

To Be Host or Not to Be: The Role of Asymptomatic Hosts in the Management of Verticillium Wilt of Potato



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Potato production: World

Potato uses

- Fresh
- Processed products: frozen, dehydrated, chips&snacks, flour, starch
- Distilled beverages: vodka
- Glue & adhesives
- Bio-fuel
- Animal feed
- Seed: Netherlands, UK



5. US
~20 million tons

China
on tons

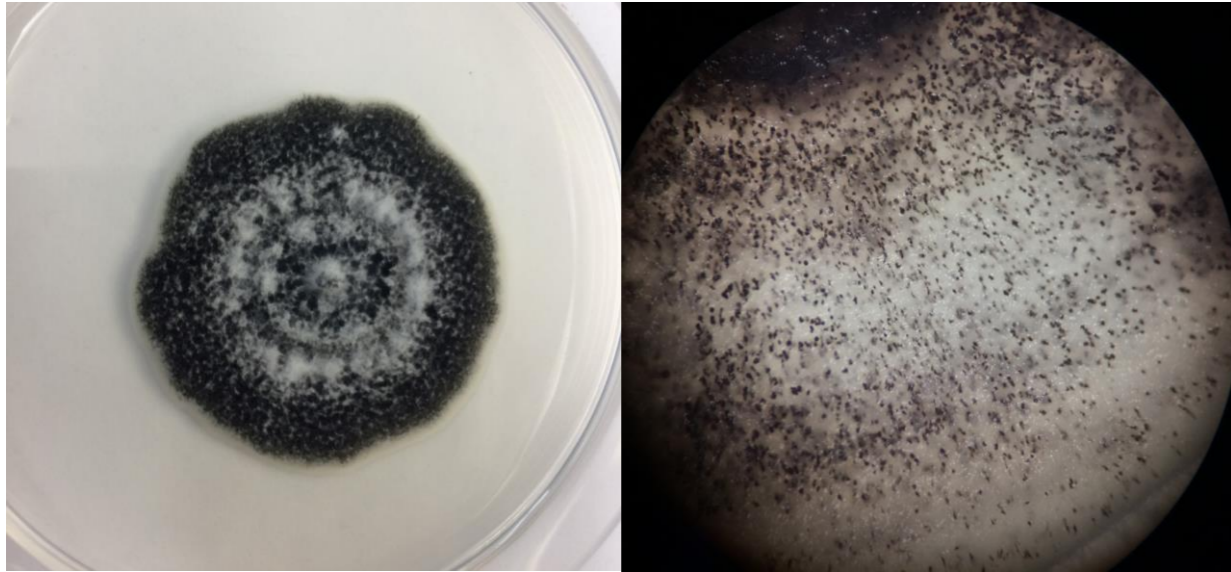
Potato production: U.S.

