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Smart selection - accelerating crop & livestock genetic gain



The magnitude of Heterosis in grain Sorghum

Daniel Otwani¹, Colleen Hunt², Alan Cruickshank, Anna Koltunow¹, Emma Mace¹ and David Jordan

¹Centre of Crop Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

²Department of Agriculture and Fisheries, Agri-Science Queensland, Hermitage Research Facility 604, Yangan Rd, Warwick 4370, Queensland, Australia

Abstract: Heterosis has been exploited for over a century for crop productivity improvement and is poised to contribute immensely to future productivity gains. Understanding the magnitude of heterosis in a specific crop x environment context is central to its useful exploitation. To address this question, a set of 112 diverse grain sorghum hybrids and their 22 inbred parents of local and exotic origins were tested in five contrasting sorghum growing environments across two years in Queensland Australia. The local parents were products of the public sorghum program run by the department of primary industries whereas the exotic parents were the product of various international programs. To assess the magnitude of heterosis, yield, plant height, days to flowering and grain weight were measured and used in the estimation of heterosis. Mid parent heterosis for yield ranged from -25 to 217% with hybrids derived from crosses between exotic parents having the highest average heterosis across all the test environments for all the measured traits except for the flowering time in one environment. Further, heterosis was not associated with site mean yield for the tested material in this study, in contrast to recent reports in maize. Of note, heterosis for grain number was 68%, in contrast to just 1.8% for grain weight heterosis. Additionally, we identified that hybrids from exotic inbred parents showed elevated heterosis levels and hypothesise that this is likely due to a combination of inferior performance of the exotic inbreds compared with local inbreds in the test environments and the greater average genetic differentiation between parents of hybrids involved in crosses between exotic parent lines compared with crosses between locally developed lines. The concepts presented in this paper reiterate that heterosis estimates should thus be interpreted in a specific genetic and environmental context to allow its useful exploitation in plant breeding.



Investigating the heritability of husk spot resistance selection traits to optimise genetic gain in macadamia

Jasmine Nunn¹, Bruce Topp¹, Olufemi Akinsanmi¹, Craig Hardner¹, Mobashwer Alam¹, Katie O'Connor²

¹Centre of Horticultural Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

²Department of Agriculture and Fisheries, Nambour, Australia.

Abstract: Husk spot is a fungal disease of macadamia pericarps caused by *Pseudocercospora macadamiae*. Infection can induce premature abscission of infected fruit, with losses of over 30% of saleable yield recorded in susceptible cultivars. Management strategies add to farm costs. Thus, husk spot resistance is a priority in the breeding of new varieties. Several disease expression variables have previously been measured to assess husk spot susceptibility; however, their genetic architecture is unknown. The objective of this study was to evaluate incidence and severity parameters in a diverse breeding population to enable selection for resistance and heritability estimation. Broad and narrow sense heritabilities were estimated for each trait to compare the levels of genetic control over the traits, and the potential for genetic gain if used for selection. Over 300 open-pollinated progeny from 32 maternal parents, and replicated clones of 24 parents were inoculated. Fruit were phenotyped for total incidence (percentage symptomatic fruit), severity (percentage fruit surface area affected), lesion number, necrotic incidence and lesion number, lesion intensity, and sensitivity to premature abscission when diseased. The most susceptible genotypes had 5-10 necrotic lesions per husk and 8 genotypes were identified with less than 5 per husk. Low heritabilities (~0.1 - 0.2) were observed over most traits, indicating low genetic control. Mean number of necrotic lesions per fruit had the highest broad and narrow-sense heritability (~0.2), and therefore may provide some genetic gain if used as a selection trait. These results will facilitate selection for husk spot resistance in the Australian macadamia breeding program.

