Genetic transformation of citrus: understanding transformation timelines, progress made and future expectations

Manjul Dutt, Assistant Professor
UF/IFAS Citrus Research and Education Center
Lake Alfred
August 20, 2025

UF IFAS UNIVERSITY of FLORIDA

Take home message

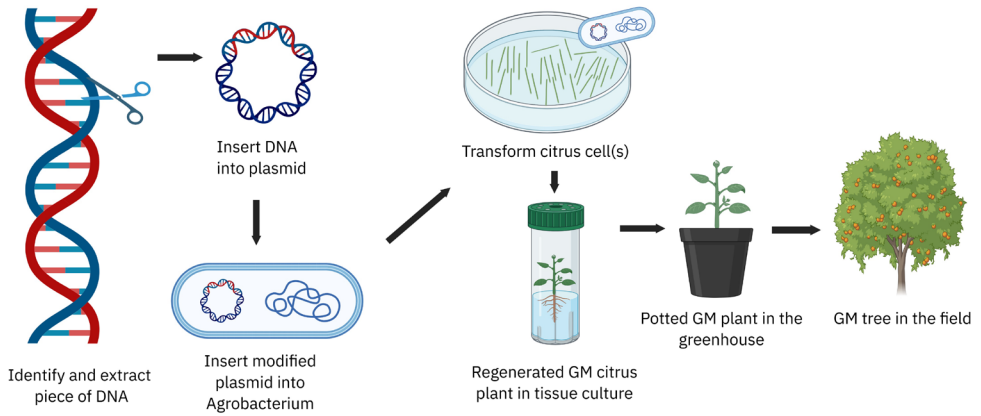
- Genetic transformation can modify the DNA of elite Citrus varieties to enhance HLB tolerance.
- It often takes 10–14 years from discovery to commercial release of a GM plant, due to extensive testing, regulatory review, and field trials.
- UF-CREC Citrus improvement program has several potential candidates that offer promise in the fight against HLB.

What is Genetic Transformation?

Genetic transformation is a process to **modify the DNA of a plant (or any organism)** to give it **new qualities**, like disease resistance, better yield, drought tolerance, or pest resistance.

How does Genetic Transformation work?

- Scientists **identify a useful gene** - for example, a gene that helps citrus survive HLB.
- They **insert that gene into the DNA** of Citrus using one of several methods.
- Citrus **grows with that new ability (HLB tolerance).**



UF IFAS UNIVERSITY of FLORIDA

Understanding Transformation Timelines

Contrary to common misconceptions, the Citrus genetic transformation and plant release process can be time-consuming.

- Year 1 – Developing the DNA constructs and inserting them into the plant's cells. Developing the modified plant in tissue culture.
- Year 2 -3 – Care for the trees in the greenhouse to support proper bud development for propagation. Conduct molecular analysis to identify superior trees.
- Year 3-4 – Propagate trees for replicated trials and evaluations (greenhouse and/or field).

Understanding Transformation Timelines

- Year 4-8 – Evaluate trees in greenhouse and/or field for HLB tolerance.
- Year 5 onwards –
  - Identify superior lines based on initial evaluations and establish larger multi-location field trials.
  - Get germplasm certified through the DPI parent tree program.
  - Initiate paperwork for eventual release.