ID + WA = 1/2 of total US production



PA ~150,000 lb vs. WA ~ 10 million lb

Verticillium wilt of potato: the pathogen

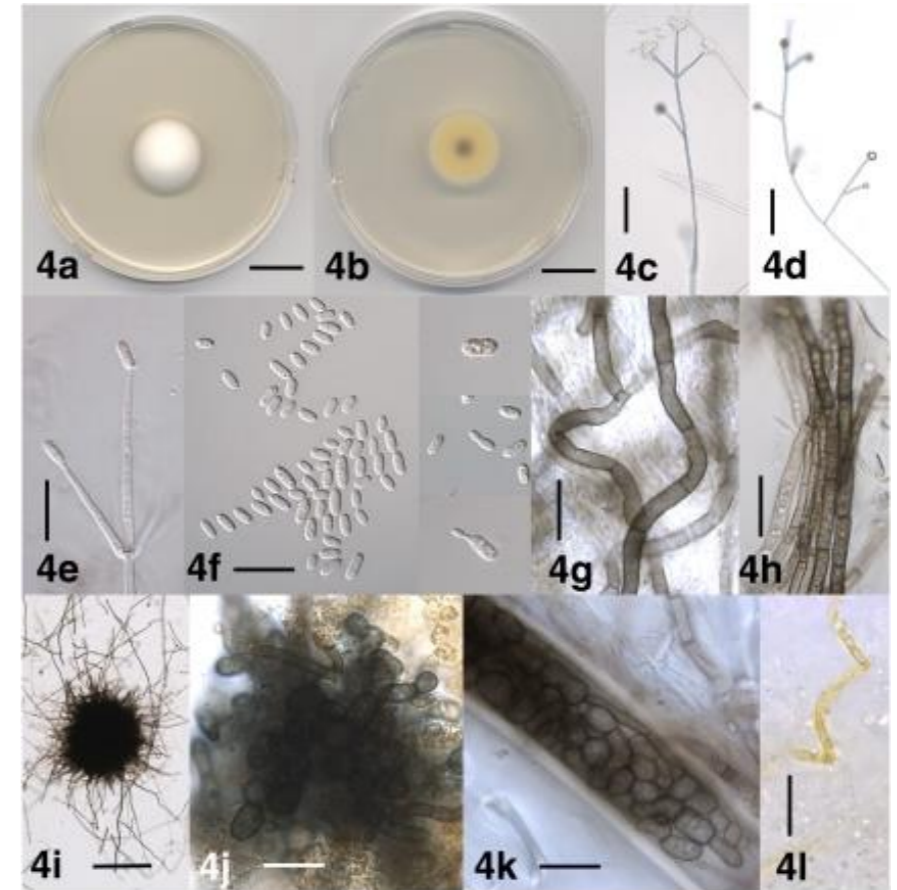


V. dahliae culture on PDA

V. dahliae microsclerotia on PDA

Verticillium dahliae

- Global distribution, especially temperate climates
- Susceptible host range ~ 300 plant species, mainly dicotyledonous
- Microsclerotia, mycelium infecting plant material
- Disseminated with soil, water, plant material, equipment, etc.

