Bad Karma

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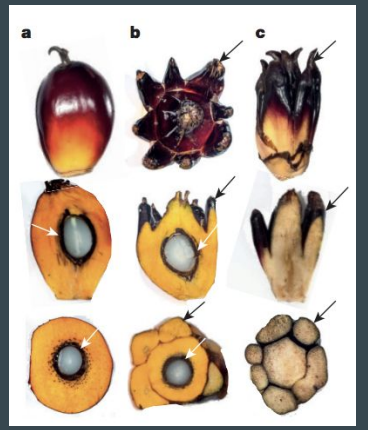
Genomics in Agriculture

- Marker Assisted Breeding
  - Find genetic markers (usually via GWAS)
  - Use them for screening breeding crosses
- Plant Pathology
  - Microbiomes of things of interest i.e. soil
  - Plant pathogens (fungi/bacteria)
- Genomic Selection
  - Basically a giant linear regression that allows one to predict breeding value of crosses
  - Pioneered in cattle
  - Great for crops with long breeding cycles.
- Cryptic or Epigenetic influences
  - Understanding non-mendelian traits.
  - Cool stuff in palm fruit
- Diversity assessments
- Gene editing

Motivation: Plant Breeding


PLANTS INOCULATION ROOM

Clones, but not identical



More on this later

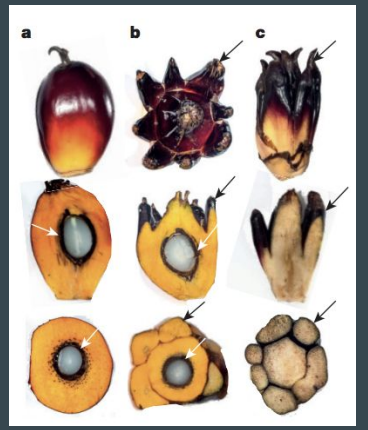
Palm Fruit

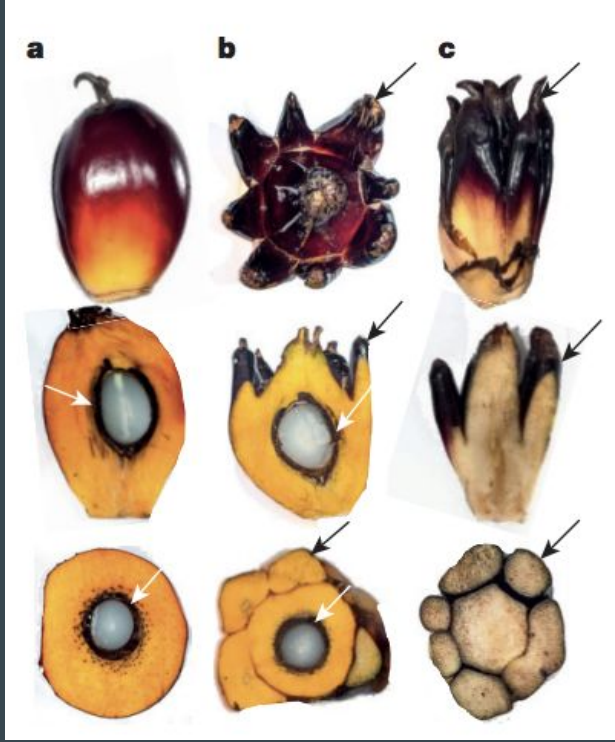
- Very economically important.
- Exceptionally high in saturated fatty acids
- In a surprising amount of products at the grocery store
- Controversial for its role in deforestation in places like Malaysia.
- Challenging for breeding: Long breeding cycles
- https://www.worldwildlife.org/pages/which-everyday-products-contain-palm-oil

Palm Oil Production

https://www.youtube.com/watch?v=Lf-GiulGlqg

Clones, but not identical





“Loss of Karma methylation and of small RNA in tissue culture contributes to the origin of mantled, while restoration in spontaneous revertants accounts for non-Mendelian inheritance” - Ong-Abdullah *et al.*