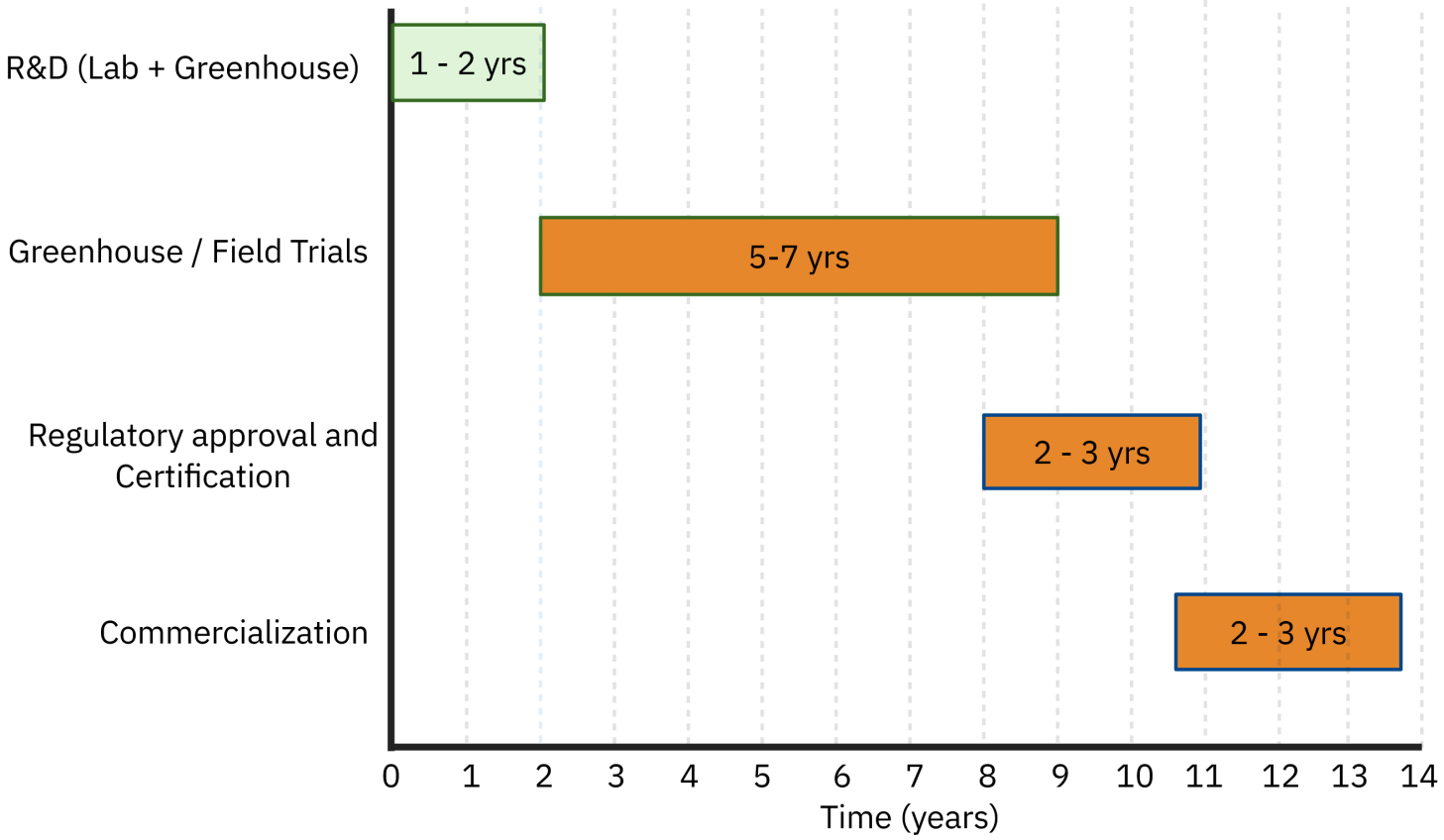
- **Year 6 onwards – the regulatory approval process**

Understanding Transformation Timelines

Cultivar registration & commercialization

- Year 8 onwards
  - Official cultivar registration.
- Planting material multiplication for large-scale nursery propagation.
- Market introduction, distribution, and outreach to growers.

