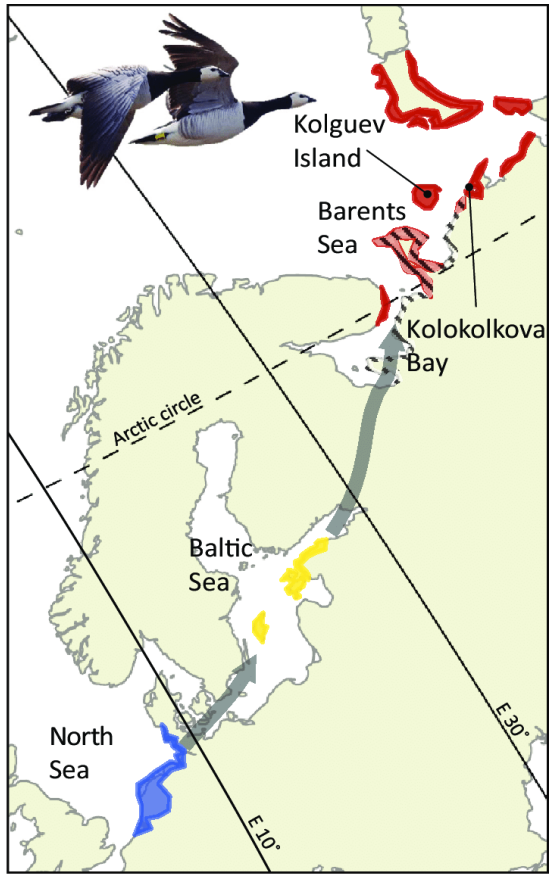
Nature, Technology and Society

Gert-Jan de Boer

April 2021

ENZA ZADEN

Our world is all about viruses. Has been and will be!

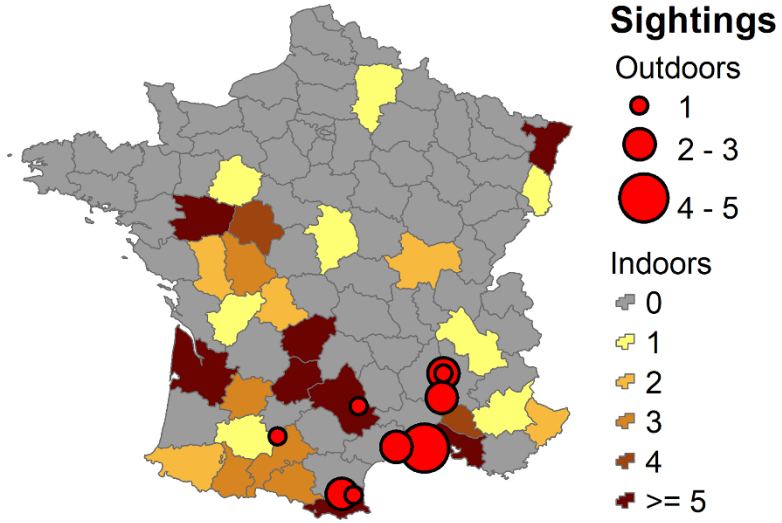


At least 20 livestock ships caught in Suez canal logjam

Concerns for animals' welfare if Ever Given blockage crisis is protracted

▲ Ships are anchored outside the Suez canal in Ain Shokhna, near Suez, Egypt, 26 March 2021. Photograph: Khaled Elfiqi/EPA

Climate change might add other challenges to crop protection



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In the news

USDA: action to protect U.S. from ToBRF

26/11/2019 - François-Xavier Branthôme

Action will prevent the introduction of To NEWS

the U.S.

ovember 18, 2019, USDA's Animal and announced it is taking immediate action rown rugose fruit virus into the United St production worth more than USD 2.3 bi lude protection against the significant dar omato crops, as already decided in some cted in Belgium, China, Italy, Greece, an, Turkey and the UK. The virus was det mato greenhouse in 2018. Tom s in tomatoes

ToBRFV: Quarantine status in effect from 1 November

24/09/2019 - François-Xavier Branthôme

Europe institutes emergency regulations on ToBRFV

The Tomato brown rugose fruit virus (ToBRFV) will get a **quarantine status (Q status), starting November 1**. From that day on, a European emergency regulation will come into force, which includes that each case of the virus in Europe must be made public. The decision was made by the European commission in July. For tomato and bell pepper, specific measures will apply, with nd movement of the virus in the EU being prohibited.

