis confirmed the unique DNA fingerprint of samples with SCWL compared with other reference phytoplasma strains (Figure 3).



Lane 1 and 11, 1 Kb Plus Marker; lane 2, SCWL I; lane 3, SCWL II, lane 4, N/A; lane 5, CWL; lane 6, SGS; lane 7, Bermuda grass white leaf (BGWL); lane 8, Tomato big bud (TBB), lane 9, Sweet potato little leaf vinca 4 (SPLL-V4); lane 10, Blank; lane 11, 1 Kb Plus marker.

Figure 3. *Mse* I restriction fragment length polymorphism of P1/P7 PCR products

Sequences analyses indicated a high similarity (> 98%) nucleotide sequence match between the samples and SCWL accessions within the GenBank database. Therefore, confirming that the sugarcane white leaf phytoplasma

was associated with the white leaf disease collected from Khon Kaen. Updated field symptom photos were taken and a diagnostic protocol was compiled as part of the training. The protocol will be peer reviewed and will be available online in the Plant Biosecurity Toolbox (http://www.padil.gov.au/pbt/).

Benefits to the Northern Territory include establishing international contacts with Thailand ranging from university academia, post graduate students to government researchers. These networks can be extended to other crops and diseases such as banana, citrus and mango and their associated diseases, such as Panama wilt and citrus greening, all of which are relevant to our local industries. Images were documented for sugarcane white leaf phytoplasma disease and the associated vectors. Knowledge of the disease vector is crucial for surveillance purposes. Although sugarcane is not grown commercially in the NT, our close proximity to neighbouring Asian countries and the existence of a closely related phytoplasma in our native grasses increases the chances of disease establishing on our shores. The white leaf disease can potentially cause 100% crop loss. Therefore, it is a high risk pest for the Australian sugarcane industry.

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Acknowledgement:

Thanks to Dr. Ratana Sdoodee for supporting the training at the Prince of Songkla University, Hat Yai, Thailand and the Department of Agriculture, Fisheries and Forestry for funding the training scholarship.

PROJECT: Identification of Exotic Moths by DNA Bar-coding

Project Officers: L. Tran-Nguyen and B. Thistleton

Location: Darwin

Keyword(s): moths, biosecurity, PCR, DNA bar-coding

Objective:

To conduct extensive training to identify exotic moths by DNA bar-coding tools and to compile a national diagnostic protocol.

Background:

At present, 17 species belong to the Lepidoptera order listed on the Northern Australia Quarantine Survey 2009 target list (http://www.daff.gov.au/aqis/quarantine/naqs/target-lists#IV). These plant pests target a variety of hosts of agricultural importance, such as citrus, mango, sugarcane, rice, banana and cotton. An example is the mango-shoot borer (*Chlumetia transversa)* (Walker), which is exotic to Australia. This particular borer has a close relative, *C. euthysticha* (Turner), which is already present in Australia. Another example is the mango fruit borer (*Citipestris eutraphera)* (Meyrick), which was recently found in Darwin. In most cases, the insects are discovered during their early life stages, i.e. egg or larva. Morphological identification requires breeding to adulthood that can be time-consuming. DNA bar-coding provides an avenue to identify the specimens quickly. The DNA bar-code information obtained from local species allows a baseline level of data for situations where an exotic insect may appear. Australia has 10 590 lepidopteran species from 89 families; DNA bar-coding has been conducted for only 1796 (17%) species (http://www.lepbarcoding.org). DNA bar-coding targets a segment of the cytochrome c oxidase subunit I gene (*COI*), since this gene appears to be among the most conservative protein-coding regions in the animal mitochondrial genome (Brown 1985; Folmer et al. 1994).

Table 1. The pyraloids selected for DNA bar-coding as part of diagnostic training

Chilo auricilius	*Chilo infuscatellus*
Chilo partellus	*Chilo polychrysus*
Chilo sacchariphagus	*Chilo terrenellus*
Deanolis sublimbalis	*Scirophaga excerptalis*
Scirophaga novella	*Citripestris eutraphera*
Citripestris sagittiferella	*Orthaga eudrusalis*

Method:

Adult moths were pinned and their genitalia were dissected to obtain morphological identification. A minimum of three legs were removed and preserved in 100% alcohol. Larval specimens, where applicable, were preserved in 100% alcohol in preparation for DNA extractions. Pyralid specimens processed to date include *Citripestris eutraphera, Deanolis sublimbalis, Chilo sacchariphagus, C. auricilius, Scirpophaga excerptalis* and *Orthaga euadrusalis*.

Two commercial extraction kits were used for DNA preparation from dried adult moth legs and larval specimens. The first method utilised a rapid extraction technique using Prepgem (ZyGem, New Zealand) where the specimen was chopped into minute pieces and transferred to a polymerase chain reaction (PCR) tube. Reactions were set up as described in Table 2.