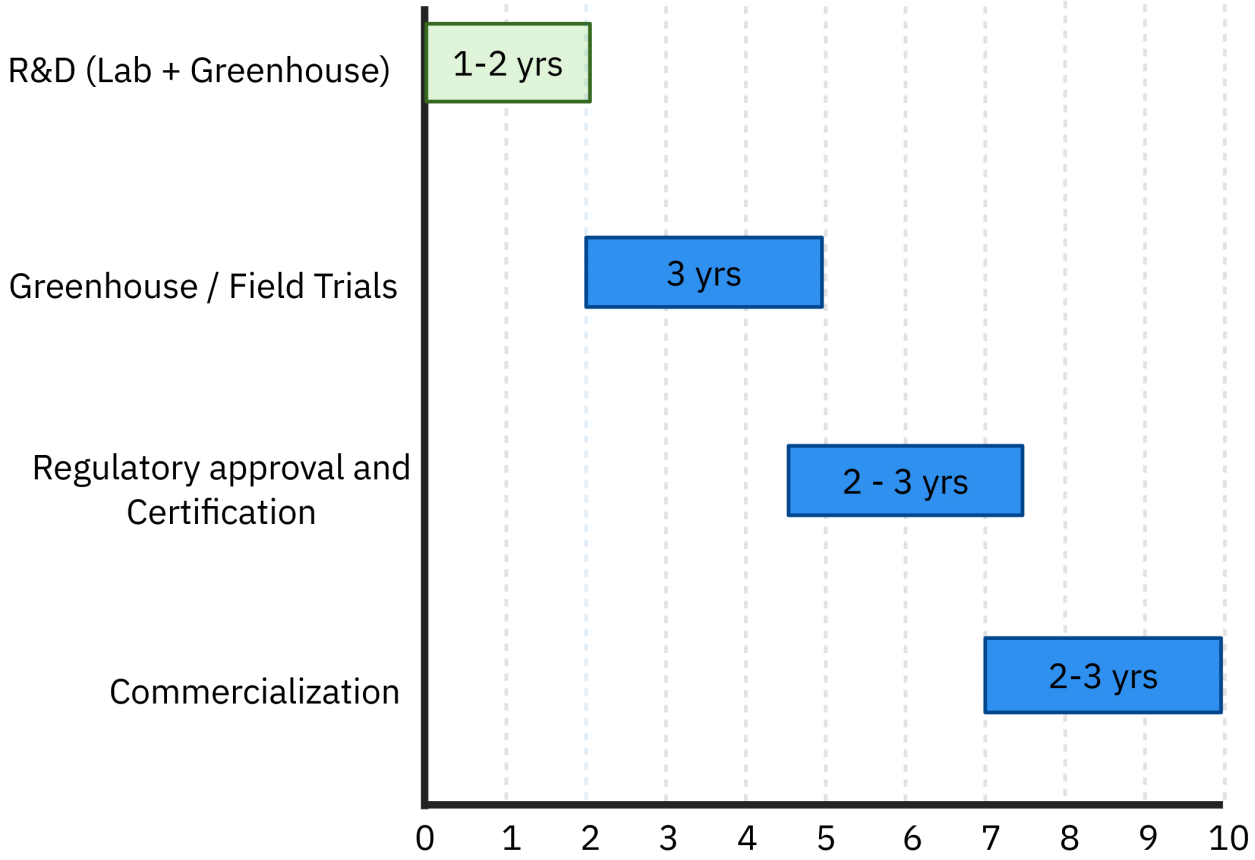
Juvenile tissue derived Citrus GMO timeline



Juvenile period is long, typically 5 to 7 years, before the tree produces flowers and fruit

UF IFAS UNIVERSITY of FLORIDA

Mature tissue derived Citrus GMO timeline



Mature tissue shortens the time it takes for a GM tree to flower and fruit

UF IFAS UNIVERSITY of FLORIDA

Progress Made and Future Expectations

- The UF-CREC citrus improvement program has evaluated hundreds of transgenic lines in the last 10 years.
  - Antimicrobial Peptides
  - Systemic Acquired Resistance (SAR) inducing proteins

- Two plant-based proteins, NPR1 and SABP2, have resulted in enhanced tree growth while reducing *Ca*Las levels in the phloem.

