The New Zealand Institute for Plant & Food Research Limited

Novel genetic marker techniques revolutionize apple breeding

Sue Gardiner (and lots of other people)

IPPS, April 2016

What does my work involve?

- I lead the Mapping & Markers Team that develops breeding tools for Plant & Food fruit breeders
- We work with breeders of a medley of crops:

  apple, pear, kiwifruit, peach, apricot, raspberry, blueberry, black currant, hops, manuka....
- Using our expertize in **genetics** and **genomics**

- **PFR goal is 'Better cultivars faster!'**

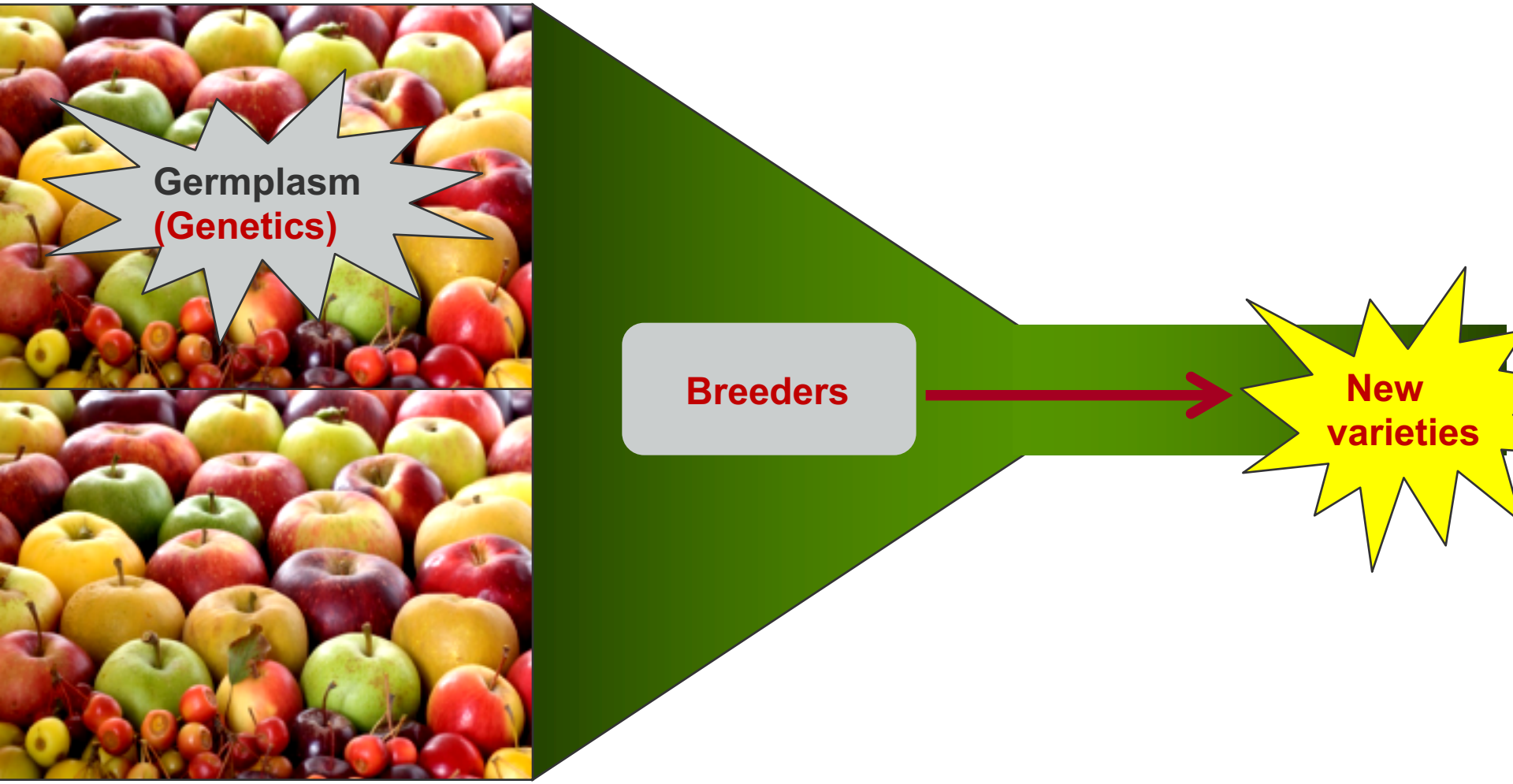
Topics for today

- **The pipeline for breeding new apple cultivars at Plant & Food Research**

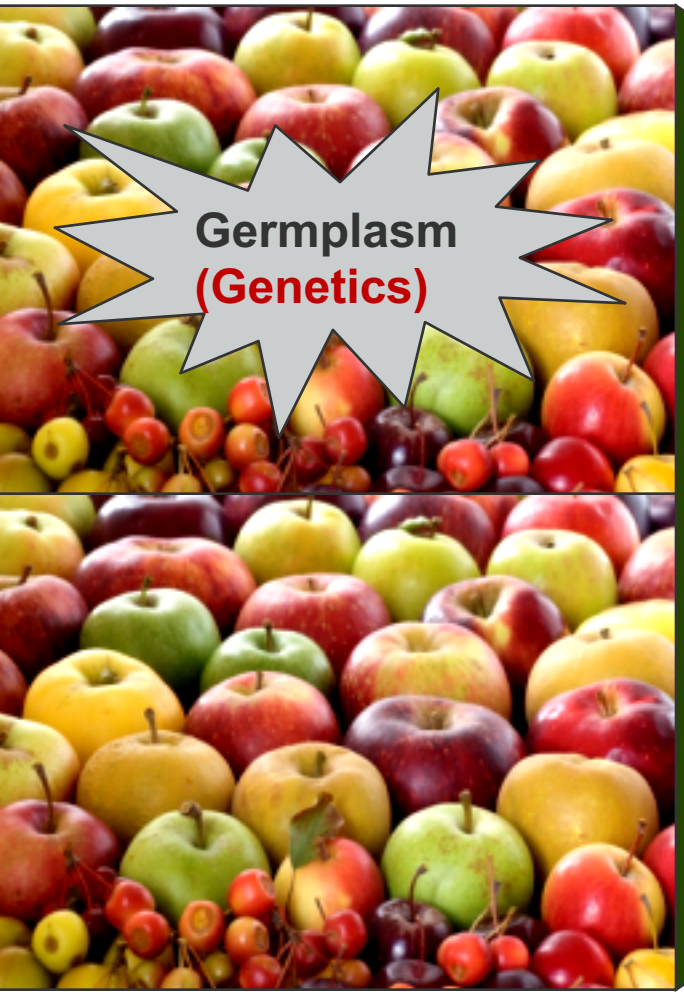
  How the Mapping & Markers Team works with industry and many other scientists to help breeders develop better cultivars faster

- **What are genetic markers?**
- **How these markers are used to select elite seedlings and increase the efficiency of breeding new cultivars**

Traditional breeding pipeline



New breeding pipeline – Industry #1



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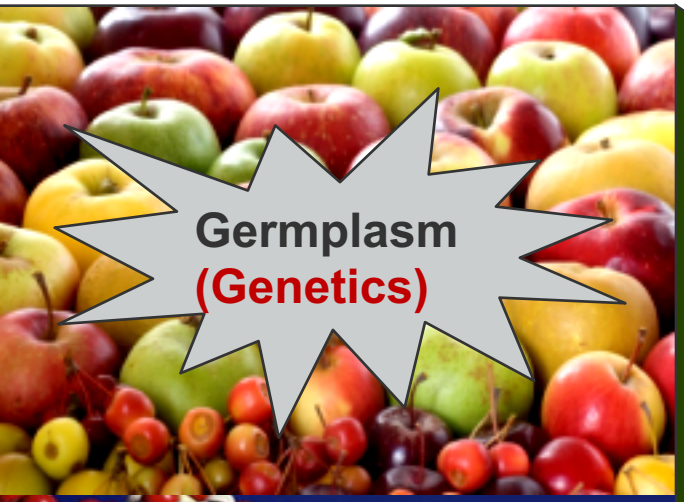
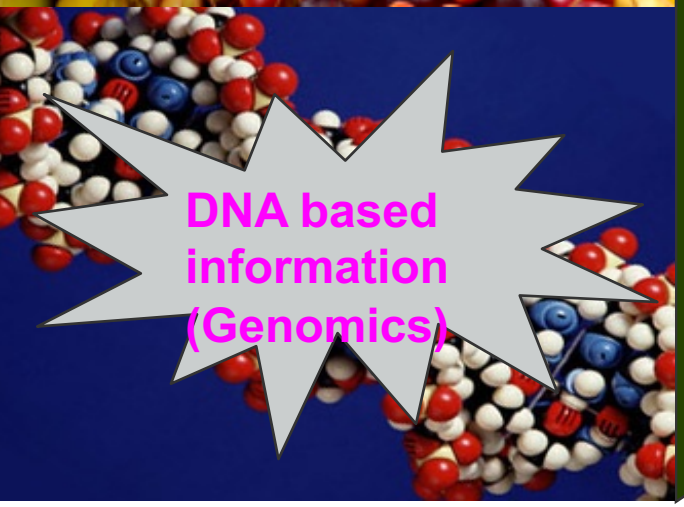
#1 Industry needs