The Initial Experiment

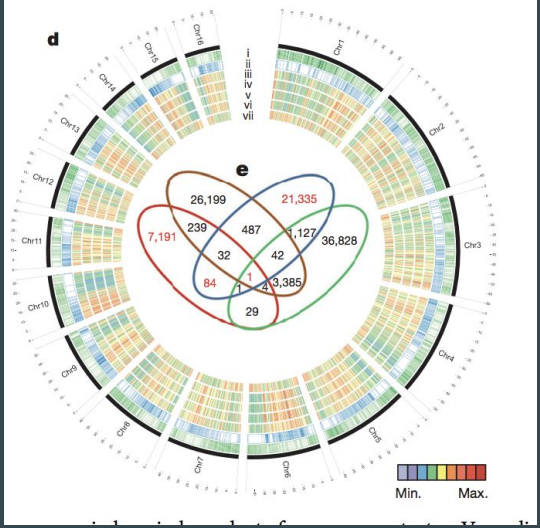
1. Collect methylation data via microarray and bisulfite sequencing for normal and mantled phenotype. (CG, CHG, and CHH, where H=A,T,or C)

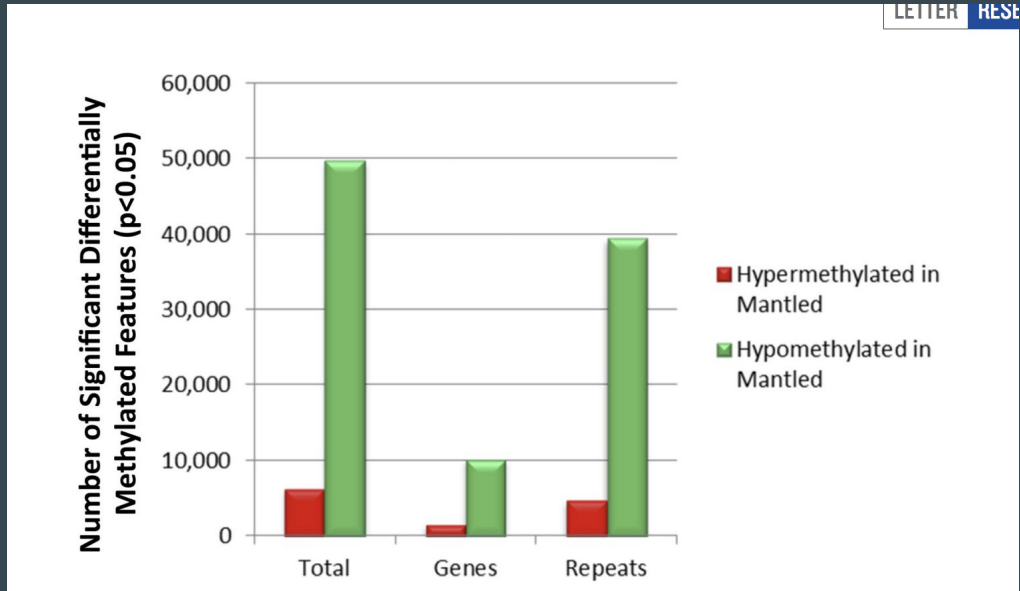
2. EWAS: Similar to GWAS, but the statistical tests is at bins across the genome. Student's t-test.