Investigating the effects of macadamia rootstocks on plant vigour

Pragya Dhakal Poudel¹, Bruce Topp¹, Mobashwer Alam¹

¹Centre of Horticultural Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Plant vigour is one of the key obstacles of efficient orchard system in macadamia (*Macadamia integrifolia*, *M. tetraphylla*, and hybrids). Vigorous trees restrict low-density planting in commercial orchards. In absence of low vigour scion and dwarfing rootstocks this problem is addressed by pruning and hedging in mature macadamias, which appear to be the major expenses for nut production. However, the ability of rootstocks to reduce tree vigour has been explained as an important advantage for many fruit-crop growers. To identify vigour managing rootstocks, this study investigates 21 diverse genotypes of macadamia including 12 elite selections from the national breeding program, an accession of the wild species *Macadamia janseni*, and eight cultivars. Scions of HAES 741 were whip-grafted onto 245 seedlings and 188 cuttings. Both rootstocks and scions were phenotyped for growth parameters (tree height, rootstock and scion height and trunk circumference, canopy width and depth) from 2017 to 2021. The variability in plant vigour traits due to the effect of rootstock genotypes and propagation types (grafted and ungrafted / seedlings and cuttings) are being evaluated. The growth parameters of the rootstocks in nursery and in the field will be compared. Results from this study will provide a basis of selecting superior rootstocks for high performance in future high density orchard systems.

AI to optimize agricultural breedings

Chensong Chen¹, Seema Yadav¹

¹Centre of Animal Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Emerging biological and genetic discoveries provide more chances to find out new advanced breeding technologies. However, it could be a large challenge for scientists to combine genotype and traits in vary levels. For example, determining breeding value for a certain SNP with given traits from a joint marker array could be quite difficult if the breeder need to consider mixed factors such as polyploid or non-additive effects. Meanwhile, the non-genetic factors such as environmental factors might contain hidden effects to certain SNPs. Previous studies have built many elegant statistical algorithms such as Bayes-A/B, GBLUP for solving breeding requirements based on complex genome constructions but this world still need more novel approaches to speed up the "Artificial evolution". This research would introduce multiple kinds of artificial intelligence strategies including convolutional neural network for optimizing current breeding system. Many of them are already playing important roles in imaging classification, audio prediction field, while the above tasks are full of discovered or hidden patterns which would be very similar with genetic prediction. The study is expected to build up a combination of conventional statistical models and machine learning approaches and try to predict traits of many agriculture species including sugarcane and cattle. By importing these digital brains with current statistical predicting methods, the system might have capacity to merge genetic features in different levels efficiently, finally increase the accuracy and speed in both genome prediction and selection.



Allele specific isoform expression in Brahman cattle

Shreya Bardoloi¹, Loan Nguyen¹, Bailey Engle¹, Ben Hayes¹, Elizabeth Ross¹

¹Centre of Animal Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Allele specific expression (ASE) is the imbalance in the expression of paternal and maternal alleles which results in phenotypic changes in the individual. The few ASE studies that have been reported in cattle have used RNAseq data to test for ASE. Since RNAseq produces short reads much of the data is unable to be associated with a haplotype of origin, as it doesn't overlap a SNP. Follow in from this, the isoform that is being expressed by each haplotype has not been considered. We hypothesized that some of the ASE was not due to different levels of expression between haplotypes, but rather that each haplotype was expressing a different isoform. Allele specific isoform expression (ASIE) can only be identified when transcriptome data is able to be characterized both in terms of haplotype of origin, and isoform. To address the hypothesis, we conducted an ASIE analysis in Brahman cattle using ISO-seq (PacBio) data obtained from a single animal. A phased haplotype level assemble of the animal was first used to assign each ISO-seq read to the haplotype of origin. For each gene, a contingency table of haplotype of origin (columns) and isoform (rows) was calculated. The values in the table were the number of ISO-seq reads for each isoform from each haplotype. A fishers exact test was used to test for independence between the isoform and the haplotype of origin. We found that 17 genes in one liver sample and 20 genes in the second liver sample and 133 genes in the thyroid showed significant relationship between isoform and haplotype of origin.