Breeders

New breeding pipeline





Genomics research

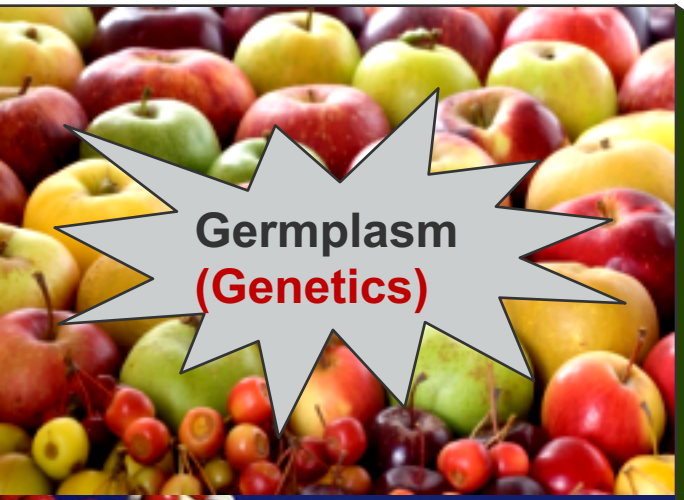
57,000 genes

Candidate genes

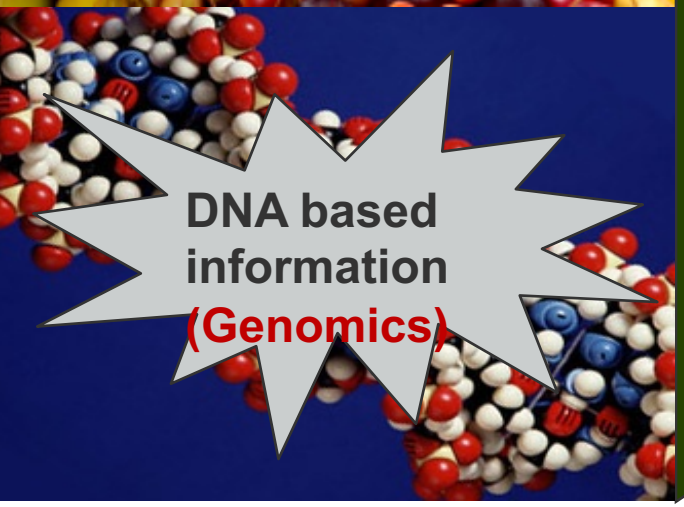
Industry needs

Breeders

New breeding pipeline



Germplasm (Genetics)



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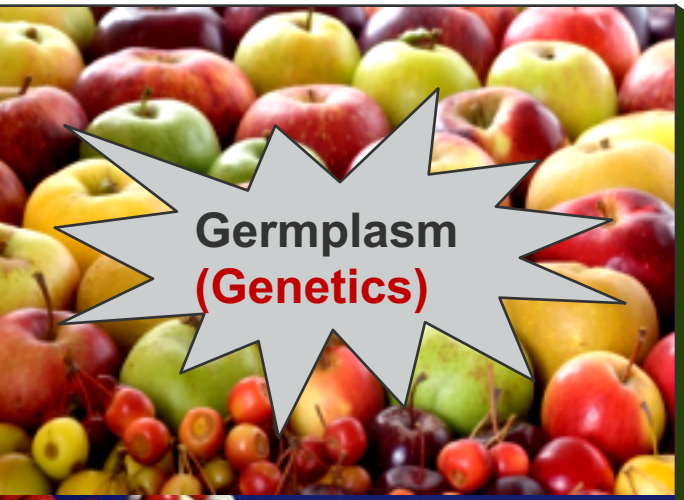

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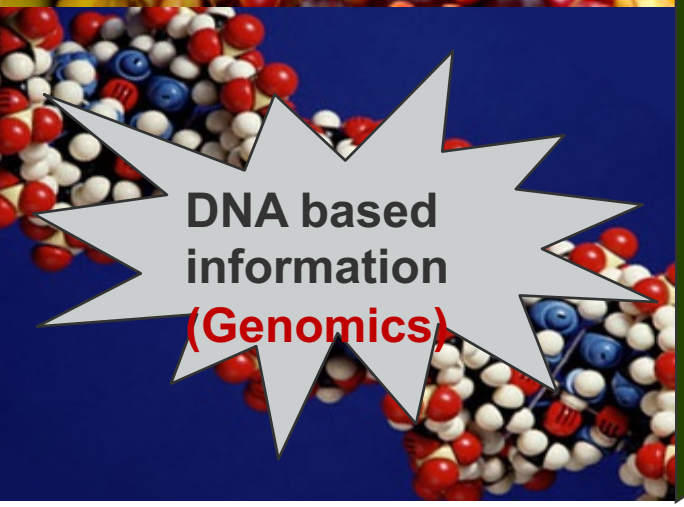

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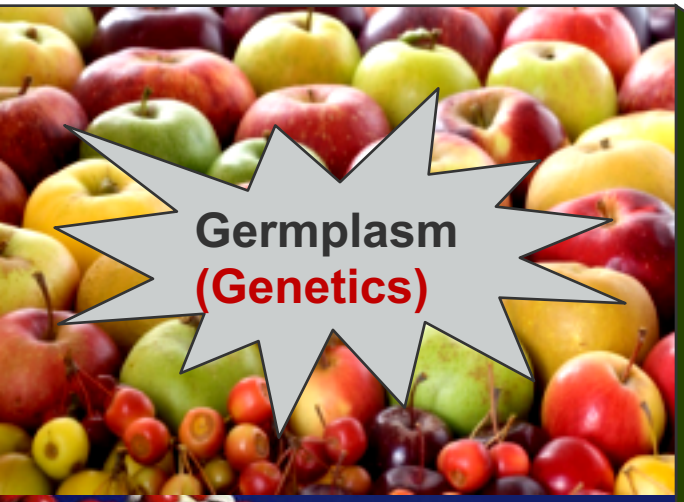

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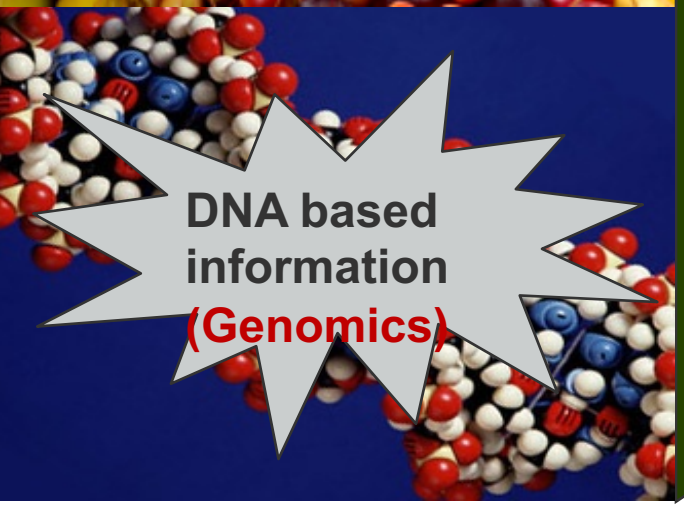

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Candidate genes

Genetic markers

Marker assisted selection

New varieties

Growers

Low-spray, crisp, juicy, flavoursome fruit to consumers

Two questions:

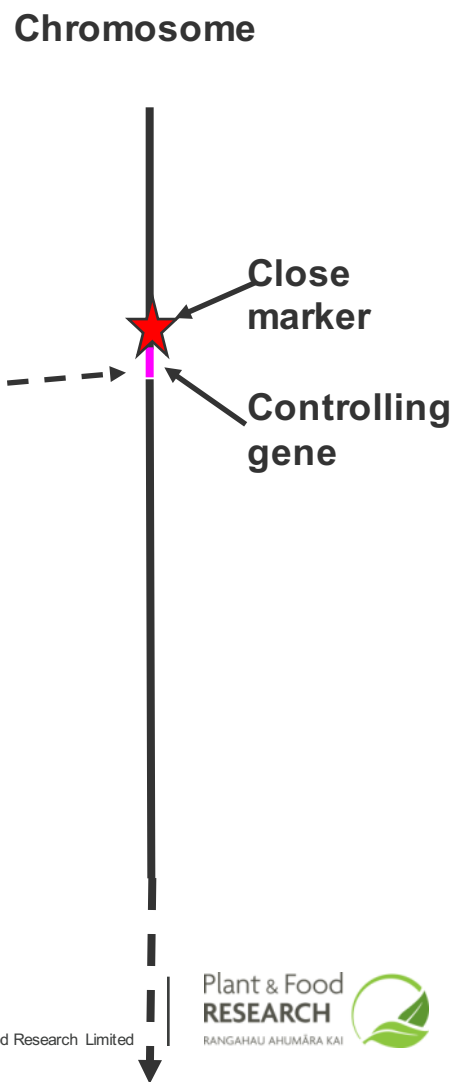
- **What are genetic markers?**
- **What does marker assisted selection involve?**

What are genetic markers?