Progress Made and Future Expectations

- UF–CREC has recently established a new 20-acre GMO evaluation field site (expandable to 50 acres) to rapidly screen large populations.
- We are evaluating not only GMO scions, but also GMO rootstocks that can potentially protect the non-GMO scion.
- Specific combinations include:
  - GMO sweet orange/grapefruit scion – Non-GMO rootstock
  - Non-GMO sweet orange /grapefruit scion – GMO rootstock
  - GMO sweet orange /grapefruit scion – GMO rootstock

Progress Made and Future Expectations

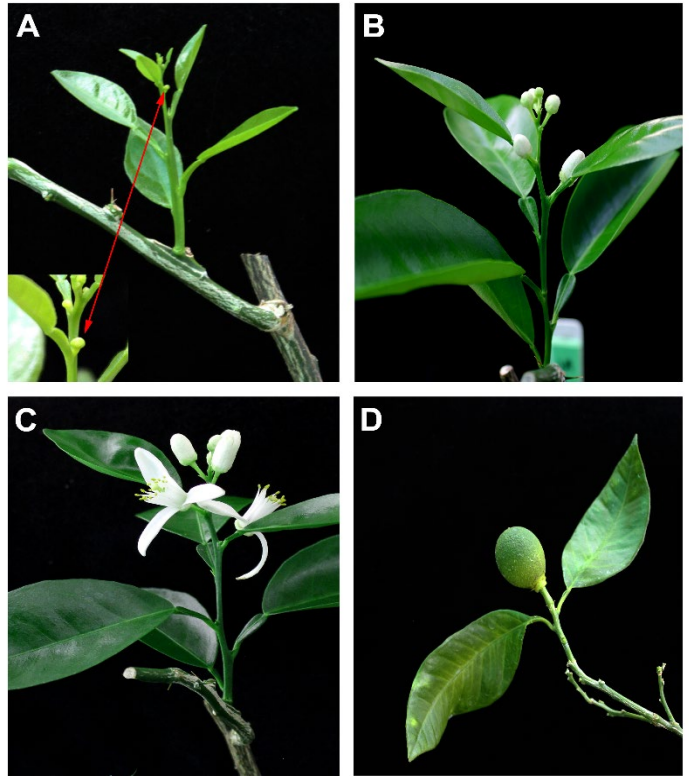
- We are generating field data that validates our preliminary observations, which will ultimately lead to GMO scions and rootstock release recommendations.

- A population of trees with stacked genes that function in different ways is being produced.
  - SAR gene stacked with BT gene
  - SAR gene stacked with BT gene and β-caryophyllene (psyllid deterrent) gene.

Progress Made and Future Expectations

How can we utilize GM technology to screen germplasm faster?

Overexpression of FT in the Phloem using an AtSUC2-CcFT3 construct can result in early flowering within 18 months of transformation.