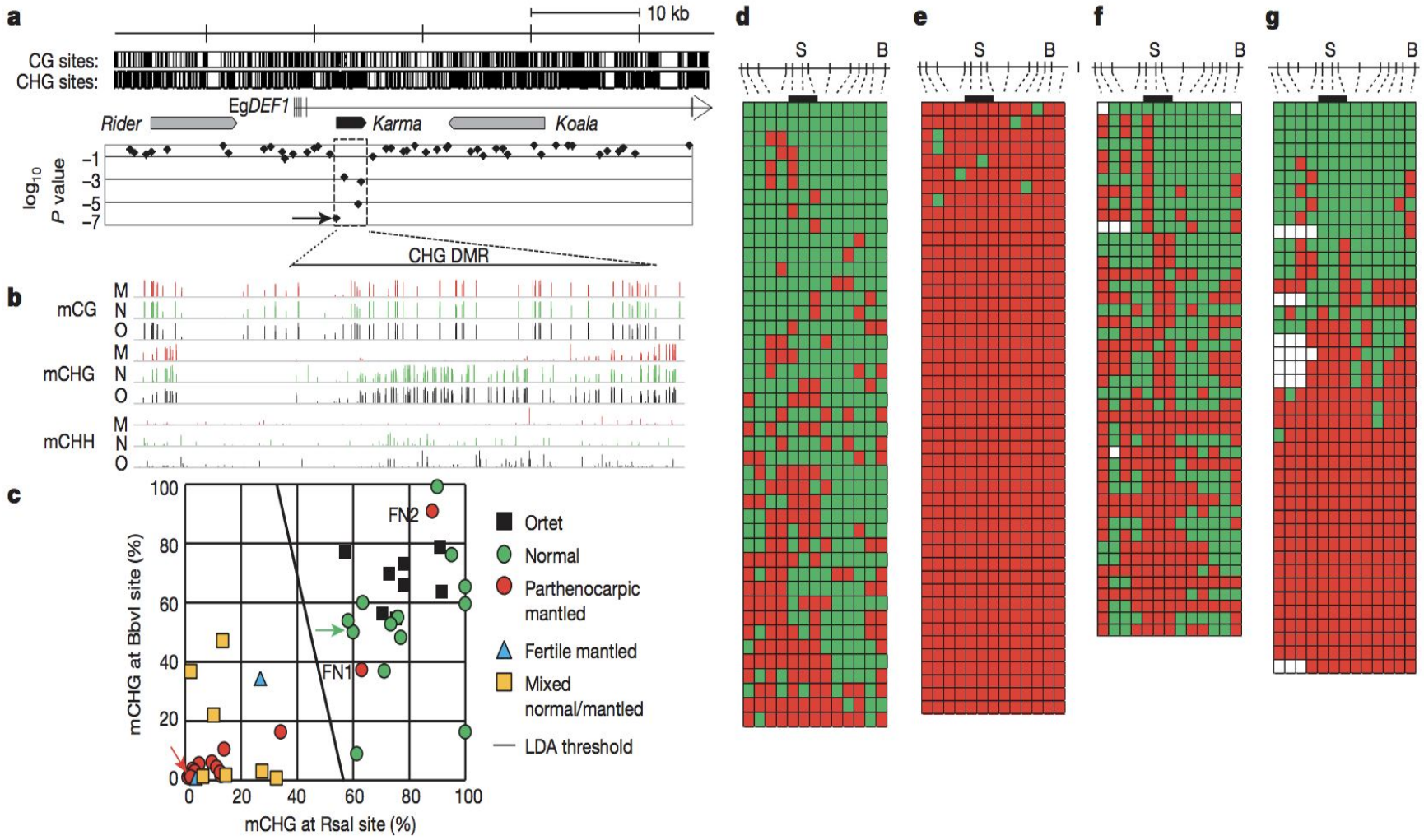
3. Analyze EWAS and find significantly differentially methylated regions

The Initial Experiment

- Breakdown of DMRs
  - Most DMRs showed hypomethylation in mantled phenotypes
  - ~75% of DMRs were in TEs and repeats (foreshadowing).
  - Found 1 DMR across all sampled populations