Table 2. Reagents used for DNA extractions as per the ZyGem protocol

Reagents	Volume for 1 reaction (µL)
Enzyme	1
10 x buffer	4
Sterile distilled water	36
Total	41

The tubes were placed in a PCR thermo-cycler and subjected to incubation at 75 ºC for 1 hour followed by 95 ºC for 5 minutes to allow the DNA to be released into the solution. The second method used the Qiagen DNA tissue extraction kit following the manufacturer's instructions. The specimens were initially crushed using a micro-pestle with buffer before being subjected to purification spin columns.

DNA samples were subjected to a PCR test targeting the *COI* gene using primers LCO1490 and HCO2198 (Folmer et al. 1994). PCR reactions were set up as described in Table 3, cycling conditions as detailed in Table 4.

Table 3. Universal DNA bar-coding *COI* PCR test

Reagent	Volume (µL)
Sterile distilled water	7.15
10 x buffer + $MgCl_2$	1
25 mM $MgCl_2$	0.4
10 mM dNTPs	0.2
2 µM LCO1490	0.3
2 µM HCO2198	0.3
HiFi Taq	0.15
DNA	0.5

Table 4. PCR cycling conditions for universal DNA bar-coding *COI* PCR test

	Universal primers (P1/P7)		
Steps	**Temperature (ºC)**	**Time**	**No. of cycles**
Initial denaturation	94	2 minutes	1
Denaturation	94	0.15 second	
Annealing	50	30 seconds	40
Elongation	72	45 seconds	
Final elongation	72	7 minutes	1

PCR products were separated on a 1% agar-rose gel and electrophoresis was conducted at 80 V for 40 minutes. Products were subsequently purified using the Qiagen PCR purification kit and sequenced using LCO1490 and HCO2198 primers. Bio-informatics analyses were conducted using the bar-code of life data systems (BOLD) database (http://www.boldsystems.org/, Ratnasingham and Hebert 2007) and multiple nucleotide alignments using Geneious software.

Results:

A 700 bp region of the *COI* gene was amplified from all samples (Figure 1). The PCR products were purified and sequenced to obtain the DNA bar-code. Sequence analyses confirmed that the specimens were pyralids but no high similarity nucleotide sequence matches were found. This was expected, since DNA bar-coding of the target pyralids for this study has not been conducted previously. Comparative studies between adult and larval specimens indicated they were 100% identical to one another, thus confirming that DNA bar-coding from larvae is potentially a useful tool to identify moths during their early life stages. This will allow preliminary diagnosis until morphological identification can be conducted.

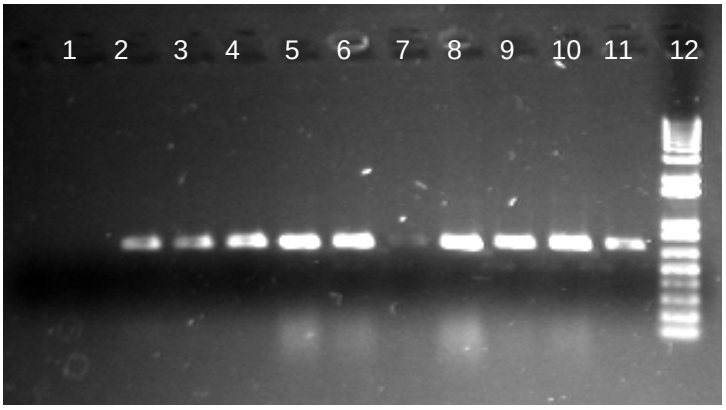

Figure 1. Cytochrome c oxidase subunit I PCR test: lane 1, sterile distilled water; lane 2, *D. sublimbalis* 61277 (leg); lane 3, *D. sublimbalis* 61279 (leg); lane 4, *D. sublimbalis* 61278 (leg); lane 5, *D. sublimbalis* 61276 (larvae); lane 6, *D. sublimbalis* 61275 (larvae); lane 7, *D. sublimbalis* RM1 (leg); lane 8, *D. sublimbalis* RM1 (larvae); lane 9, *C. eutraphera* 1 (larvae); lane 10, *C. eutraphera* 2 (larvae); lane 11, *C. eutraphera* 2 (leg); lane 12, 1 Kb DNA marker

A study of tussock moths in New Zealand also showed that DNA bar-coding tools could be used to identify insects during their immature life stages (Armstrong et al. 2003). The molecular tools could also be used to provide a rapid and accurate identification of morphologically similar insect species (Armstrong and Ball 2005).

This study will provide DNA bar-codes for the listed exotic pyraloids, thus building a DNA database for use if an incursion occurs in the Northern Territory (NT). The information will enable rapid identification, thus increasing the NT's capacity to respond to an incursion and formulate eradication strategies. A diagnostic protocol will be compiled in collaboration with AQIS, peer reviewed and will be available online in the Plant Biosecurity Toolbox (http://www.padil.gov.au/pbt/).

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Acknowledgement:

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PUBLICATIONS, CONFERENCE PAPERS AND PRESENTATIONS

Scientific Journal/Proceedings Publications

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