







Plant pollination networks DNA barcoding applications


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 **THE UNIVERSITY
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 **State Herbarium
of South Australia**

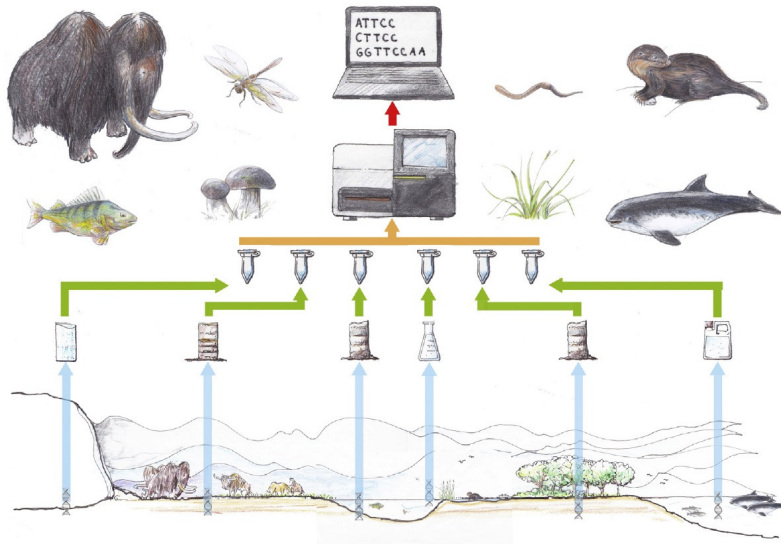
Advanced DNA, Identification and Forensic Facility ADIFF

- Illegal wildlife trade
 - Species identification
 - Timber Tracking
- Human identification
 - Crime scene investigation
 - Degraded remains
 - Forensic odontology
- Environmental Analysis
 - Soil Analysis
 - Diet identification
 - Conservation management
 - Human health



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 **Flinders
UNIVERSITY**
 **Government
of South Australia**
 Department of Environment,
Water and Natural Resources
 **Australian
museum**  **South Australian
Museum**
 **agrf**
 Empowering Australian Genomics

DNA – To monitor environmental, agricultural and public health outcomes



(Thomsen & Willerslev 2015)

Bioinformatics and analysis

Next Generation Sequencing

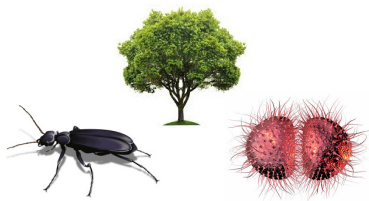
DNA extraction &
library preparation

Environmental samples

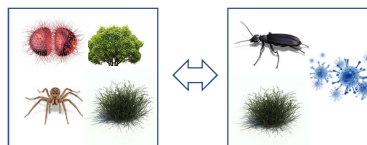
- Soil
- Leaves
- Pollen
- Air
- Human skin & nose

What we may want to know?

Who is present in a sample?



How do samples compare?

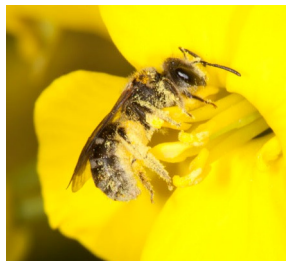


What is the role of these taxa?





Secure crop pollination through revegetation



Australian Government
Department of Agriculture
and Water Resources



AgriFutures™
Securing
Pollination

Apple, Canola, Almonds, Lucerne

- Pollinator assessment

- What bees pollinate crops? Honey-Native
- What native plants support them?
- When should they flower? Competition?