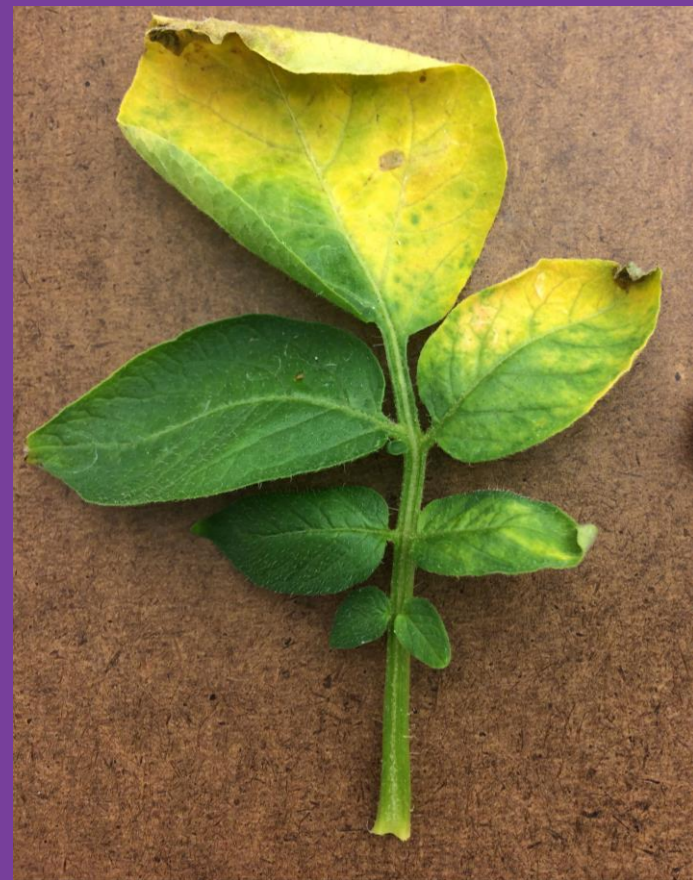


V. albo-atrum culture, hyphae, conidia, microsclerotia, melanized mycelium

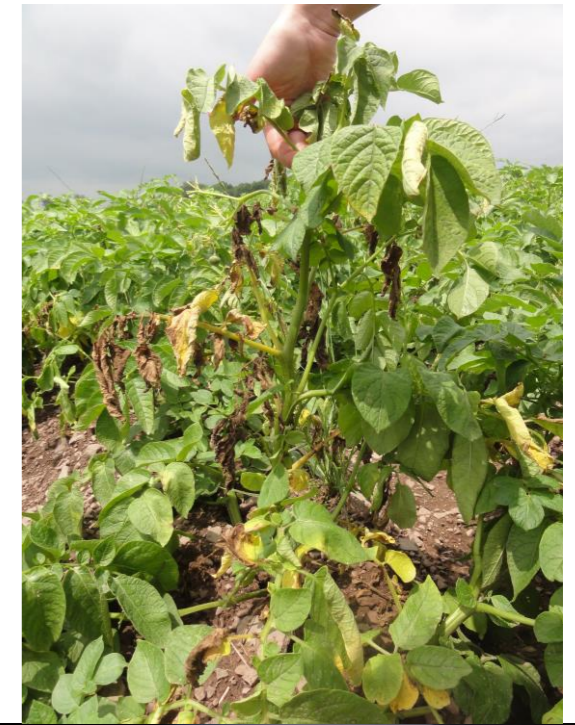
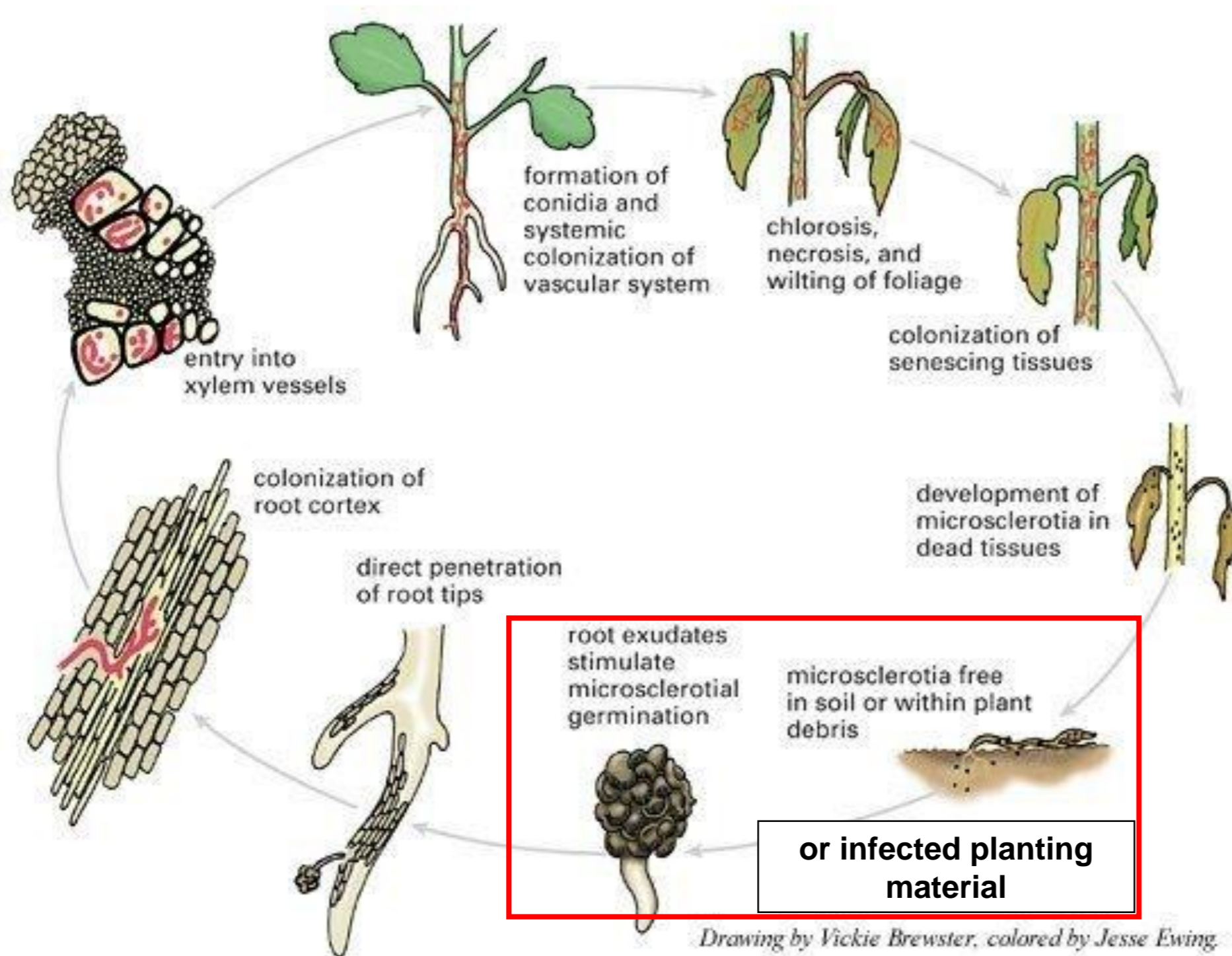
Verticillium albo-atrum

- Canada, Germany, UK, northern Europe and northern USA
- Susceptible host range: potato
- Microsclerotia and melanized mycelium
- Disseminated same as *V. dahliae*

Verticillium wilt of potato: symptoms



Verticillium wilt of potato: disease cycle



Verticillium wilt of potato: integrated disease management



Soil disinfestation: fumigation, solarization, biofumigation.