Johanna Diener, Westhof Bio-Gemüse:

"TOBRFV is actually very similar to the Corona-virus"

The German tomato season is in full swing: large harvest volumes in the Netherlands and in Germany are causing volume pressure and falling market prices, says Johanna Diener from Westhof Bio-Gemüse GmbH & Co. KG. The market situation for conventional produce is also affecting the organic tomato market.

NVWA vindt ToBRFV in zaad uit Peru

GLASGROENTEN JOOST VAN WINSEN 11 FEB 2020 OM 11:34UUR

Het Tomato brown rugose fruit vir

(NVWA) aangetroffen op tomatenza

Consequences of a new quarantine disease

- OPERATIONAL COST & COMPLEXITY INCRESASE across the value chain, from research, to seed production, and tomato producers and traders.
- SOCIAL STIGMA & ECONOMIC IMPACT due to TRADE DISRUPTION to growers.
- COST PRICE OF SEED INCREASES making it more difficult to produce for emerging markets with a low seed price.

VEGETABLE GROWERS NEWS

COVID-19 | BY REGION | VEGETABLES | LABOR | CROP PROTECTION | CROP MANAGEMENT | TECHNOLOGY

JUN 5, 2020

APHIS restricts imports of tomato, pepper due to ToBRFV

USDA's Animal and Plant Health Inspection Service has amended Federal Order for U.S. Imports of tomato (Solanum lycopersicum) and pepper (Capsicum spp.) hosts of Tomato brown rugose fruit virus (ToBRFV).

Effective June 5, 2020, after issuance of this Federal Order, the USDA's Animal and Plant Health Inspection Service (APHIS) is amending the restrictions for the importation of tomato (Solanum lycopersicum) and pepper (Capsicum spp.) hosts of Tomato brown rugose fruit virus (ToBRFV).

Specifically, APHIS is amending the import requirements for tomato and pepper fruit for consumption by adding restrictions for tomatoes and peppers from the Dominican Republic, France, and Spain. APHIS has detected ToBRFV in tomato fruit imported from the Dominican Republic, and received official reports of the disease in France and Spain.

SeedWORLD PRO

Tomato Trade And Blockade: Ancillary Pieces

In 2019 the United States Department of Agriculture updated the import requirements for tomato and pepper seeds twice. This module provides a step-by-step overview of what happened and the implications for seed companies. This PRO content also walks you through how to understand the documents released, with annotated documents and comprehensive insights – learn to read and interpret these documents, just like our experts do.