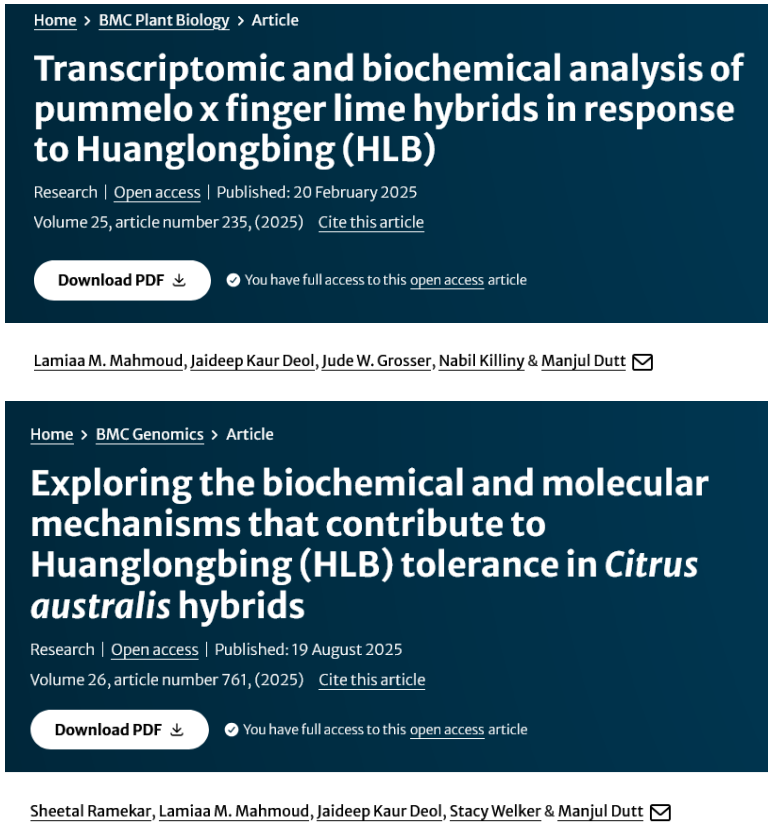
Precocious flowering one-year old non-transgenic 'Valencia' scion grafted onto *AtSUC2-CcFT3* transgenic rootstock.

(A) Flower bud emergence within 21 days following budding. Insert shows enlarged image of emerging flower buds
(B) fully expanded flower buds,
(C) fully open flowers,
(D) developing sweet orange fruit

UF IFAS UNIVERSITY of FLORIDA

Looking deeper into HLB-resistant species – How can they help us?

- Several wild citrus as well as some citrus relatives are resistant to HLB.
- Understanding their mechanism of resistance is providing us with
  - Genes to develop GMOs.
  - Genes for CRISPR.



Home > BMC Plant Biology > Article

Transcriptomic and biochemical analysis of pummelo x finger lime hybrids in response to Huanglongbing (HLB)

Research | Open access | Published: 20 February 2025

Volume 25, article number 235, (2025) Cite this article

Download PDF

You have full access to this open access article

Lamiaa M. Mahmoud, Jaideep Kaur Deol, Jude W. Grosser, Nabil Killiny & Manjul Dutt

Home > BMC Genomics > Article

Exploring the biochemical and molecular mechanisms that contribute to Huanglongbing (HLB) tolerance in *Citrus australis* hybrids

Research | Open access | Published: 19 August 2025

Volume 26, article number 761, (2025) Cite this article

Download PDF

You have full access to this open access article

Sheetal Ramekar, Lamiaa M. Mahmoud, Jaideep Kaur Deol, Stacy Welker & Manjul Dutt

Article | Open access | Published: 28 November 2024

Identification of CAP genes in finger lime (*Citrus australasica*) and their role in plant responses to abiotic and biotic stress

Lamiaa M. Mahmoud, Nabil Killiny & Manjul Dutt

Scientific Reports **14**, Article number: 29557 (2024) | Cite this article

2237 Accesses | **4** Citations | **2** Altmetric | Metrics

ORIGINAL RESEARCH article

Front. Plant Sci., 20 October 2022

Sec. Plant Breeding

Volume 13 - 2022 | https://doi.org/10.3389/fpls.2022.1019295

Insights into the mechanism of Huanglongbing tolerance in the Australian finger lime (*Citrus australasica*)

Kyle C. Weber[1†] Lamiaa M. Mahmoud[1,2†] Daniel Stanton[1] Stacy Welker[1] Wenming Qiu[3]

Jude W. Grosser[1] Amit Levy[1] Manjul Dutt[1*]

[1] Citrus Research and Education Center, University of Florida, Lake Alfred, FL, United States

[2] Pomology Department, Faculty of Agriculture, Mansoura University, Mansoura, Egypt

[3] Institute of Fruit and Tea, Hubei Academy of Agricultural Sciences, Wuhan, China

UF IFAS UNIVERSITY of FLORIDA

THANK YOU

UF IFAS UNIVERSITY of FLORIDA