Genetic markers can be compared to DNA fingerprints

- **A specific marker is associated with a specific trait**

  - it is either derived from a gene controlling expression of a trait eg. *dwarfing*; *red flesh; resistance to woolly apple aphid* or to *apple scab*



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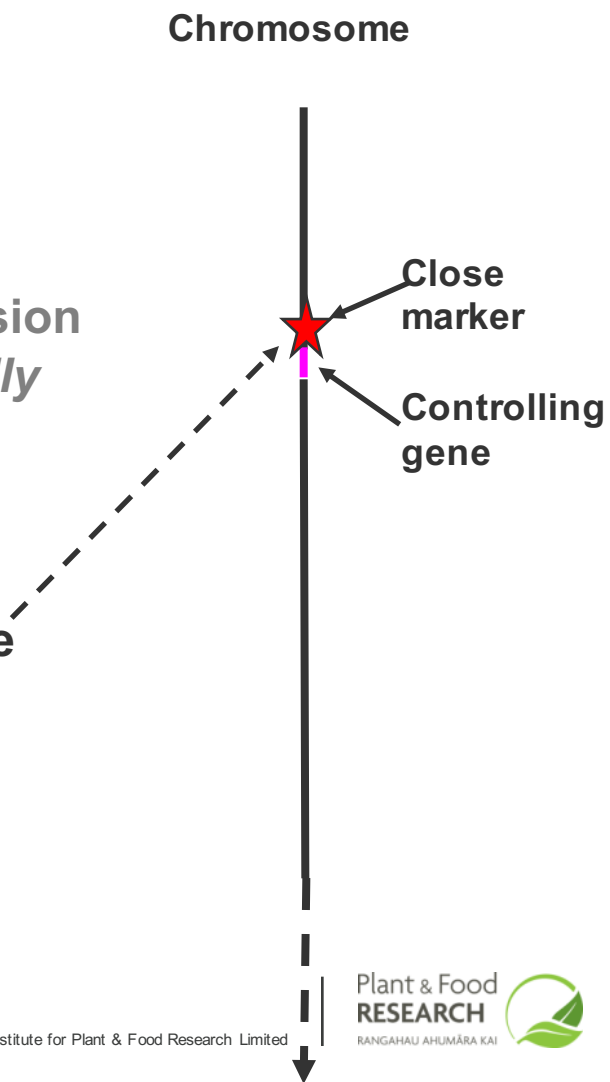

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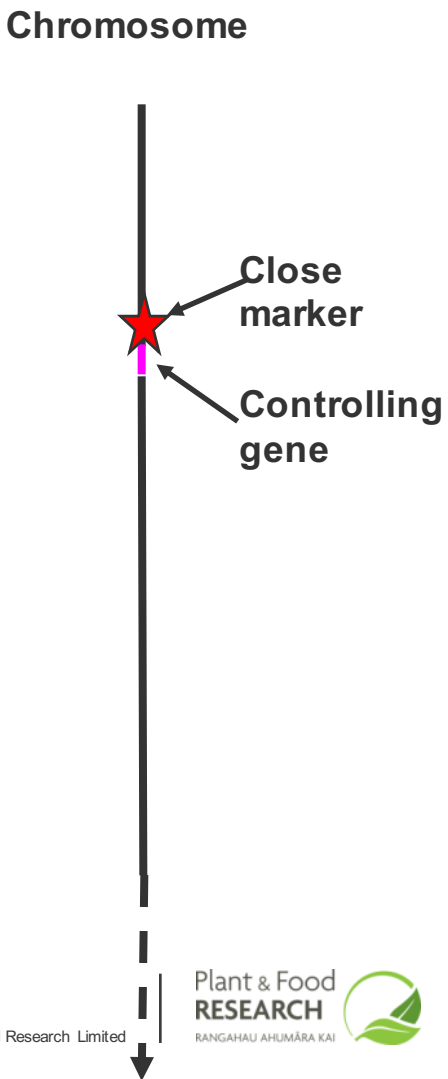
- **To test for presence of the trait, screen DNA extracted from each seedling, with the marker**



What is marker assisted selection?

Marker assisted selection is the use of markers to select seedlings carrying the trait of interest

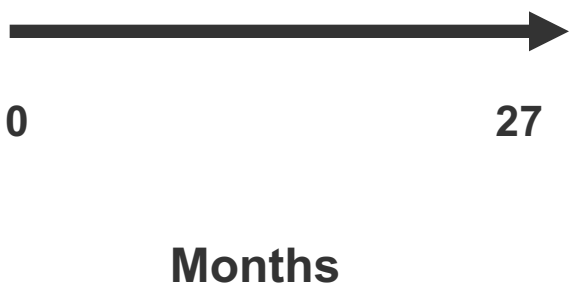
- **The seedlings need not express the trait before selection**

- **Seedlings that do not carry the marker are culled (50% of population for Mendelian inheritance)**

What comes next?

Marker assisted selection

Environmental promotion of flowering



- **Two flowering 27-month-old seedlings <u>predicted to have red flesh, using marker assisted selection</u> (AND top ranked fruit quality using genomic selection).**
- ***I will explain genomic selection another time!***

Marker assisted selection is used by PFR breeders:

For > 10 years marker assisted selection (MAS) employed as a tool to breed 'better apple cultivars faster'

- **Screen for seedlings with desired combination of 'must have' traits (*resistance to apple scab, powdery mildew, woolly apple aphid….with red fruit colour…)* expressed by the adult tree (6,000+ seedlings p.a)**

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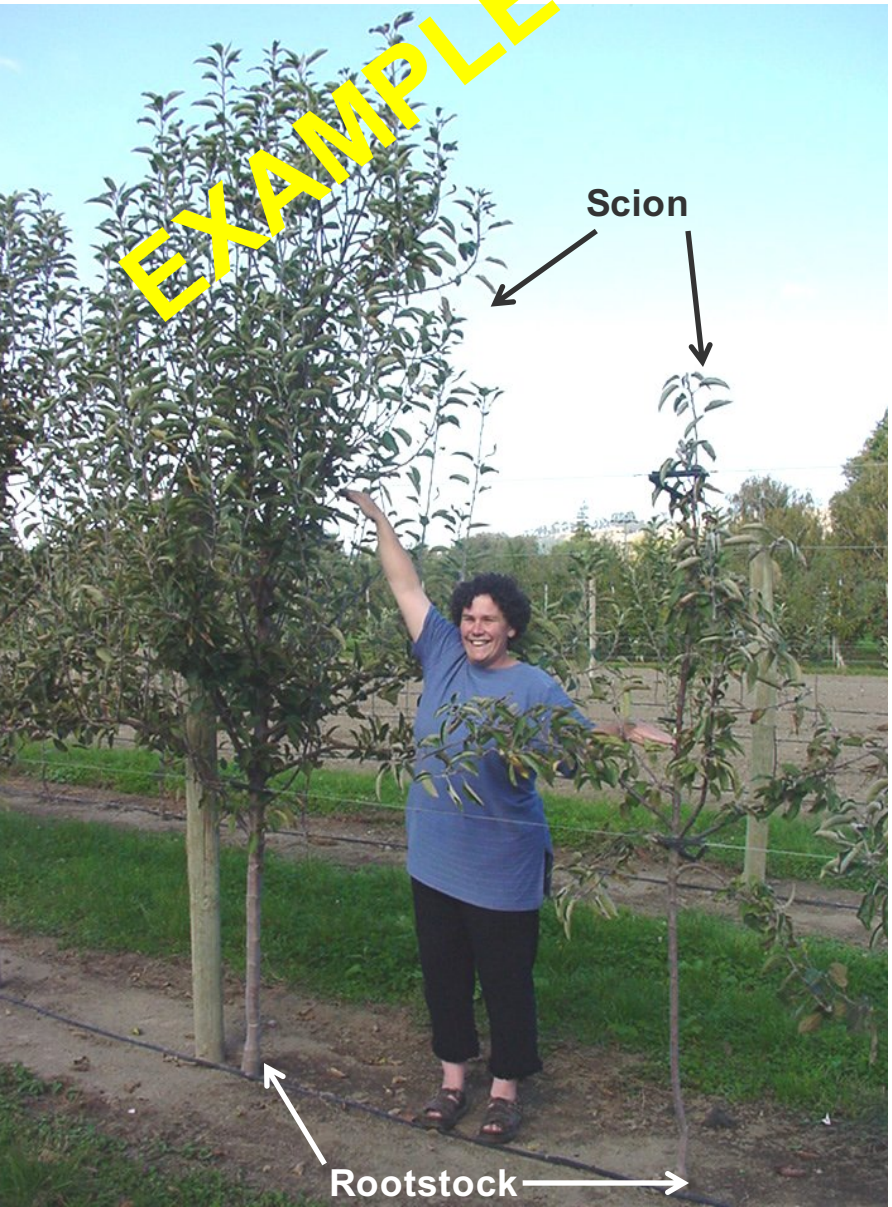
MAS helps apple breeders by:

→ **Minimizing orchard evaluation costs by reducing number of seedlings to be trialed in orchard**

→ **Reducing number of breeding cycles - e.g. can identify plants with pyramided resistances easily**

(Plants with two different resistances to same pest have a longer lasting, more durable resistance)

Commercial apple production relies on grafting of scion variety to dwarfing rootstock



→ ***dwarfing, pest + disease resistance, fruit production traits (time to fruiting, yield…)***

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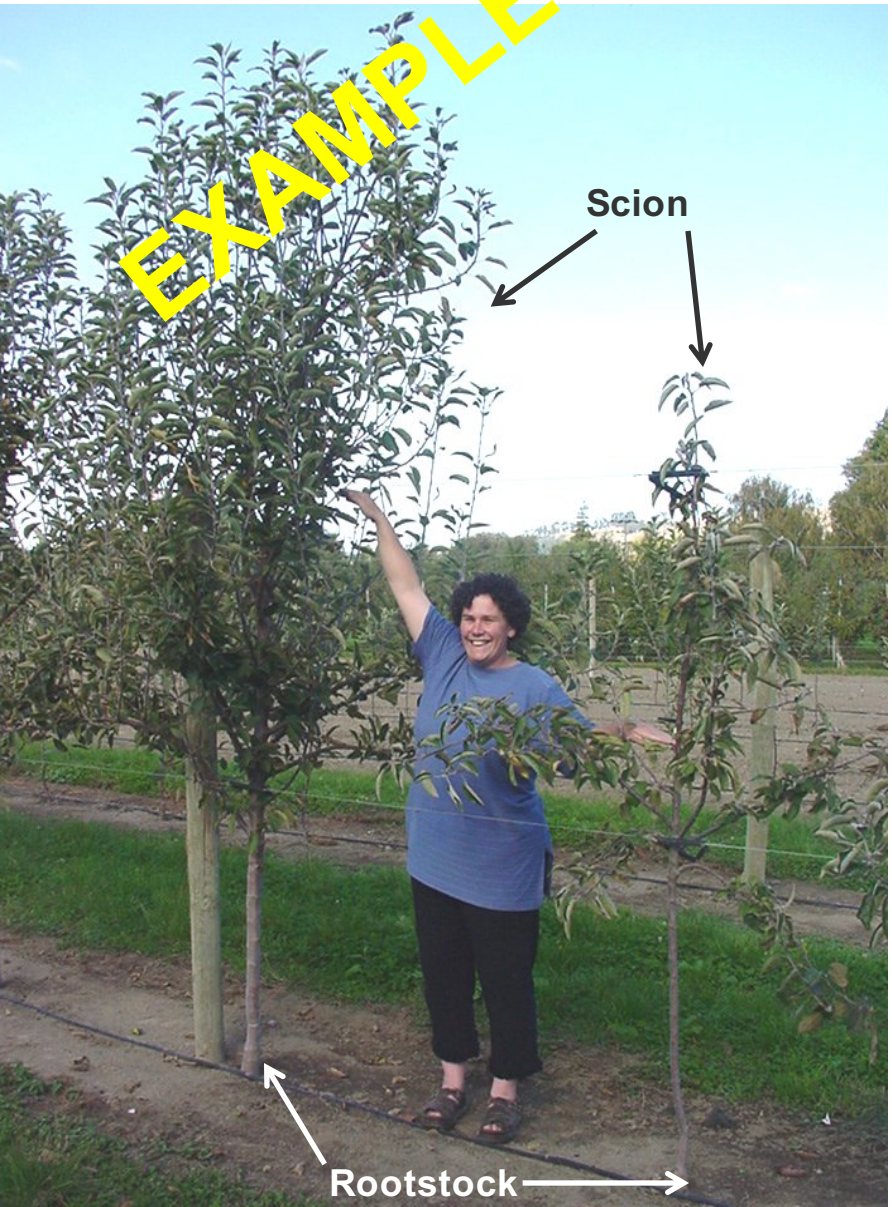

→ *dwarfing, pest + disease resistance, fruit production traits (time to fruiting, yield...)*

Breeder assesses rootstock seedlings on basis of traits expressed in grafted scion

- **Time consuming**
- **Labour intensive**
- **Expensive**

Impact of MAS in rootstock breeding

>> than for cultivar breeding.....

Example – 696 apple rootstock seedlings segregating for:

- **Fire blight resistance: *FB_R5***

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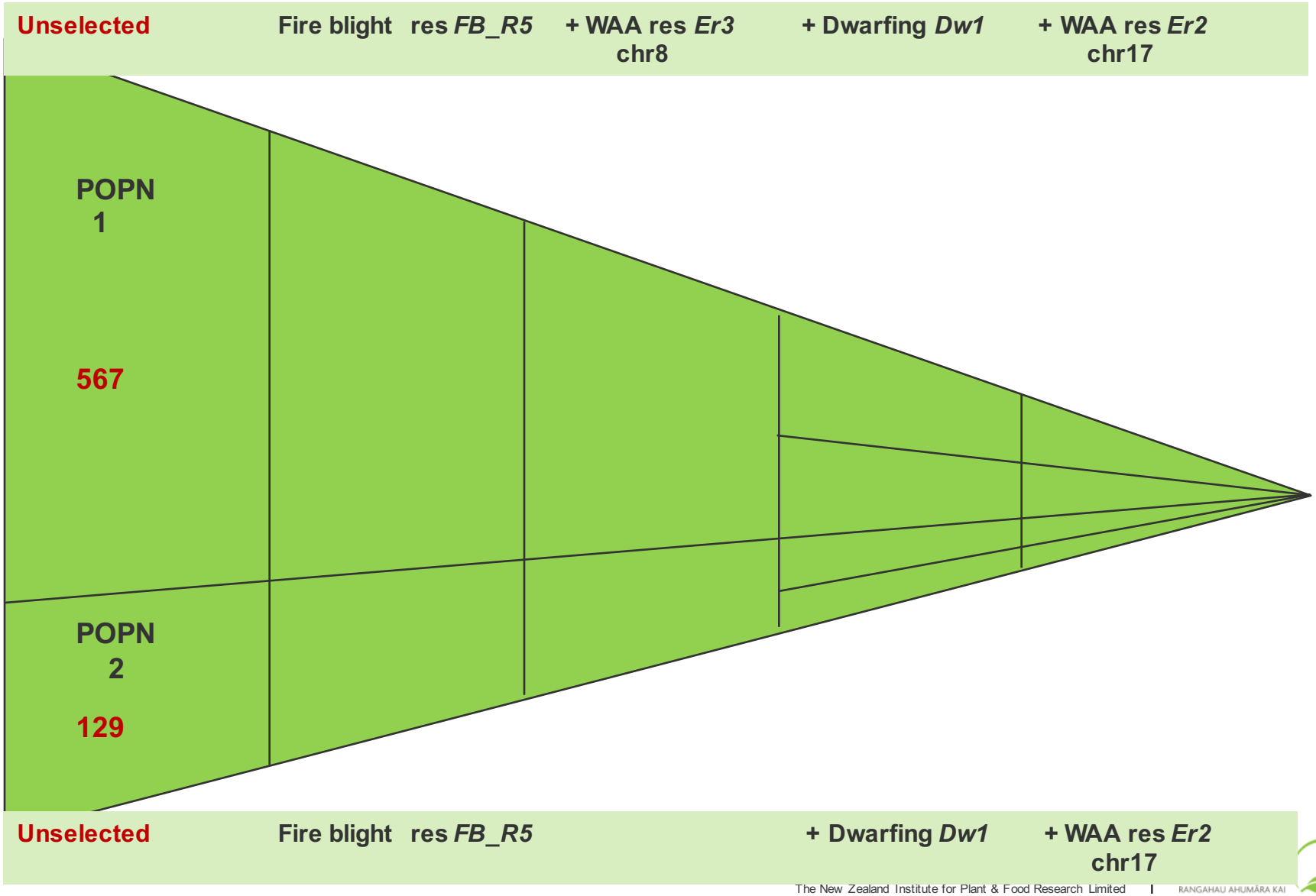
- **Fire blight resistance: *FB_R5***