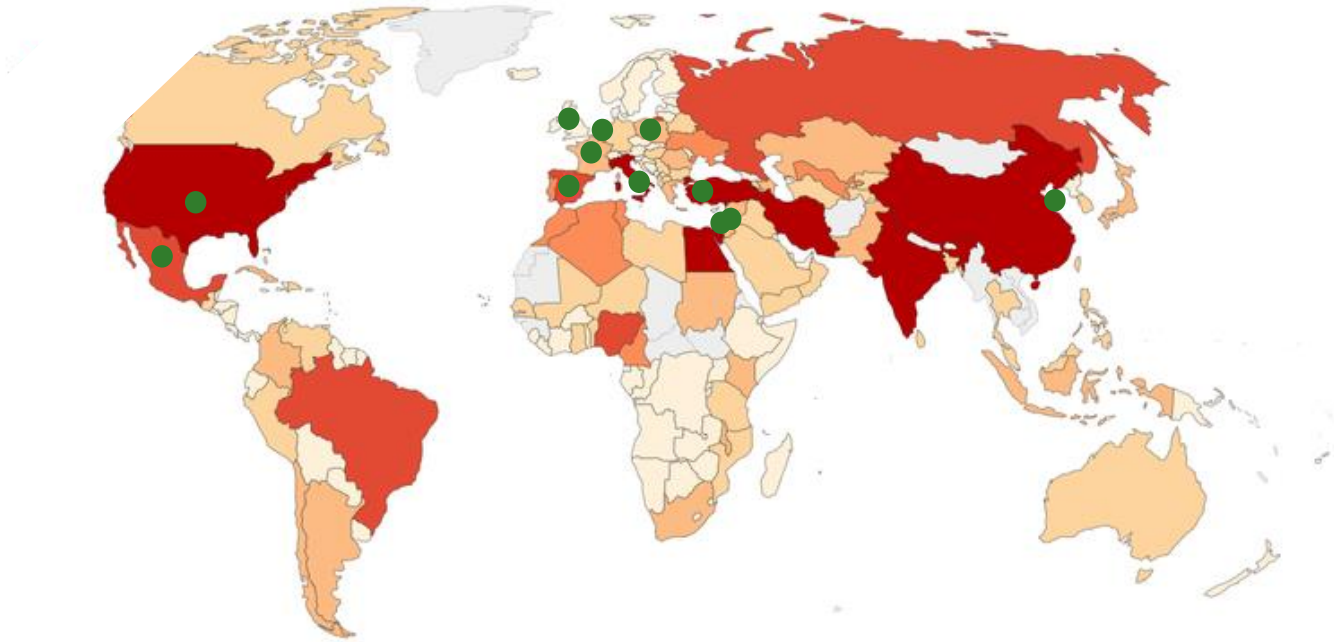
Fresh Plaza

Mexico adopts measures to prevent tomato virus from spreading

Mexico's agricultural authorities have established a series of actions to prevent the so-called rugose tomato virus from spreading in the country.

The National Health, Food Safety and Quality Service (Senasica), an agency of the Ministry of Agriculture and Rural Development (Sader), established phytosanitary measures, such as restricting the importation of seeds for experimental and research purposes, as well as modifying 233 kinds of combinations and requirements for the import of seeds, plants, seedlings, and cuttings of tomatoes, chili, and eggplant.

A global crop, a global threat

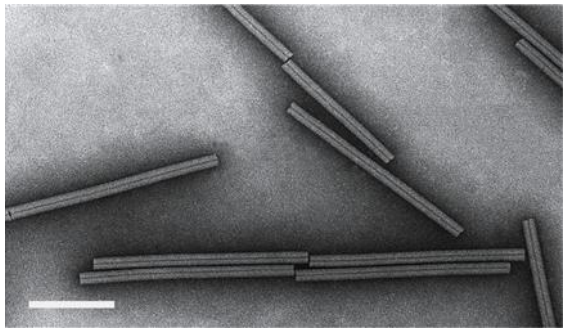


virus presence

ToBRFV has spread fast globally.

Source: https://gd.eppo.int/
Source: our world in data.

The tobamoviruses

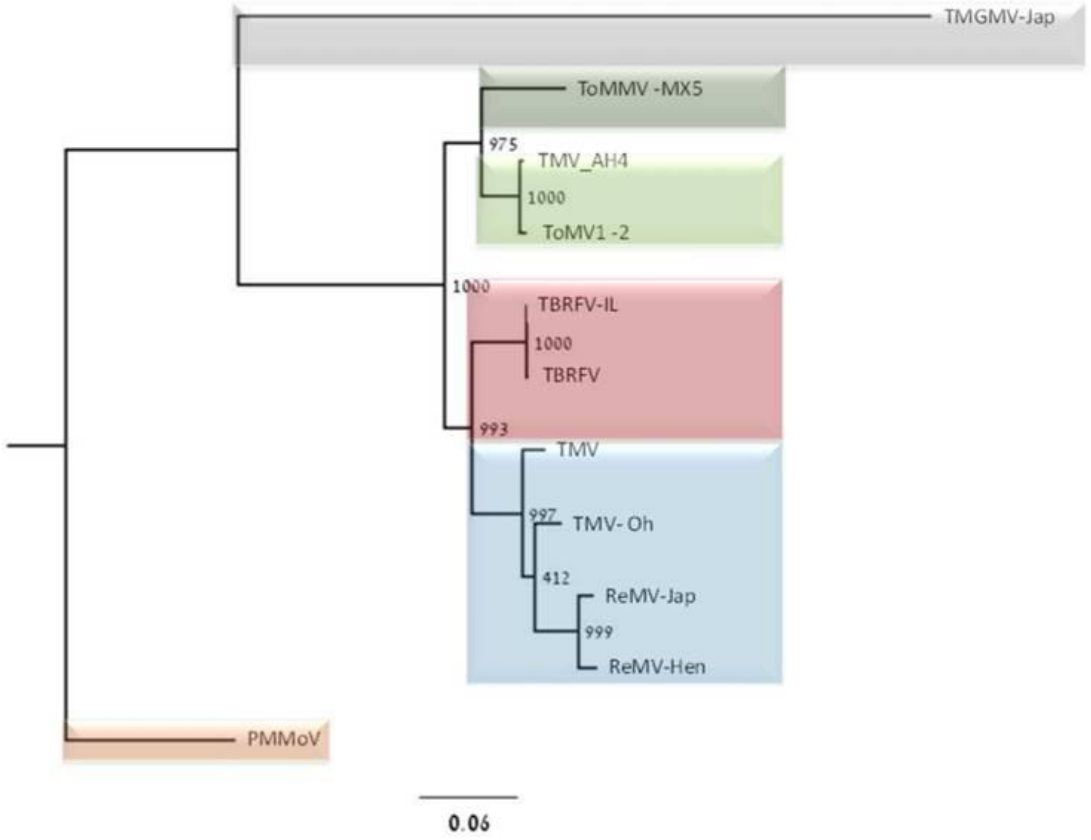


- Genus Tobamovirus, family of Virgaviridae
  - Single stranded genomic RNA
- 6.3-6.6 kb genome size