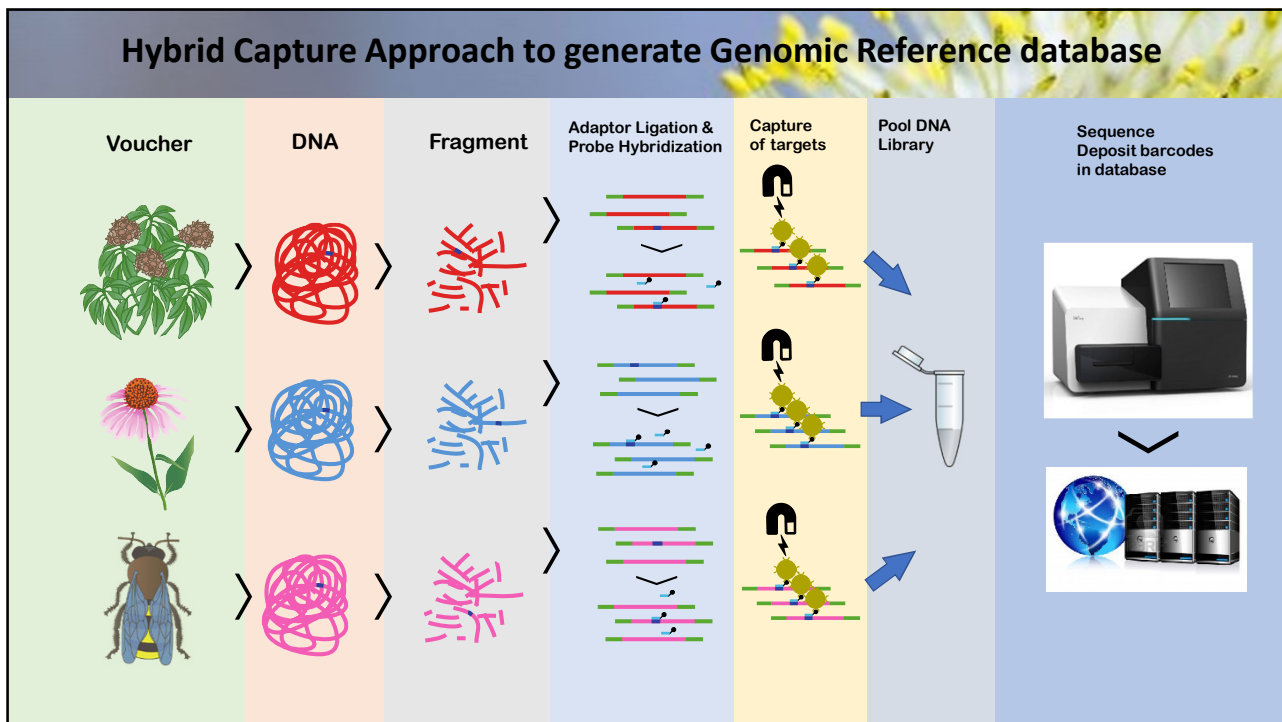
- DNA Barcoding

- Develop barcodes to identify pollen and bees

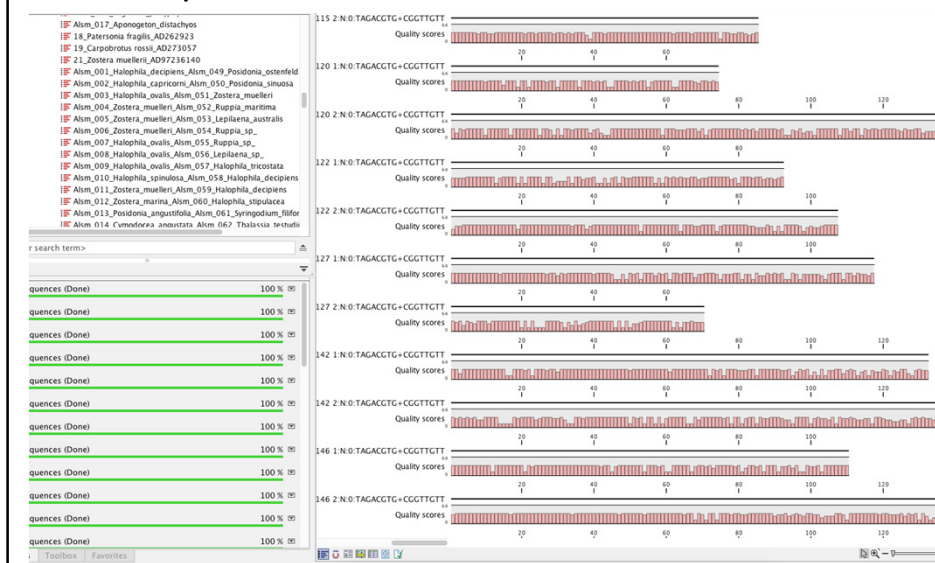
- DNA Metabarcoding

- Harvest pollen to identify host plants and ID bees





Dramatic increase in data we can gain from specimens...