Campylobacter fetus venerealis host microbiome and genomics predictions for Bovine Genital Campylobacteriosis (BGC) Immunity

Mst Sogra Banu Juli¹, Ala Tabor¹, Ben Hayes¹, Gry Boe-Hansen², Mehrnush Forutan¹

¹Centre of Animal Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

²School of Veterinary Science, University of Queensland, Australia, 4067

BGC is caused by *Campylobacter fetus venerealis* and is the most common venereal disease of cattle transmitted by asymptomatic carrier bulls. BGC can result in reproductive failure and economic losses in cattle by decreasing pregnancy rates, early embryonic death, sporadic abortions, and permanent sterility. It is considered a 'quiet profit take', and easily neglected in a herd. BGC can reduce the gross profit margin of a farm by about 66% during the first year of infection and 36% after disease establishment. However, economic assessments of BGC have been difficult to perform due to the lack of reliable diagnostic methods. It is of particular concern for northern cattle herds in Australia and is categorised as a 'List B' notifiable disease by the OIE. Dysbiosis in vaginal microbiota can change the vaginal ecosystem and increase susceptibility to reproductive diseases including BGC. Additionally, BGC susceptibility, like other polygenic diseases, can be determined by Single Nucleotide Polymorphisms (SNPs) in genes that are involved in the immune response to infection. Those SNPs can be identified through Genome-Wide Association Studies (GWAS). Here we will analyse the association of bovine genetics with BGC susceptibility/resistance to find genetic markers that can be used in breeding programs to select for resistant genotypes. Also, we will investigate the composition of vaginal microbiota to identify BGC immune biomarkers which can be used for assay development. The outcomes of this study will provide the Australian beef industry with knowledge and tools required to reduce the economic losses associated with infectious reproductive failure.

Using the boring trait to target the expensive trait: A case study in bovine fertility

Babatunde Olasege¹, Laercio R. Porto-Neto², Ben J. Hayes³, Marina R. S. Fortes¹

¹*School of Chemistry and Molecular Biosciences, The University of Queensland, Saint Lucia Campus, Brisbane, QLD, 4072, Australia*

²*Commonwealth Scientific and Industrial Research Organization, Agriculture & Food, St. Lucia, Brisbane, QLD 4067, Australia*

³*Centre of Animal Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067*

The key phenotypes related to female fertility in beef cattle are often difficult-or expensive to measure. However, some traits in male are easy to measure and have been shown as a good predictor of female fertility. So far, genomic regions driving these correlations are still unknown. The objective of this study is: 1) identify set of single nucleotide polymorphisms (SNPs) driving the genomic correlation between scrotal circumference (SC, termed 'boring trait') and age at puberty (AGECL, termed 'expensive trait'), and 2) investigate how accurately the boring trait can predict expensive trait using the driver SNPs set identified and then compare the accuracy with the high-density SNP array using GBLUP and bayesR. We used phenotype consisting of 1022 (training [TR]: 818; Validation [Val]: 204) animals for SC and 996 (TR: 786; Validation Val: 200) animals for AGECL in Brahman population genotyped with high-density chip. Using a correlation of 500-SNPs effects in a 100-SNPs sliding-window based methodology in the TR dataset for the two traits, genomic regions driving correlation between SC and AGECL were identified. Further, no difference was observed between the accuracy of prediction of AGECL from SC in GBLUP (0.23) and bayesR (0.23) using the driver set of SNPs discovered. However, prediction accuracy was higher using the driver set of SNPs (0.23) compared to using the high-density SNPs (0.13). Conclusively, expensive traits can be predicted from a boring trait with better accuracy using pre-selected SNPs with either GBLUP or bayesR. Notwithstanding, larger sample-size will be required to improve prediction accuracy.



Building Blocks for Designer Ideotypes: Dissecting Plant Architecture Genes

Yasmine Lam¹, Karen Massel¹, Zachary Aldiss²

¹Centre of Crop Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

²School of Chemistry and Molecular Biosciences, The University of Queensland, Saint Lucia Campus, Brisbane, QLD, 4072, Australia

Improving sustainable food and feed relies on many factors including the ability to maximize yields in the face of stress due to climate change. Plant architecture is highly plastic in nature and allows plants to be well adapted to a variety of environments, providing alternate strategies to efficiently capture resources from the surrounding environment. However, given the rate of climate change and increasing population, crop systems are facing major challenges. The studies showcased hereinafter, using two cereal crops of high importance, sorghum and barley, utilize cutting edge gene editing technology to target key plant architecture regulating genes to 1) better understand the function of developmental genes, and 2) with that knowledge be able to build designer ideotypes for a variety of environmental scenarios. CRISPR/Cas9 knockouts targeting a gene involved in auxin transport altered root angles significantly, while a gene involved in floral determinacy was targeted to alter canopy structures. These outcomes demonstrate how gene editing technologies can be useful in generating novel genetic variants, and paired with breeding and predictive modelling, can effectively help crops adapt to the rapidly changing climate.