- 37 species in tobamovirus group
- Consists of two groups
  - Tobamovirus group 1 - Solanaceae
  - Tobamovirus group 2 – Cucurbit viruses

- Tobamoviruses are very stable and can survive for long times

The tobamoviruses are a large group and are very stable



Source: Luria et al., 2017. PLOS ONE

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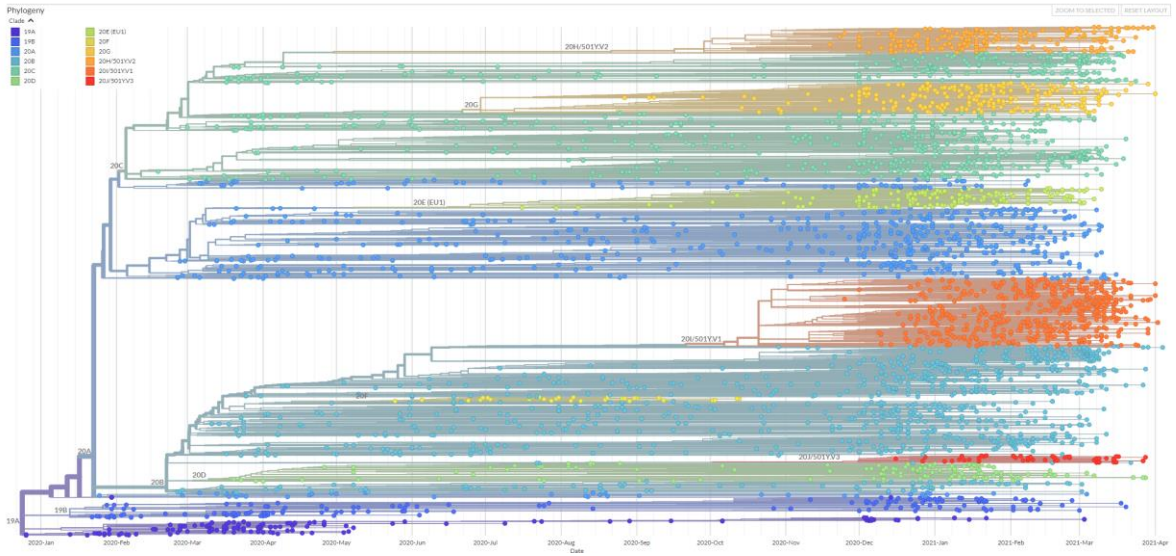
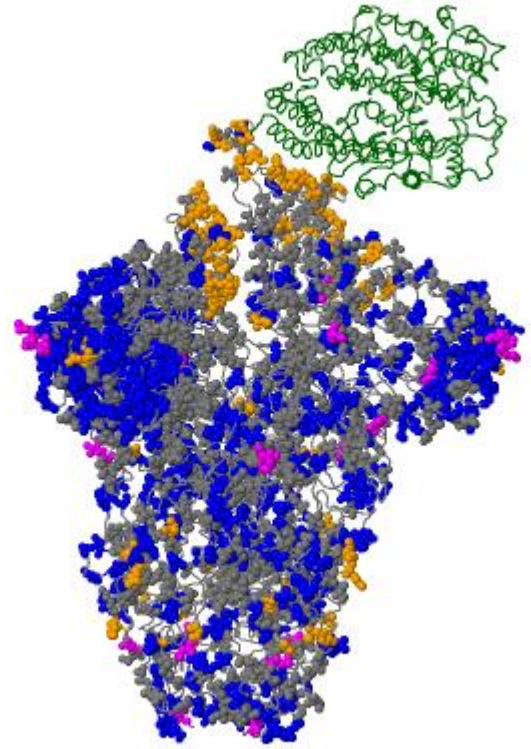
Covid-19 as an example of rapid evolution and adaptation





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Discovery of a new tomato virus