Last run:
 200 million sequences
 30.11 billion bases
 80 specimens
 380 million average

For same cost (~\$100 p sample):

- We could do 2-3000 bp with Sanger sequencing
- Now we get:
 - 10s of thousands of bp
 - 70% of Chloroplast
 - All traditional barcoding loci
 - Exon and Intron data for dozens of nuclear UCE's (Ultra Conserved Elements)

Taxon 65 (5) • October 2016: 1081–1092
Tartaric acid
amc

Universal target enrichment baits for anthozoan
Hart & al. • Targeted enrichment of old DNA from herbarium specimens to long-

METHODS AND TECHNIQUES

Retrieval of hundreds of nuclear loci from herbarium specimens

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DNA for Agilent's hybrid capture-based targeted massively parallel sequencing

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MyBait probe design

- Used available transcriptomic and genomic resources
- Mapped *Eucalyptus leucoxylon* transcriptome sequence to the *Arabidopsis thaliana* genome
- Genes with >85% mapping similarity
- Select only single copy loci in reference genomes (cf. 60 spp on Phytozome)
 - 760 genes found
 - 30 nuclear gene regions selected for first trial with exons and intron regions in '~200 bp vicinity'



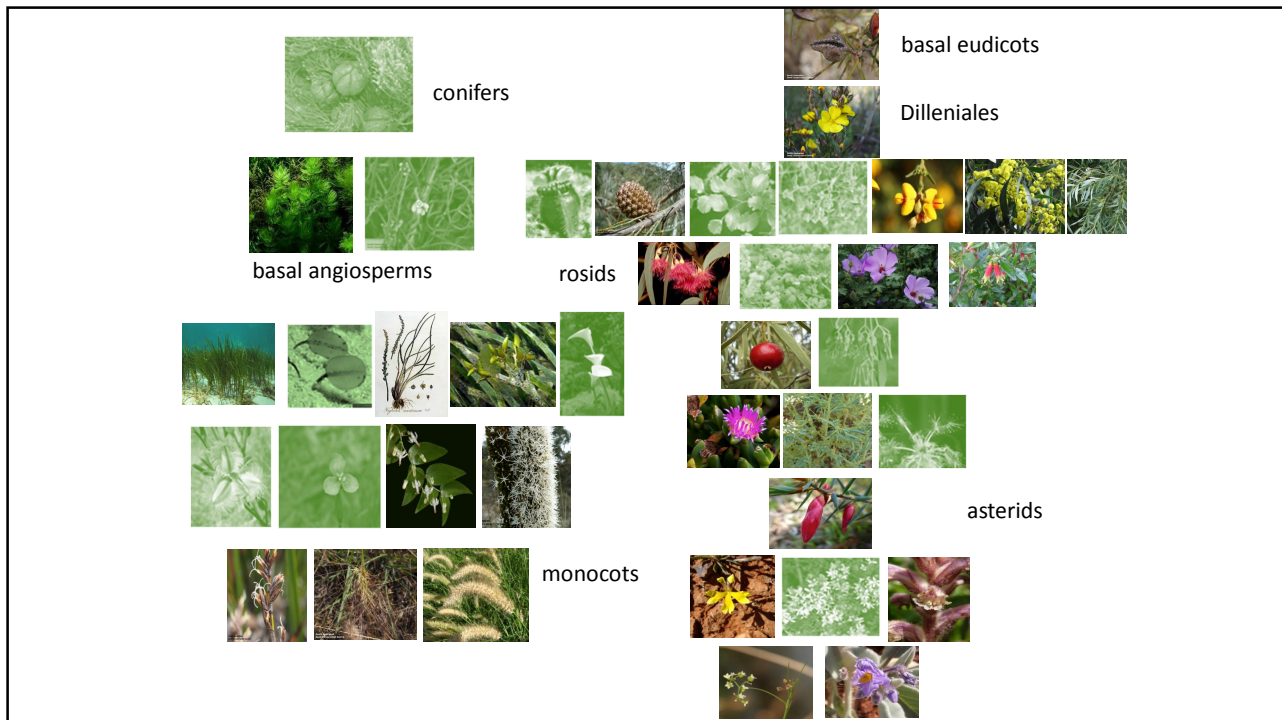
Eucalyptus leucoxylon transcriptome



85% & 1x

Arabidopsis thaliana genome



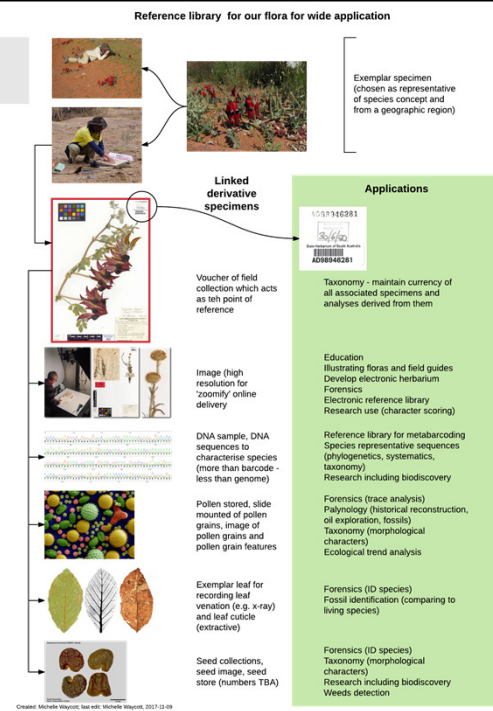


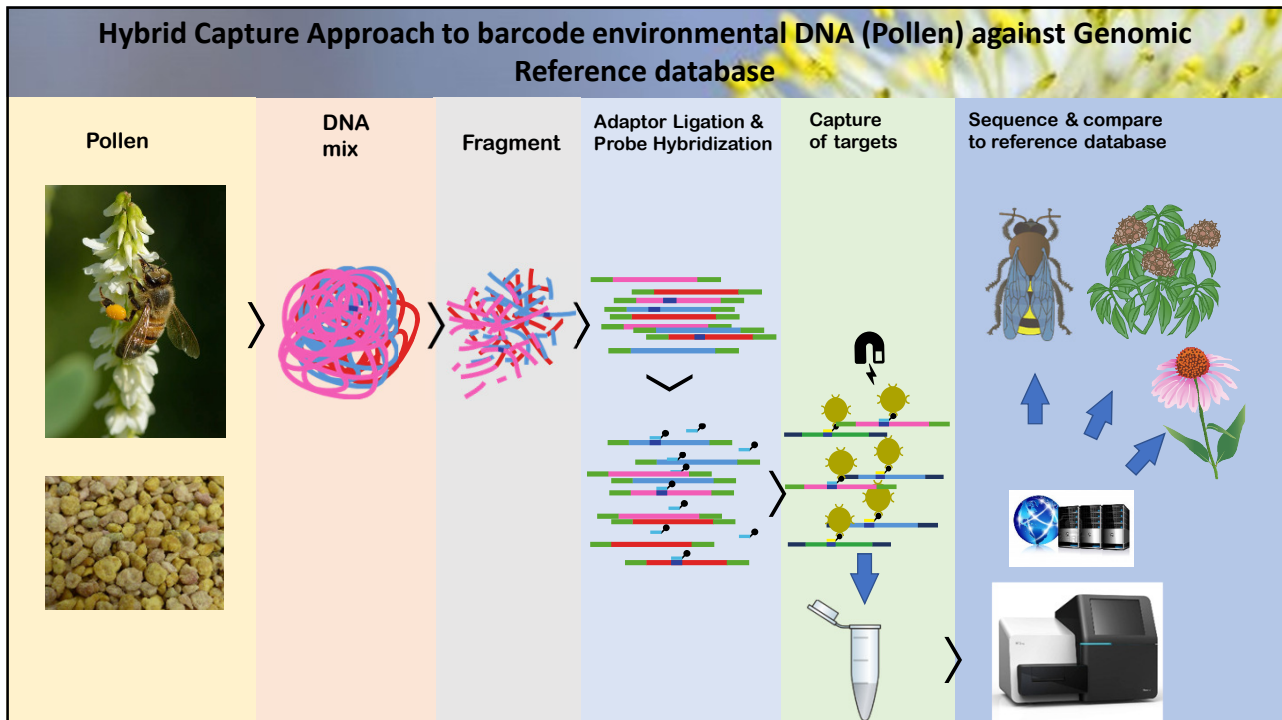
Plant Sampling Process

State Herbarium Exemplar Project

- Vouchers and photos from field
- High resolution image
- Pollen images
- Leaf venation
- Seed collection (image and storage)
- DNA sample > DNA sequence
 - Develop bait based DNA reference library toolkit
 - Currently in final stages of pilot testing

Allow use of wider range of vouchers including older specimen





Apply to examine pollination networks in and around crops



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Australian Government
Department of Agriculture and Water Resources

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AgriFutures™ Securing Pollination

hort frontiers Strategic partnership initiative | **POLLINATION FUND**

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Regional Landcare Facilitator

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Natural Resources Northern and Yorke

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