A toolkit to rapidly modify root systems through single plant selection

Charlotte Rambla¹, Samir Alahmad¹, Lee Hickey¹

¹Centre of Crop Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

The incorporation of root traits into elite germplasm is typically a slow process. Thus, innovative approaches are required to accelerate research and pre-breeding pipelines targeting root traits to improve yield stability in different environments and soil types. Marker-assisted selection (MAS) can help to speed up the process by selecting key genes or quantitative trait loci (QTL) associated with root traits. However, this approach is limited due to the complex genetic control of root traits and the limited number of well-characterised large effect QTL that can be targeted. Coupling MAS with phenotyping could increase the reliability of selection. Here we present a useful framework to modify root traits in elite germplasm. In this wheat exemplar, a single plant selection (SPS) approach combined three main elements: phenotypic selection (in this case for seminal root angle); MAS using KASP markers (targeting root biomass); and speed breeding to accelerate each cycle. To demonstrate the effectiveness of the approach, we applied SPS in a backcrossing program for the purpose of rapidly modifying the root system of elite bread wheat line Borlaug100. Within 18 months, BC2F4:F5 introgression lines were developed and displayed a full range of root configurations, while retaining similar above-ground traits to the recurrent parent. The SPS approach enables researchers and plant breeders to rapidly manipulate root traits of future crop varieties, which could help improve productivity in the face of increasing environmental fluctuations. The newly developed elite wheat lines with modified root traits provide valuable materials to study the value of different root systems to support yield in different environments and soil types.



Plant and livestock disease prevention and management

Capsicum gene expression, symptom development and virus accumulation in response to capsicum chlorosis virus infection in local and systemic infection

Fernanda Y Borges Naito¹, Shirani M K Widana Gamage², Neena Mitter¹, Ralf G Dietzgen¹

¹Centre of Horticultural Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

²University of Ruhuna Matara

Chalcone synthase (CHS), cytochrome P450 (CYP), Tetraspanin 8-like (TSP8), Thionin-like (Thio) and WRKY40 transcription factor (WRKY40) were identified as being differentially expressed in capsicum during capsicum chlorosis virus (CaCV) infection. Studying symptom development and the expression of those genes over time during CaCV infection in susceptible capsicum and correlating their expression with virus accumulation is an important step to create strategies for CaCV management. Relative gene expression and virus titre in Yolo Wonder and Warlock cultivars were determined by real-time PCR at 3, 7 and 12 days post inoculation, and symptoms were visually scored. In Yolo Wonder, local symptom development was faster and virus accumulation was higher in the early stages of infection, but virus titres were similar once systemic infection was established for both cultivars. A strong positive temporal correlation was identified between symptom development and virus titre. CYP and Thio expressions are also correlated with CaCV titre over time in Yolo Wonder and similarly expression of TSP8, Thio and WRKY40 in Warlock. In terms of functionality, CHS expression may be linked to symptom development, CYP appears to be important in early antiviral defence and TSP8 may be involved in virus movement. The current investigation provides useful information on potential gene targets for novel strategies to control CaCV infection.

Accelerating the development of animals with superior phenotypes using organoids and gene editing