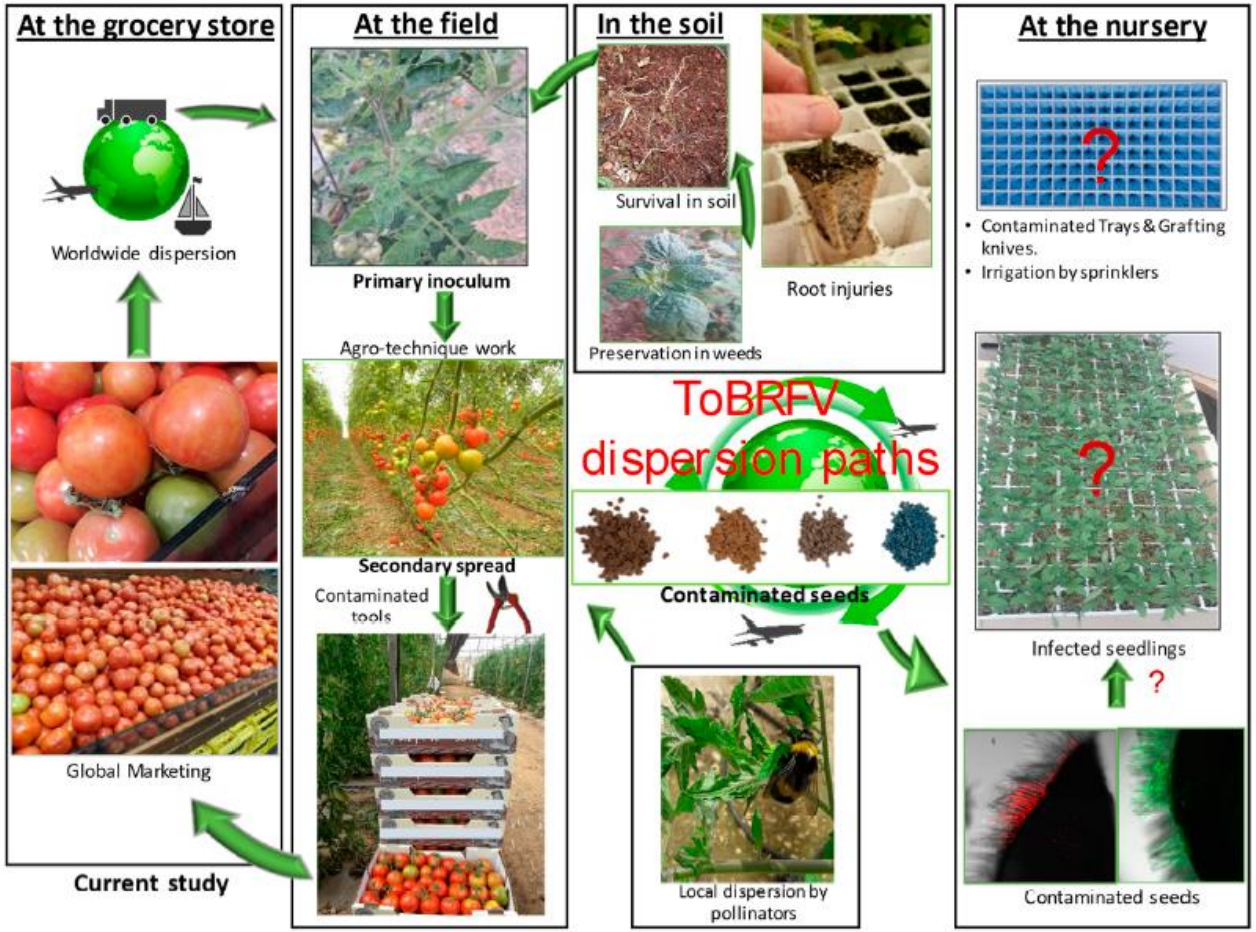
- September 2014: outbreak with a new virus in tomato in Israel
- Two diagnostic samples arrived for analysis in Enkhuizen
  - Saudi Arabia (Jan. 2015)
  - Jordan (June 2015)
- Looked like tomato or tobacco mosaic virus (ToMV / TMV) -> ToMV Elisa positive
- However, both virus isolates break ToMV / TMV resistance!
- Sequencing: tobamovirus with 80-85% identity with ToMV and TMV -> new virus!
- First described in a paper by Salem *et al*. in Nov. 2015: **Tomato Brown Rugose Fruit Virus** (ToBRFV)

ToBRFV: a new Tobamo virus found in 2014

Tobamovirus epidemiology

- Virus spreads:
  - Seeds
  - Water
    - Irrigation water
  - Pollen
  - Bumblebees
  - Mechanically/Contact
    - Hands and tools
    - Tobacco/cigarettes
    - Fresh fruits/Tabasco (sauces)

- Co-infections with other viruses make symptoms worse and plants more susceptible
- Up to 100% yield loss



*Source: Plants **2020**, 9, 623; doi:10.3390/plants9050623*

Virus is very contagious and spread by all kind of media.