- **Woolly apple aphid resistances: *Er3*, *Er2 on different chromosomes***

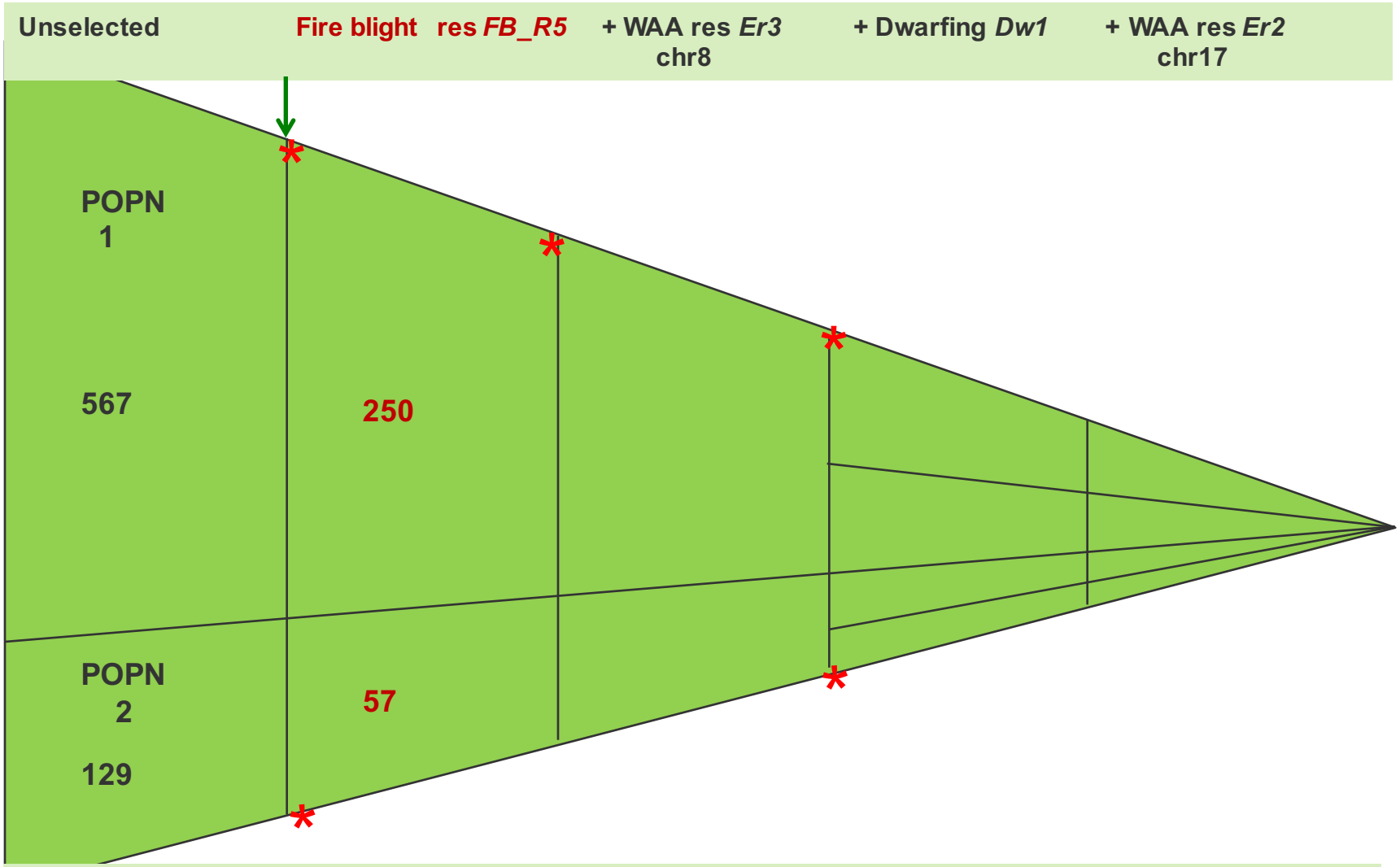
- **Dwarfing of grafted scion: *Dw1***

Goal: Identify seedlings carrying all 4 traits

Example – MAS on 696 apple rootstock seedlings



Efficient rootstock MAS – 'cherry pick' at steps*



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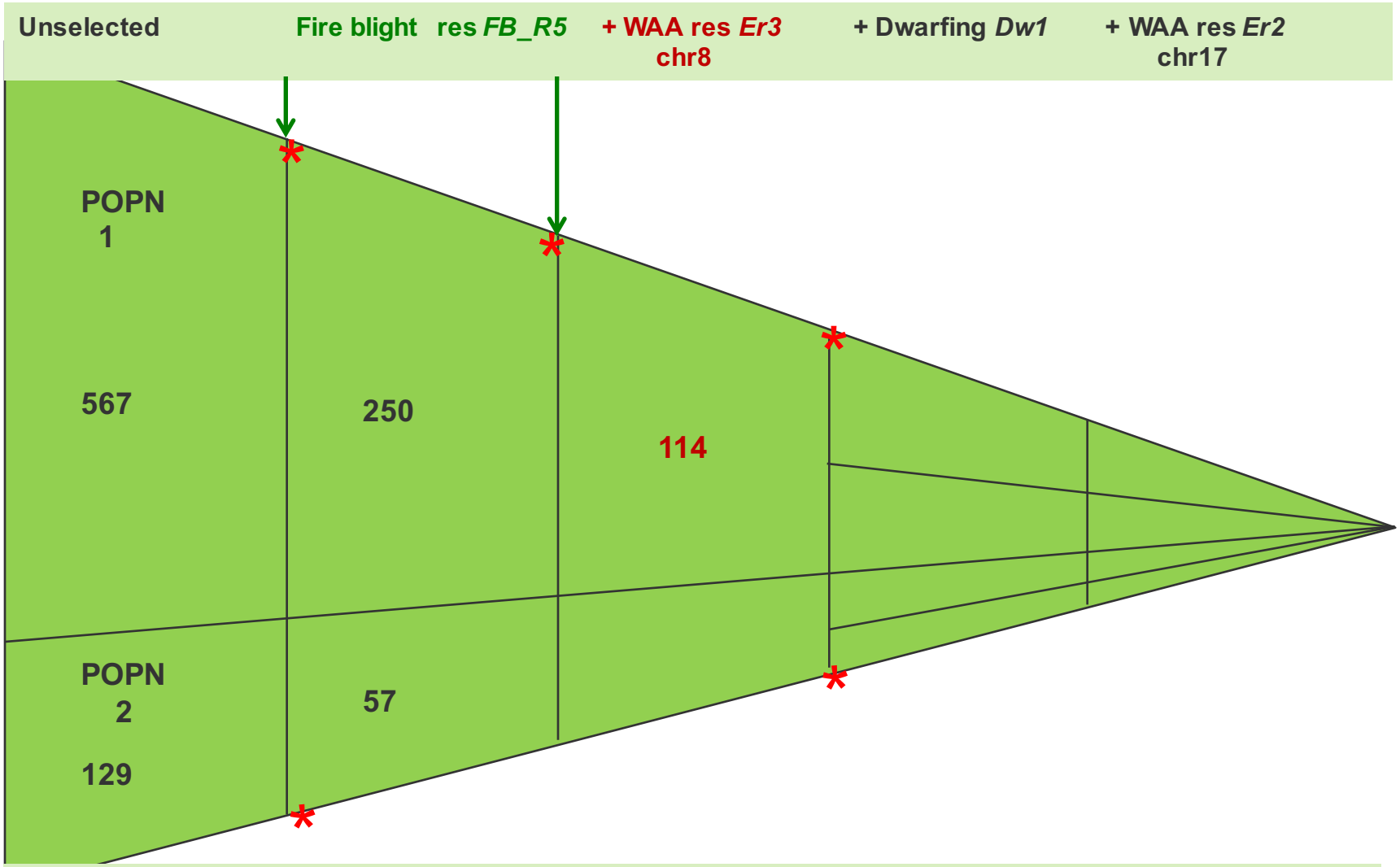