Tatiana Briody¹, Tim Mahony¹

¹Centre of Animal Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Increasing global population and diminishing resources has put considerable pressure on the agricultural sector to be more efficient. However, pests and diseases stand in the way of such a goal, accounting for approximately \$3billion in annual losses for the Australian livestock industry. Despite recent advances in gene editing technologies, developing lines of cattle resilient to infections such as bovine viral diarrhoea virus and bovine herpesvirus 1 is costly, time consuming, and arguable unethical. Organoids offer a potential solution to these issues. Organoids are cell-based structures grown in the laboratory that can be used to study complex phenotypes and biological interactions. My project will investigate developing lung organoids from cattle skin samples to model the impact of gene editing aimed at increasing resistance to respiratory pathogens. I will conduct comparative analyses of integrating and non-integrating approaches for the reprogramming of bovine fibroblasts to induced pluripotent stem cells (iPSCs). I aim to develop a robust protocol for the identification of pluripotency and maintenance of bovine iPSC stocks, which can be used for the development of multiple organoid types. Following CRISPR-mediated gene editing of iPSCs and subsequent differentiation into lung organoids, I will assess cellular responses to infection and quantify virus resistance. In doing so, I can evaluate the effectiveness of animal-specific genetic changes to infection in a much quicker time and in an ethical manner. The potential implication of this research is to demonstrate the benefits of gene editing to develop animals with superior phenotypes, ensuring the long-term productivity of Australian livestock.

Effects of elevated temperature on capsicum chlorosis virus resistance in capsicum

Wei-An Tsai¹, Jonathan Peters¹, Neena Mitter¹, Ralf G. Dietzgen¹

¹Centre of Horticultural Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Capsicum chlorosis virus (CaCV) is an important pathogen affecting capsicum plants in the major commercial production areas in Queensland, Australia. Although CaCV-resistant capsicum breeding lines were established several years ago, no studies on how abiotic stresses affect stability of resistance to CaCV have been conducted. Since climate change presents emerging abiotic threats to future crop production, the present study was undertaken to investigate how elevated temperatures affect CaCV-resistant capsicum's response to CaCV. We found that defence responses of CaCV-resistant plants were compromised at high temperature (HT, 35oC) in 8/14 plants. Systemic necrotic spots were seen at 10 dpi in these resistant CaCV-infected plants grown at HT, while systemic leaves of all plants grown at ambient temperature (AT, 25oC) were free of symptoms. To identify miRNAs involved in resistance-breaking at HT, we compared miRNA expression profiles in CaCV- or buffer-inoculated plants grown at HT or AT. A total of 105 known and 83 novel miRNAs were identified from 12 small RNA libraries. The levels of Can-miR408a-3p, Can-miR397-5p, and Can-miR398b-3p were suppressed by CaCV infection at AT, while they were induced by CaCV infection at HT. Gene ontology enrichment analysis of miRNA target genes indicated that the lignin metabolic pathway, regulated by Can-miR408a-3p and Can-miR397-5p, was involved in the temperature-mediated resistance-breaking response. This study provides first insights into the involvement of miRNAs that may be critical to temperature-sensitive tospovirus resistance.

Cloning and expression of conserved *Mycoplasma bovis* antigenic sequences into the BoHV-1 vaccine vector - Development of a BoHV-M. bovis bivalent vaccine.

Yastika Banerjee¹, Timothy Mahony¹, Karl Robinson²

¹Centre of Animal Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

²Centre of Horticultural Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Bovine respiratory disease (BRD) is a pervasive issue of feedlot cattle in Australia causing acute respiratory distress and significant economic losses; it is a multifactorial disorder caused by several stressors and pathogens. My project aims to genetically engineer Bovine Herpesvirus as a bivalent live viral vaccine against BRD using cutting edge CRISPR/Cas9 technology. This vaccine will target both BoHV-1 and the bacterium *Mycoplasma bovis* in a single vaccine. This will be achieved by 1) cloning and expression of codon optimised Variable surface protein A (VspA) of *M. bovis* as a Green Fluorescent Protein (GFP) fusion protein in RK-13 cells; 2) insertion of the GFP-VspA expression cassette into the BoHV-1 viral vector backbone using CRISPR/Cas9 in RK-13 cells; 3) Generation of non-GFP fused BoHV-1-VspA recombinants using CRISPR/Cas9 followed by 4) characterisation of the growth performance and expression of BoHV-1-VspA recombinants in cell culture. The expression of a bacterial antigen from a viral backbone able to illicit and immune response will be a significant development in combating BRD in Australian feedlot cattle.