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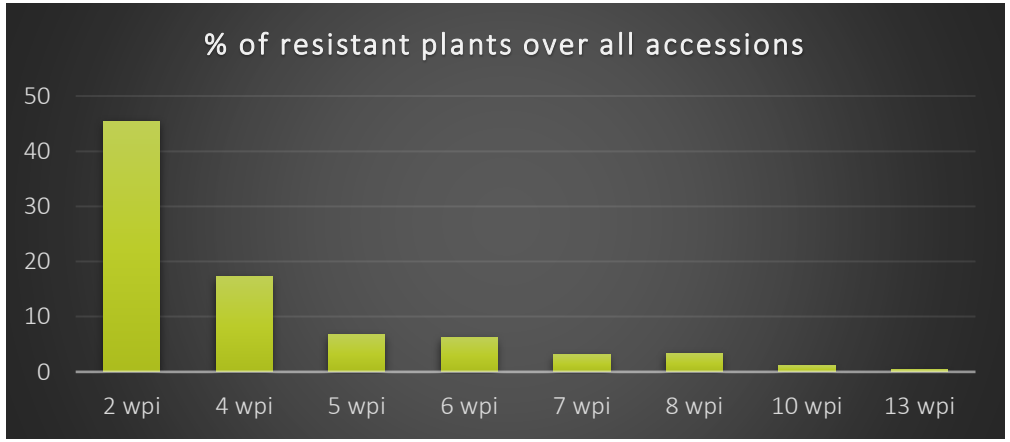
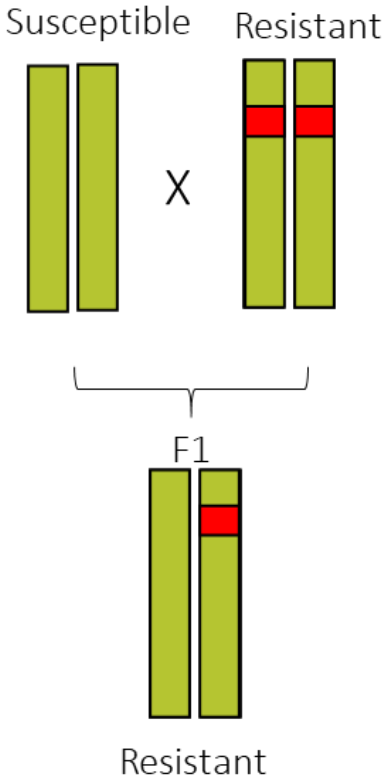
Nature to the rescue: The quest for a resistance source