a
10 kb
CG sites:
CHG sites:
Eg*DEF1*
Rider
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−1
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CHG DMR
b
mCG
mCHG
mCHH
M
N
O
c
mCHG at BbvI site (%)
mCHG at RsaI site (%)
FN1
FN2
Ortet
Normal
Parthenocarpic mantled
Fertile mantled
Mixed normal/mantled
LDA threshold
d
e
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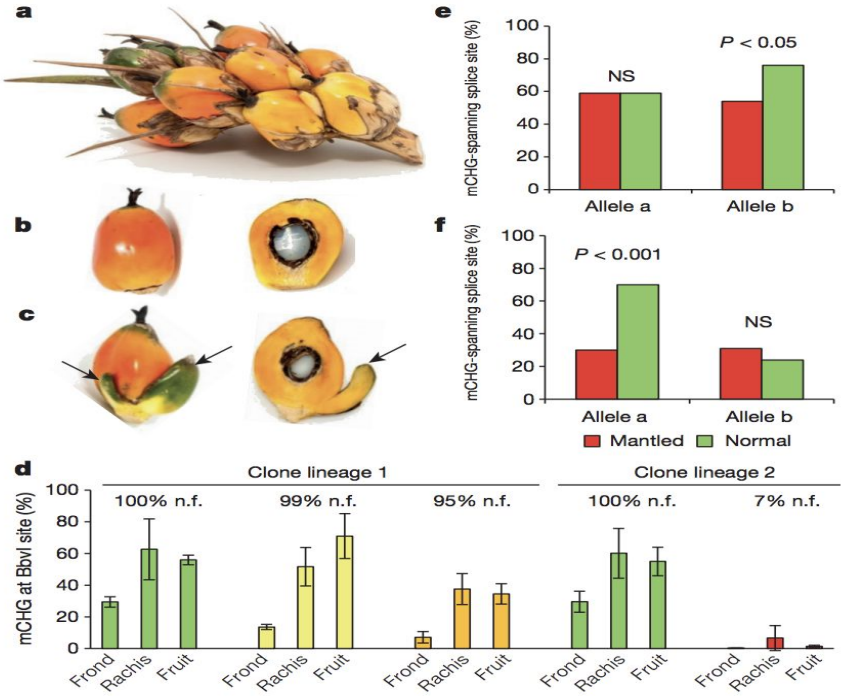
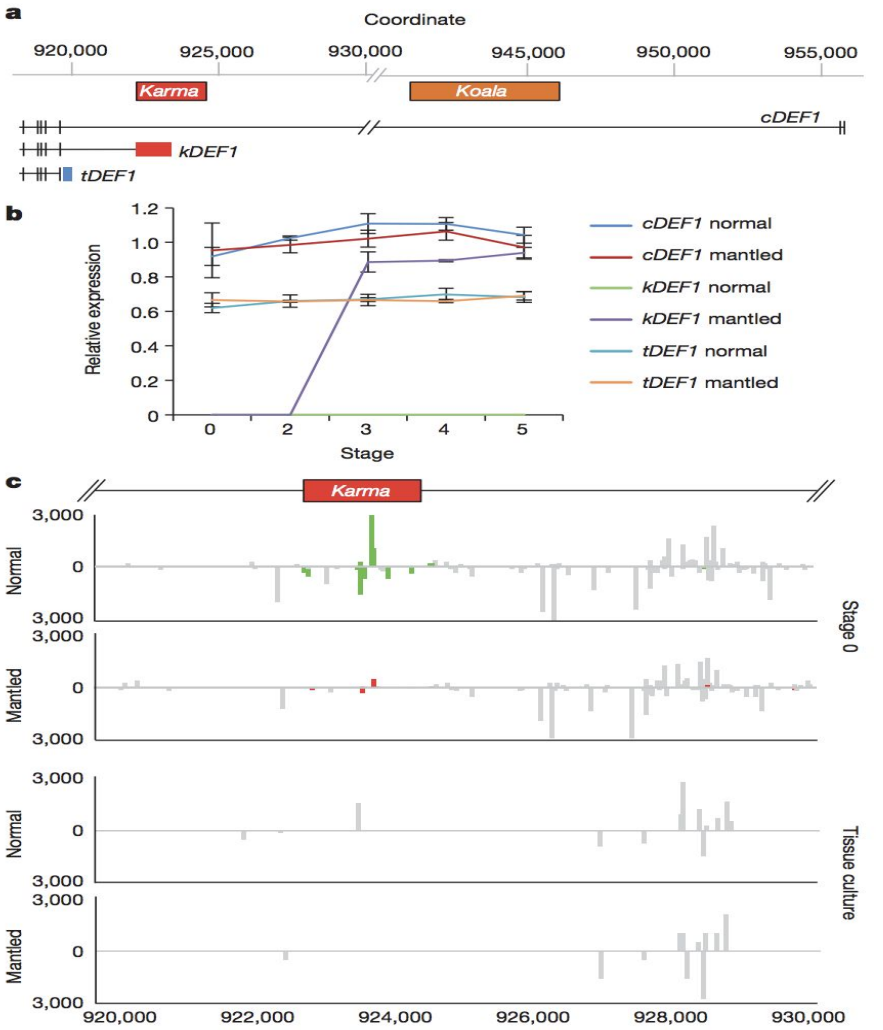


Figure 3 | *Karma* methylation in revertant palms. **a**–**c**, Spikelet from a revertant ramet (**a**) including normal (**b**) and fertile (**c**) mantled fruit with one or two pseudocarpels (arrows). **d**, Density of CHG methylation (percentage mCHG) at the BbvI site (Methods) in ramets yielding 100% normal fruit (n.f.) (green), revertant ramets yielding 99% (yellow) or 95% (orange) normal fruit and a mosaic ramet yielding 7% (red) normal fruit per bunch. Error bars denote s.d. (biological replicates of fronds ($n = 4$), rachis sections ($n = 8$) or fruit ($n = 2$)). **e**, **f**, Percentage mCHG for the three CHG sites found in the unique common microarray feature in normal (green) and subtly mantled (red) fruit from revertant ramets yielding 99% (**e**) or 95% (**f**) normal fruit per bunch (two-tailed Fisher's exact test; NS, not significant). Alleles were analysed separately based on a heterozygous single nucleotide polymorphism (SNP) within the bisulfite sequencing amplicon.



Takeaways

- Methylation, alternative splicing, small RNA, transposable elements all act together to cause this very expensive phenotype.
- Demonstrated a potential to query and diagnose this phenotype with qPCR at an early age.
- The vast majority of similar phenotypes are yet to be understood, and it's easy to understand why (this paper is the exception to the rule, hence cover of Nature).
- Think about how improved genomic resources could really aid with these sorts of tasks.