BioClay: A non-GM RNAi strategy to protect plants from pests and pathogens

Srinath Balasubramanian¹

¹Centre of Horticultural Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Crop production is one of the key elements to sustain our life on earth. Saying so, great attention is needed to protect crops in a sustainable way. Agricultural chemicals have so far combated pests but there has been a trade-off to human and environmental health, while technologies such as genetically modified (GM) crops which overcome issues of chemical residues failed to receive consumer acceptance and remain to be a controversial topic. Consequently, to tip the balance between health and consumer acceptance, an innovative strategy was devised using RNA silencing machinery in a non-GM approach. RNA interference (RNAi) technology has made significant progress in crop protection because of its high target specificity which allows growers to knock-down pests more precisely than agrochemicals and without any off-target effects. While RNAi technology is available on the market through transgenic crops, non-transgenic alternatives such as topical application of RNA (SIGS - Spray Induced Gene Silencing) are gaining attention due to its feasibility, low cost, and acceptance. When dsRNA/siRNA are topically applied they are internalized by pests either directly or indirectly which results in gene knock-down. To protect topically delivered naked dsRNAs/siRNAs from degradation, tools such as BioClayTM, dsRNA bound on clay particles, are used to ensure its efficiency in the field. BioClay seems to be a promising tool in crop protection addressing many of the issues with current pest control strategies.

Does allergic response to buffalo fly cause skin lesions in cattle?

Muhammad Noman Naseem¹, Muhammad Kamran¹, Ali Raza¹, Constantin Constantinoiu², Conny Turni¹, Mst Sogra Banu Juli¹, Emily Mantilla Valdivieso¹, Michael McGowan³, Rachel Allavena³, Ala Tabor¹ and Peter James¹

¹Centre of Animal Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

²James Cook University, College of Public Health, Medical & Veterinary Sciences, Townsville (4810), Australia.

³The University of Queensland, School of Veterinary Science, Gatton (4343), Australia.

Buffalo flies (BF) are major blood feeding ectoparasite of cattle causing economic and welfare impacts to the Australian herds. Lesions associated with BF infestations are termed as buffalo fly lesions (BFLs), manifested as dermatitis or wounds most commonly near the eye, neck and on the belly. BFLs range in severity from raised, dry, hairless scabbed areas to severe bloody wounds. There is a poor correlation between BF numbers and BFLs development which indicate it might be the individual response to BF, making them susceptible for BFLs. This study investigated the role of individual immune responses in the development of BFLs. Based on the presence and severity of BFLs, 28 Brangus steers were divided into 2 groups: lesion susceptible (LS) and lesion resistant (LR). Each steer was injected intradermally with different concentrations of BF (100, 10 $\hat{1}\frac{1}{4}$ g), *Onchocerca gibsoni* (100, 10 $\hat{1}\frac{1}{4}$ g) and house fly (100, 10 $\hat{1}\frac{1}{4}$ g) antigens. Differences in skin response, gross and histological changes measured in the 2 groups are reported. LS group had significantly stronger allergic response to BF antigens as compared to LR. LS steers started developing skin lesion after 48-72 hours at the BF antigen injection sites. Histology of skin sections revealed complete destruction of superficial skin layer, hair follicles and sweat glands in LS group. All these changes indicate type-I allergic reaction specific to buffalo fly's antigen, in LS steers. Clarification of the pathogenesis of BFLs will help to identify the factors that underlie differences in predisposition to lesion development in susceptible and resistant cattle.

Geographic expansion and transmission of banana Blood disease

Jane Ray¹, Subandiyah S., Rincon-Florez V., Carvalhais L.C., Prakoso A.B., O'Dwyer C.,
Drenth A.

¹Centre of Horticultural Science, Queensland Alliance for Agriculture and Food Innovation,
University of Queensland, Australia, 4067

Blood disease caused by *Ralstonia syzygii* subsp. *celebesensis* is a bacterial wilt of banana. The disease causes significant crop losses in Indonesia and Malaysia and is a priority pathogen absent from Australia. The leaves of infected banana plants wilt and discolour, and the fruits rot, making them inedible. Our research aimed to determine; (i) the past and current distribution of blood disease, (ii) if infection occurs through male and/or female parts of the banana inflorescence, (iii) if local disease dispersal occurs through mechanical transmission by insects or human activities, and (iv) the role of contaminated planting material in long-distance dispersal. Our survey results demonstrate that Blood disease has significantly expanded its geographic range in the last 20 years, such that it is an emerging threat to global banana production. We show that infection occurs through the male and female parts of the banana inflorescence. That local dispersal is predominantly through mechanical transmission of the bacterium by insects, birds, bats and tools from diseased to healthy banana plants and that long-distance dispersal is through the movement of contaminated planting materials. Our findings provide insight into the biology and epidemiology of banana Blood disease to underpin the development of improved disease management and eradication strategies to manage the disease where it occurs and prevent spread to areas still disease-free.