- ~800 wild *Solanum* accessions screened



Nature provides the solution: ToBRFV HR Resistance

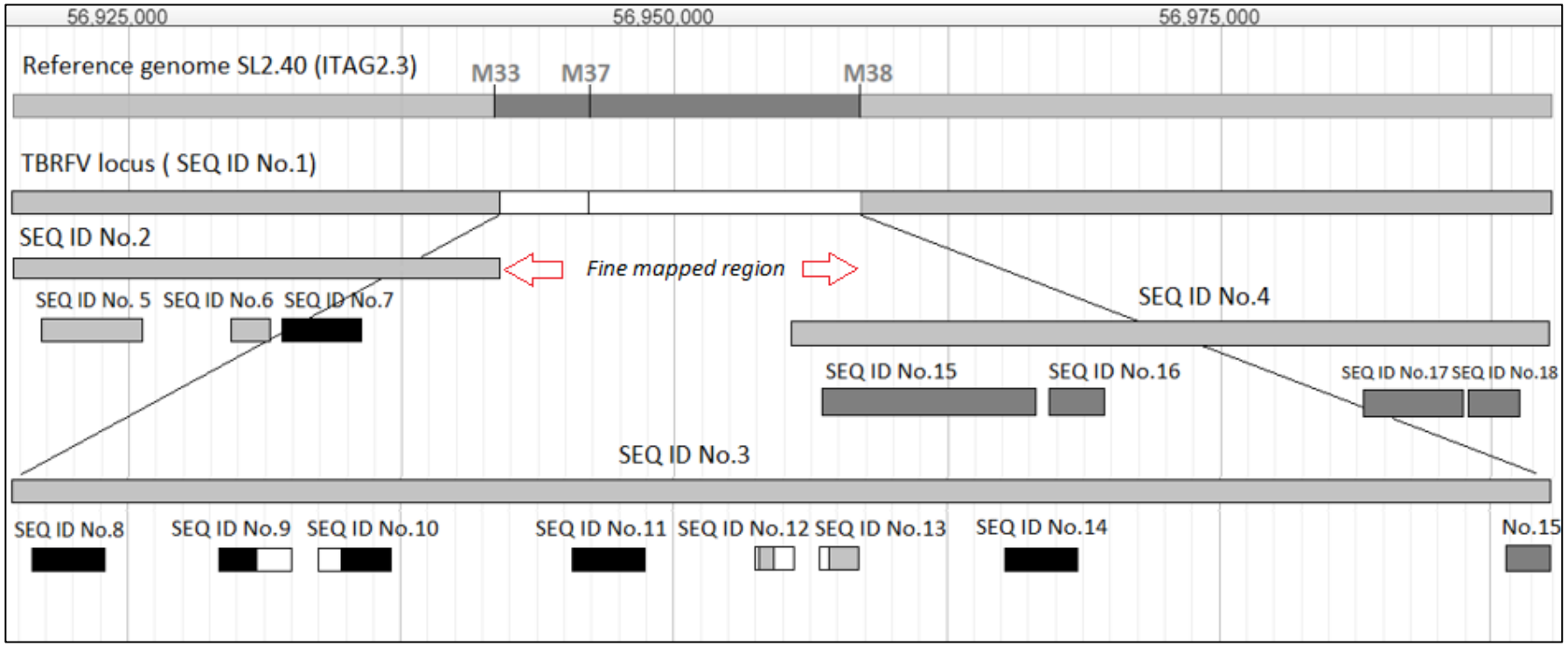
- Found in a wild Tomato accession
- Single gene giving full dominant resistance (like Tm22)
- Introgression in cultivated tomato plants.
- Traditional breeding techniques.

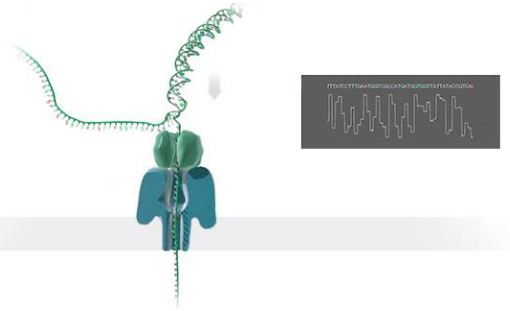


Finding a needle in a haystack followed by hard work to get it in our lines

ToBRFV locus (enabling technology).

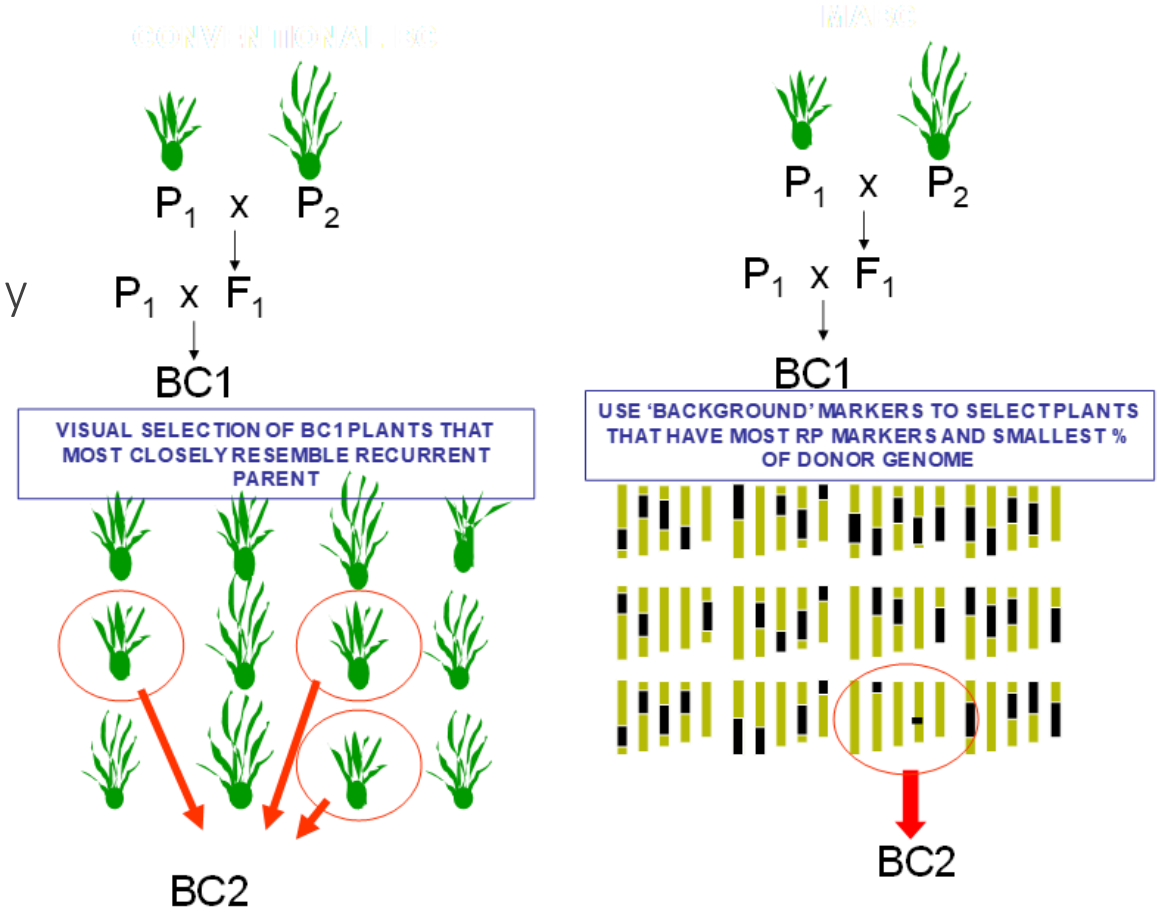
- ONT Sequencing and assembly (Keygene).
- IsoSeq data for further refinement.
- Resistance source contains a region that is 68 kb larger than SL2.40.