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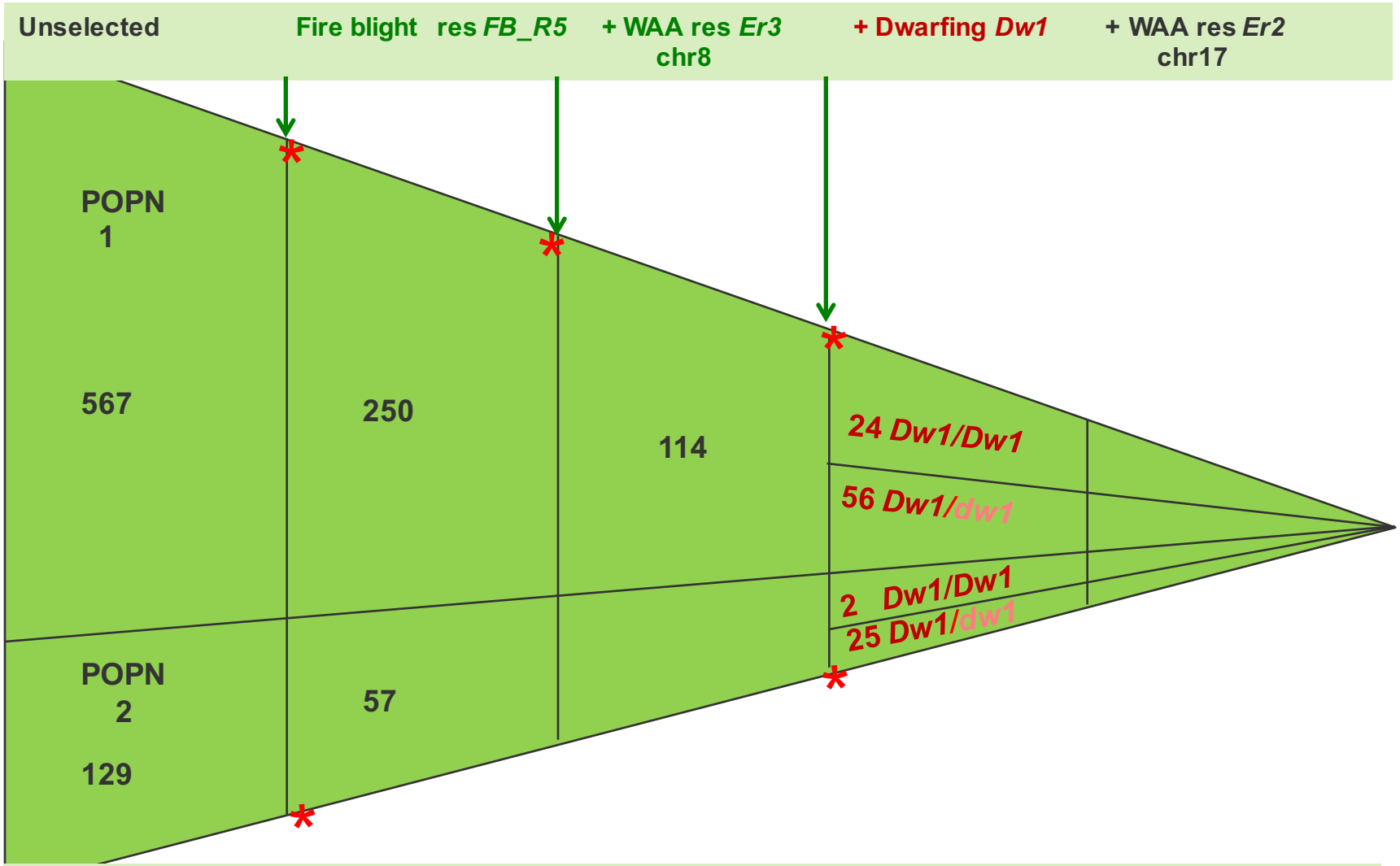

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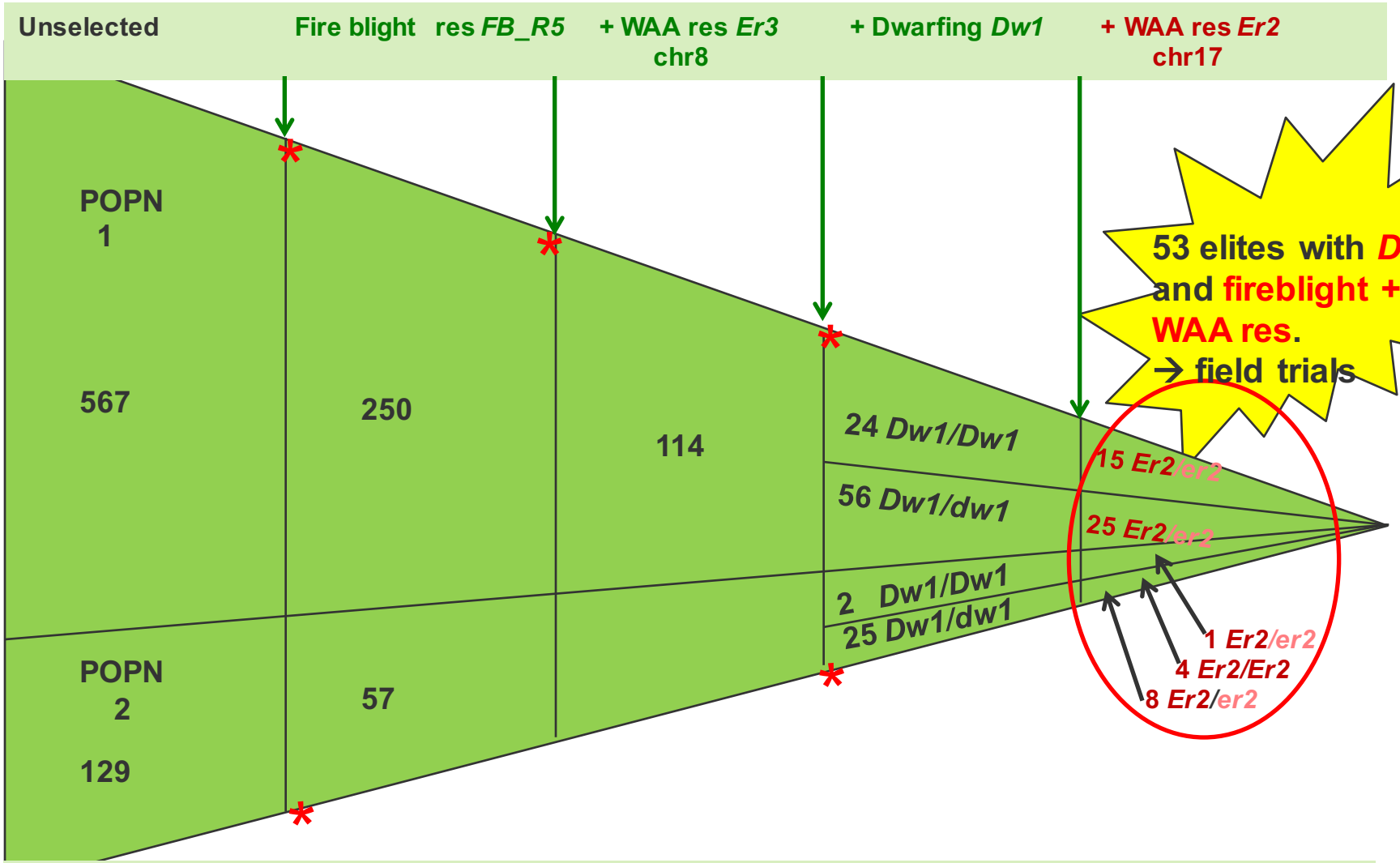