Shaping future food markets



Innovative trademarks and designations can represent the ethical values underlying QAAFI's research with Indigenous people and industry partners

Christopher Sauer¹

¹Centre of Nutrition and Food Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

QAAFI research practices include embracing both our industry and Indigenous partners with ethical values that support the development of a responsible, sustainable and inclusive Australian foods industry. These values overlap with a new generation of trademarks and designations that expand upon categories of environmental and economic concerns. For example, the UNESCO Creative Cities designation represents that a city's activities align with sustainable development goals such as developing social capital and community cohesion. Another example are Responsible Innovation labels that communicate how organizations and researchers uphold values such as inclusive deliberation, reflexivity and responsiveness. Can trademarks and designations communicate QAAFI's research practices and values to the public?



Future flavours from the past: sensory profiles of summer fruit from an Arnhem Land Aboriginal community

Selina Fyfe¹, Yasmina Sultanbawa¹, Horst Joachim Schirra¹, Heather E Smyth¹

¹Centre of Nutrition and Food Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Native fruit of Australia have long been eaten by Aboriginal communities and have the possibility of being used as unique and interesting flavours in future products. The green plum (*Buchanania obovata*), red bush apple (*Syzygium suborbiculare*) and wild peach (*Terminalia carpentariae*) are all harvested and eaten by the remote East Arnhem communities in Australia. The purpose of this study was to provide sensory descriptions for the three fruit and a better understanding of ways this fruit could be used in food products. Free choice profiling was performed on each sample by 15 panellists who tested each sample twice over two days. Samples were assessed as whole or cut up pieces of fruit, as puree and as an ingredient (freeze-dried powder) in both semolina and yoghurt. Reliable sensory descriptions were obtained for each sample format. The green plum and red bush apple had highly desirable attributes and unique profiles. The green plum has flavours of sweet, tart, stewed apple and citrus in each form. The red bush apple has flavour notes of sour, sweet, spiced tea, raspberry, apple, floral and herbaceous. The results of this study and product descriptions will be useful to sustain employment opportunities for remote Aboriginal communities through food industry uptake of these unique fruits in novel future food products.

User perspectives of Australian Native Foods

Clare Wijngaarden¹, Heather Smyth¹, Kamalesh Adhikari¹

¹Centre of Nutrition and Food Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Customers and end users are critical players in any FMCG value chain. It is important to understand what drives interest in a product and category in terms of how and which products are used, sought out, and ultimately valued by different groups of consumers. For the native food industry this information (market insight) can help stakeholders at all stages of the value chain: from primary growers (e.g. guiding on which foods to grow and how to grow them), for intermediaries (e.g. identifying opportunities on how to value add, process and pack foods), to product developers (e.g. what sort of products to incorporate them in) and marketers (e.g. what communication and education is required to best promote and appeal to their target audiences). Overall, market insight will help identify any key barriers or pain points associated with product use, as well as highlighting opportunities and direction for market initiatives moving forward. The aim of the present research is to collect a snapshot of general market interest and usage relating to Australian native foods. Results reported here will help inform future studies relating to both general and food specific research around determination of demand and value for Uniquely Australian Foods.