Technology allows for fast introgression of the resistance in our varieties by MABC

- MABC - commonly used pipeline at ENZA ZADEN when a new trait is discovered
- Fast introgression with help of molecular markers to quickly introgress a new trait in a (fi) elite parental line
- Basis is classic recurrent backcrossing
  - with the help of molecular markers the highest % recurrent parent (RP) is calculated/selected after each backcross (BC)



From viral detection (2014) to trialing resistant varieties (2020)

In our experiments, the tomato crops remained symptom-free and virus-free

- Resistant in lab test
- Resistant in field tests Jordan 2019-2020 and more recently in Mexico.
- After infection, no virus present in the plant (PCR)
- No symptoms

HR resistance can help eradicate the virus.

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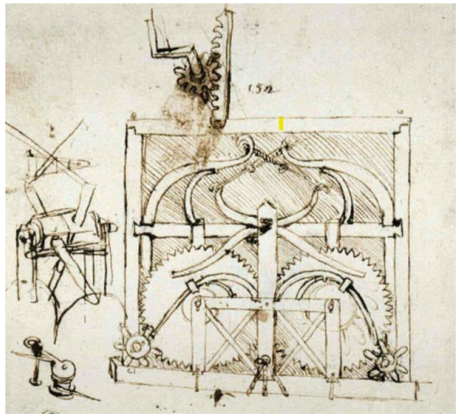
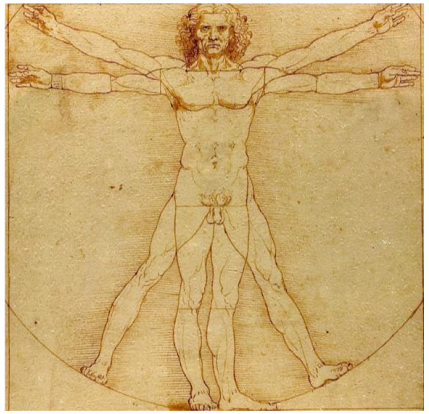
Society – Nature - Technology

Technology allows us to make better use of nature



Fragile trust in technology



Nature poses Challenges but also offers solutions

Together, in the fight against ToBRFV

Stay connected with us, and register at:
www.enzazaden.com/keep-me-informed-ToBRFV

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Regulatory landscape

Japan: Japan Determines Genome Edited Tomato Will Not be Regulated as GE

December 17, 2020 | Attaché Reports (GAIN)

Euroseeds
Embracing Nature

ii. Policy developments in different Countries around the world

Europe:

- Ongoing EU Commission stakeholder consultation (Enza participation)
- Study to be ready in April 2021
- France: the French Minister of Agriculture regarding the products of new breeding techniques. He expresses his support to no longer regulate them as GMO's on EU level.

USA:

- Further clarification on the governmental requirements for commercialisation of GE products