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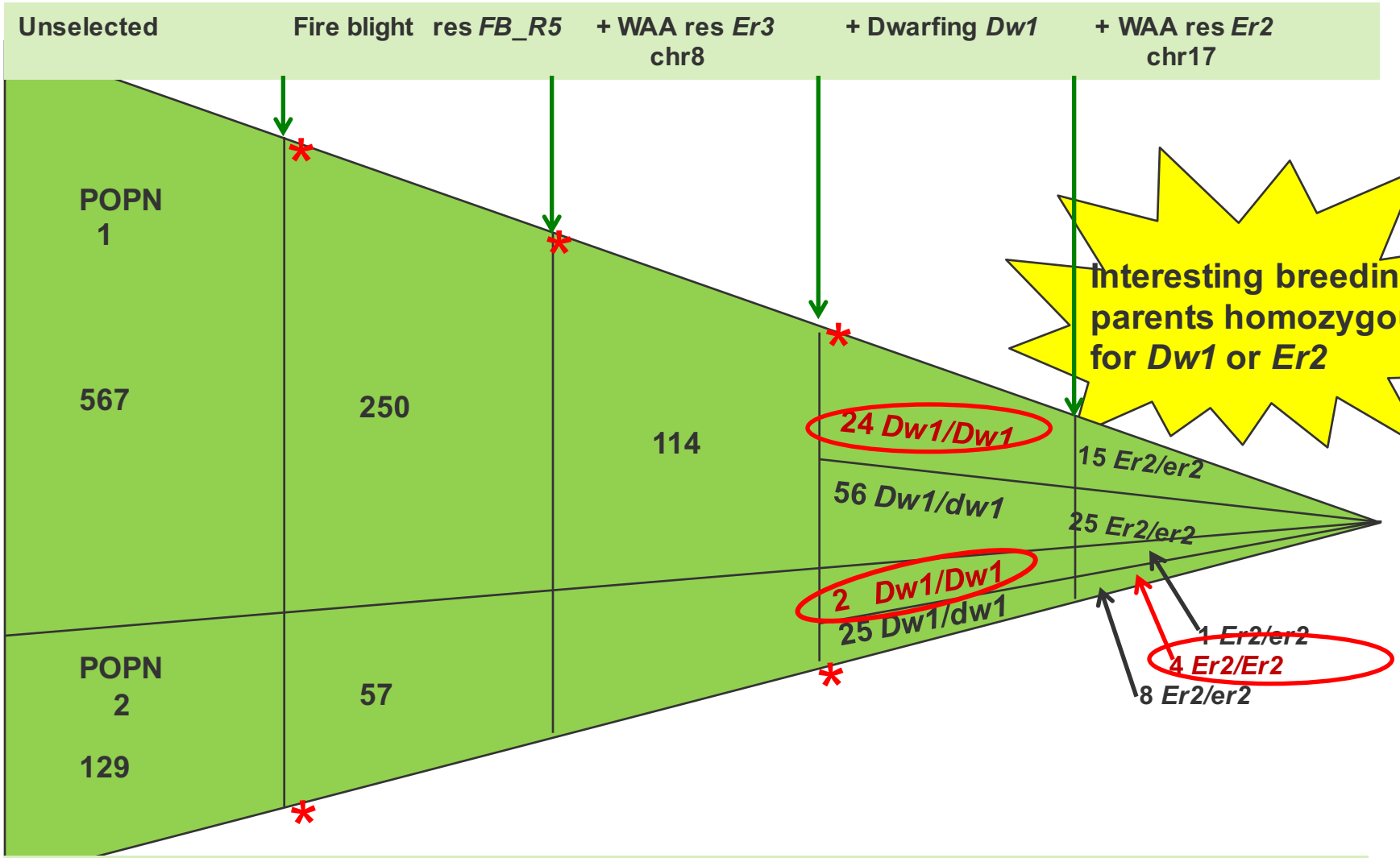


Apple rootstock MAS – numbers reduced

- **696 seedlings → 53 elites carry dwarfing trait AND resistance to fire blight and woolly apple aphid**
- **→ Only 7.6 % of population requires grafting for expensive, time-consuming breeder assessment**

Apple rootstock MAS – summary

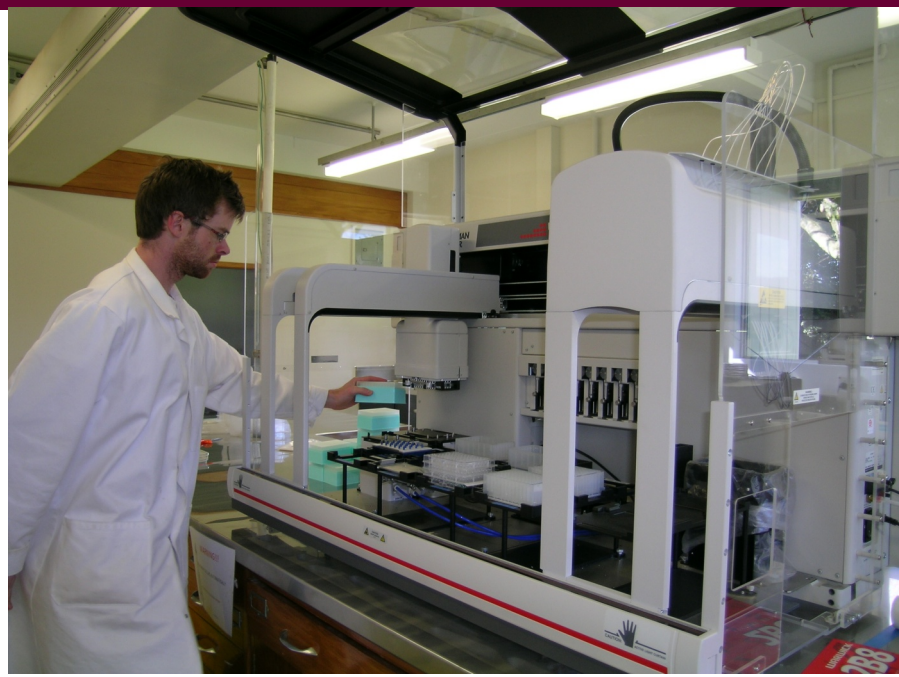
- **696 seedlings → 53 elites** carry dwarfing trait AND resistance to fire blight and woolly apple aphid
- **→ Only 7.6 % of population requires grafting** for expensive, time-consuming breeder assessment

- **40** elite dwarfing, fire blight-resistant seedlings **carry pyramided woolly aphid resistances *Er3* and *Er2*** → enhanced durability of resistance
- **20 elites** homozygous for dwarfing (*Dw1/Dw1*) → **potential ideal breeding parents**

Automation → key for efficient MAS

Slipstream Automation performs:

- **DNA extraction**
- **Marker screening**
- **Cherry picking of +ve seedlings after each step (cost reduction)**



Time: MAS < 2 weeks

Leaf → results to breeder

Rootstock breeding – orchard trials old way *vs* new way (-/+ MAS)

In both cases: First cull seedlings for negative characters: spines, burr knots (1 year)

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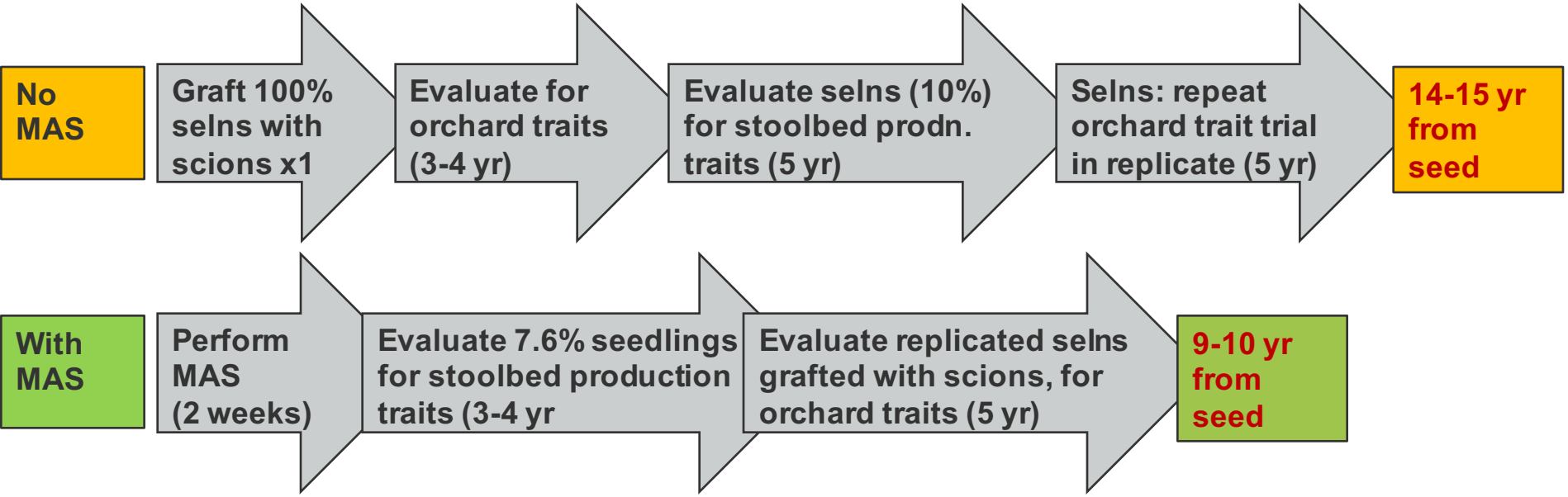