Anthocyanin-rich novel purple sweetcorn for healthy life and food security

Apurba Ray¹

¹Centre of Crop Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067

Purple sweetcorn is not found anywhere in the world because of an extremely close genetic linkage between a non-functional anthocyanin biosynthesis gene, anthocyaninless-1(a1) and the super sweet gene, shrunken-2(sh2). As a result, purple corn is almost always starchy (non-sweet), and sweetcorn is always non-purple (white or yellow). The anthocyanin biosynthesis is also regulated by Myb and bHLH transcription factor (TF) genes. To develop purple super sweet sweetcorn it is essential to break down the tight genetic linkage between a1 and sh2 genes. Furthermore, biochemical as well as molecular experiments are also essential to know the nutrients as well as structural and regulatory genes. To do this, five consecutive field experiments were performed, and heterozygous purple sweetcorn is developed in the third field experiment. In the fifth field experiment, a homozygous purple sweetcorn line is developed. By biochemical analysis it is observed that the developed purple sweetcorn line produced similar amount of anthocyanin as in its purple maize parent, and sugar as in its white sweetcorn parent. In addition, it is found that at least one copy of the TF is required for the biosynthesis of anthocyanin, which is confirmed by SSR molecular marker assay. The developed novel purple sweetcorn has the highest amount of anthocyanin in comparison to other purple crops and can be used as a healthy fruit to ensure global food security.



Characterization of synbiotics and their impacts on human gastrointestinal fermentation

Hai Tran¹, Mark Turner¹, Deirdre Mikkelsen¹, Barbara Williams¹, Nidhi Bansal²

¹*School of Agriculture and Food Science, The University of Queensland, 4067*

²*Centre of Nutrition and Food Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067*

A synbiotic is a combined product of prebiotics and probiotics with potentially synergistic benefits. Unfortunately, the selection of synbiotic components has not always been a scientifically sound process. This study aimed to provide insights into interactions between probiotics and prebiotics, which might ultimately contribute to the beneficial modulation of the human gut microbiota. The behaviours of selected prebiotics (inulin, oligofructose, and lactulose) and probiotics (*Lactobacillus casei* CRL431 [CRL431], and *Bifidobacterium animalis* subsp. *lactis* BB12 [BB12]) were investigated. A carbohydrate-free medium was used to investigate the ability of probiotics to utilize prebiotics. In addition, the impacts of these synbiotics on human faecal fermentations were also evaluated, including levels of post-fermentation products such as short-chain fatty acids and ammonia. All three prebiotics were utilized by CRL431 and BB12. As the major energy source in these human faecal batch fermentations, the prebiotics had a major influence on the gas profiles and post-fermentation products. However, the presence of probiotics only had minor impacts on these fermentations. Analysis of the faecal microbial composition is still pending. This work demonstrates that prebiotics have a potent influence over gut microbe activity while the addition of probiotics (individually or in a synbiotic formula), which we show can metabolize these prebiotics, have minimal effects.

Optimising trehalulose level in Australian native stingless bee honey

Jiali Zhang¹, Mary Fletcher¹, Natasha Hungerford¹, Tobias Smith², Hans Yates³

¹*Centre of Food Science, Queensland Alliance for Agriculture and Food Innovation, University of Queensland, Australia, 4067*

²*School of Biological Science, The University of Queensland, 4067*

³*Queensland Health*

The stingless bee honey has been regarded as functional food of high value. Indigenous people use stingless bee honey treating wounds, burns or respiratory illness. In more recent studies, it has been proved that stingless bee honey has excellent therapeutic potential as an antidiabetic and antimicrobial agent. Our research team has identified the therapeutic component in stingless bee honey to be trehalulose, a rare sucrose isomer. The trehalulose level in stingless bee honey samples from Australia, Malaysia and Brazil is around 13-44 g per 100 g honey. The origin of this rare disaccharide has been discovered in this research. By experimentally feeding various sugar solutions to live stingless bees, we observed the complete conversion from sucrose to trehalulose. The experimentally produced honey contents trehalulose (64-72%), erlose (18-23%), fructose (9-12 %) and minor glucose. Remarkably, feeding 1:1 glucose/fructose solution did not result in trehalulose formation, which means bees cannot create the 1,1 glycosidic linkage of trehalulose directly. Therefore, stingless bee with natural access to floral nectar with high sucrose content will produce higher trehalulose level in honey. Feeding bees with commercial refined sugars has widely used to prevent starvation of the hive in winter or to artificially encourage foraging and breeding. However, the indirect adulteration of stingless bee honey will result lacking in key natural phytochemical. The "fake" honey can be readily distinguished via isotope ratio mass spectrometry.