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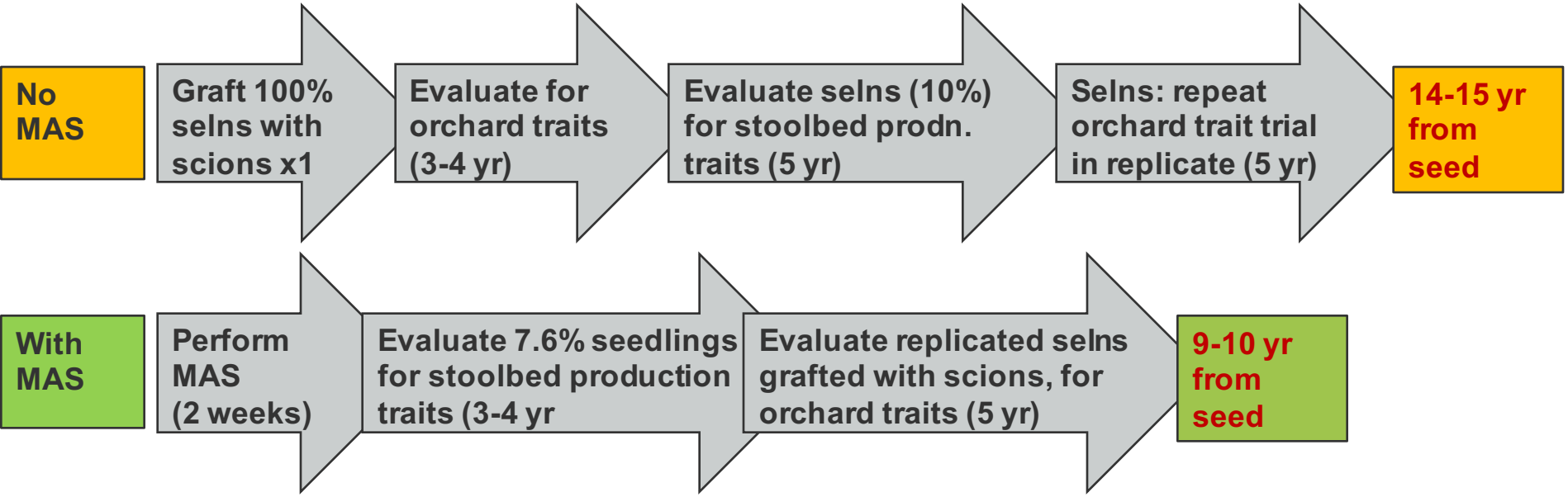

With MAS: takes 5 years less; fewer seedlings to assess →costs less

- Change in order of steps, omitted x1 orchard trait trial

Outcome of Plant & Food apple breeding innovation:

- **‘Better cultivars faster’**
- **More profit for New Zealand**

The New Zealand Institute for Plant & Food Research Limited

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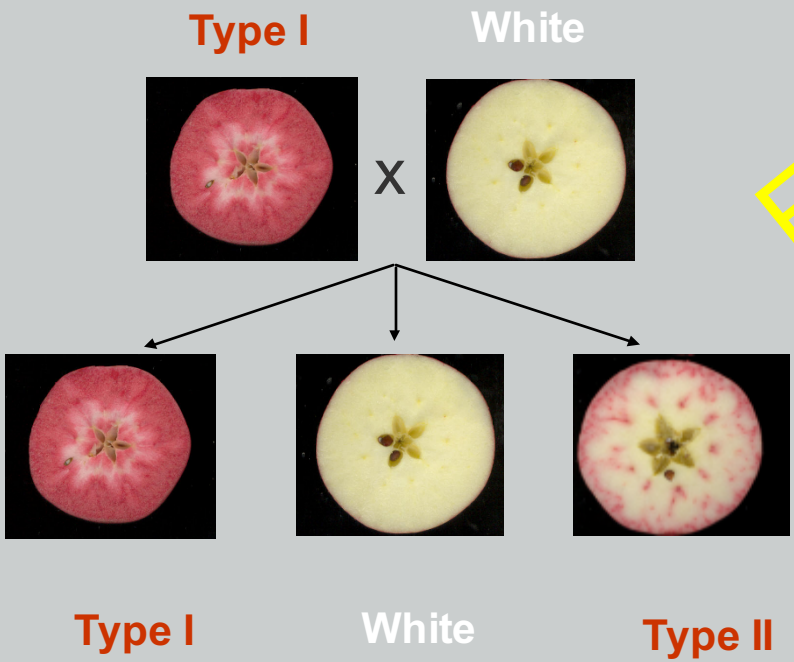
How do we test for association of a candidate gene marker with a trait?

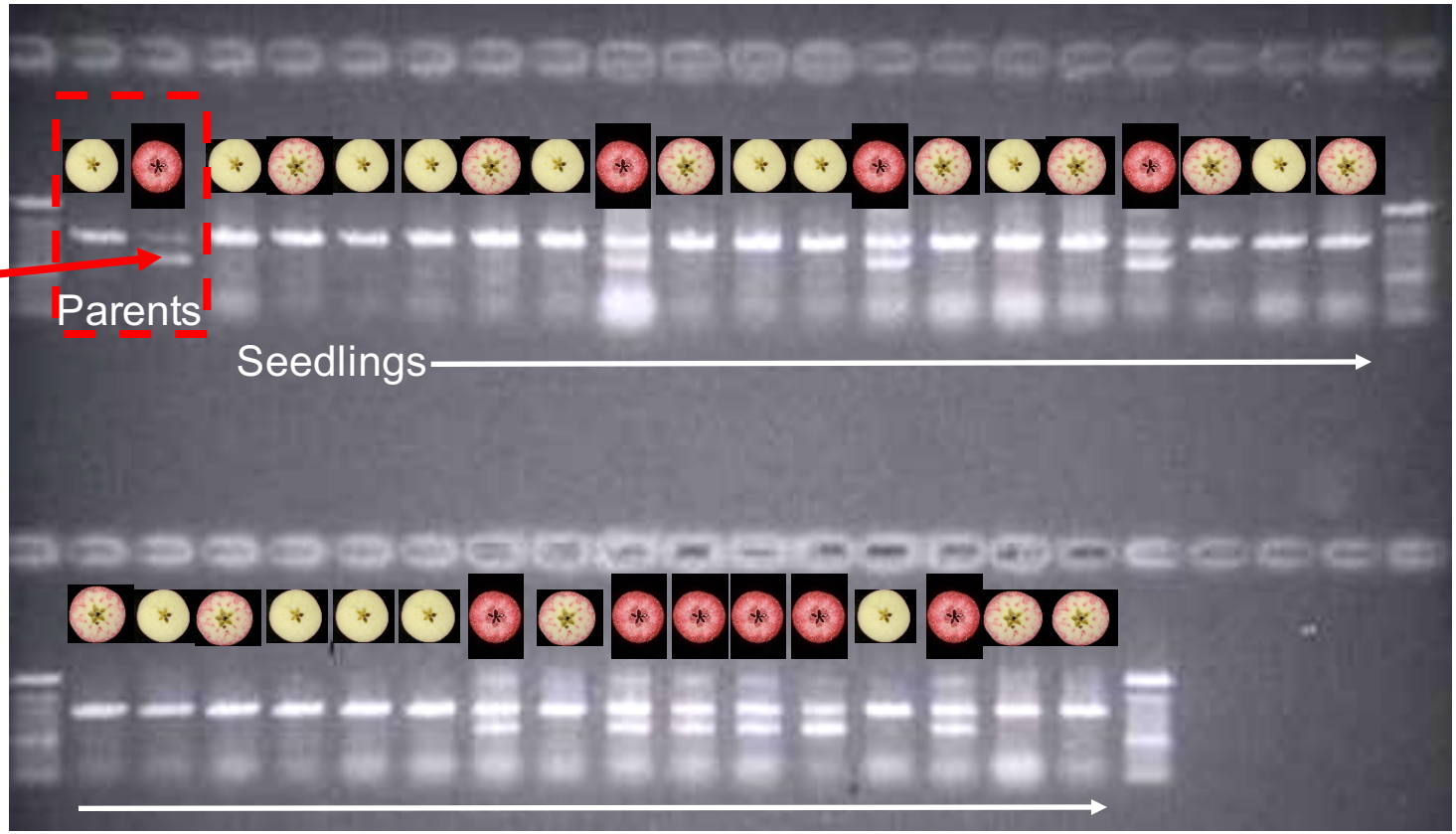
Develop a mapping population segregating for the trait of interest:

Example



Screening of a possible marker for Type I red flesh

***MdMYB10* marker screen – marker is linked to Type I red flesh, unlinked to Type II.**











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