5-10 years crop rotations with non-susceptible hosts to separate disease cycles



- Moderate host resistance
- Cultural practices to improve plant and soil health

Management of Verticillium wilt using crop rotation

- Traditional crops rotated with potatoes: **monocots, some leguminous.**

** Examples of rotation sequences in PA potato farms:

potato -> rye / timothy (cash crop) -> sudangrass (green manure)

potato -> wheat / soybean (cash crop) -> corn (cash crop)

- Growers and researchers have had inconsistent results:

Inoculum levels, disease incidence and severity depending on the rotational species and the field

- **Asymptomatic secondary hosts** (e.g. rotational crops and weeds): barley, oat, rye, wheat, corn, mustard species, sorghum, sudangrass, pea; weeds such as *Amaranthus* spp., *Chenopodium album*, *Datura stramonium*, *Solanum nigrum*, *Plantago major*, *Portulaca oleraceae*, etc.
- **Unknown diversity** of *V. dahliae* populations associated with asymptomatic hosts.

Variation of pathogenicity and genetic diversity of *V. dahliae* populations

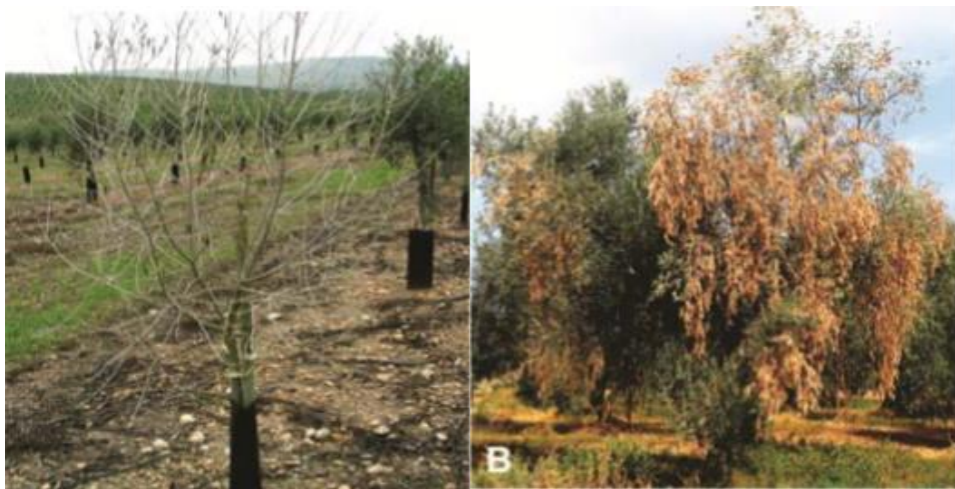
- Vegetative Compatibility Groups (VCGs)
- Defoliating (D) and Non-Defoliating (ND) Pathotypes
- Races 1 and 2 in tomato, lettuce



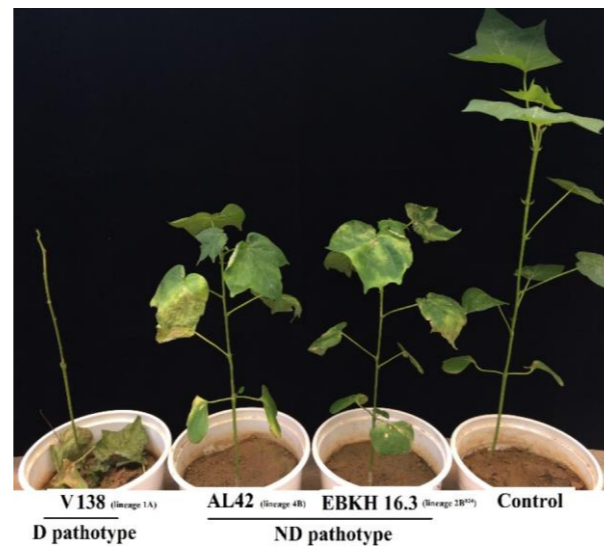
4B vs. 4A in potato



1A vs. 2A vs. 2B³³⁴ in artichoke



pathotype D vs. ND in olive

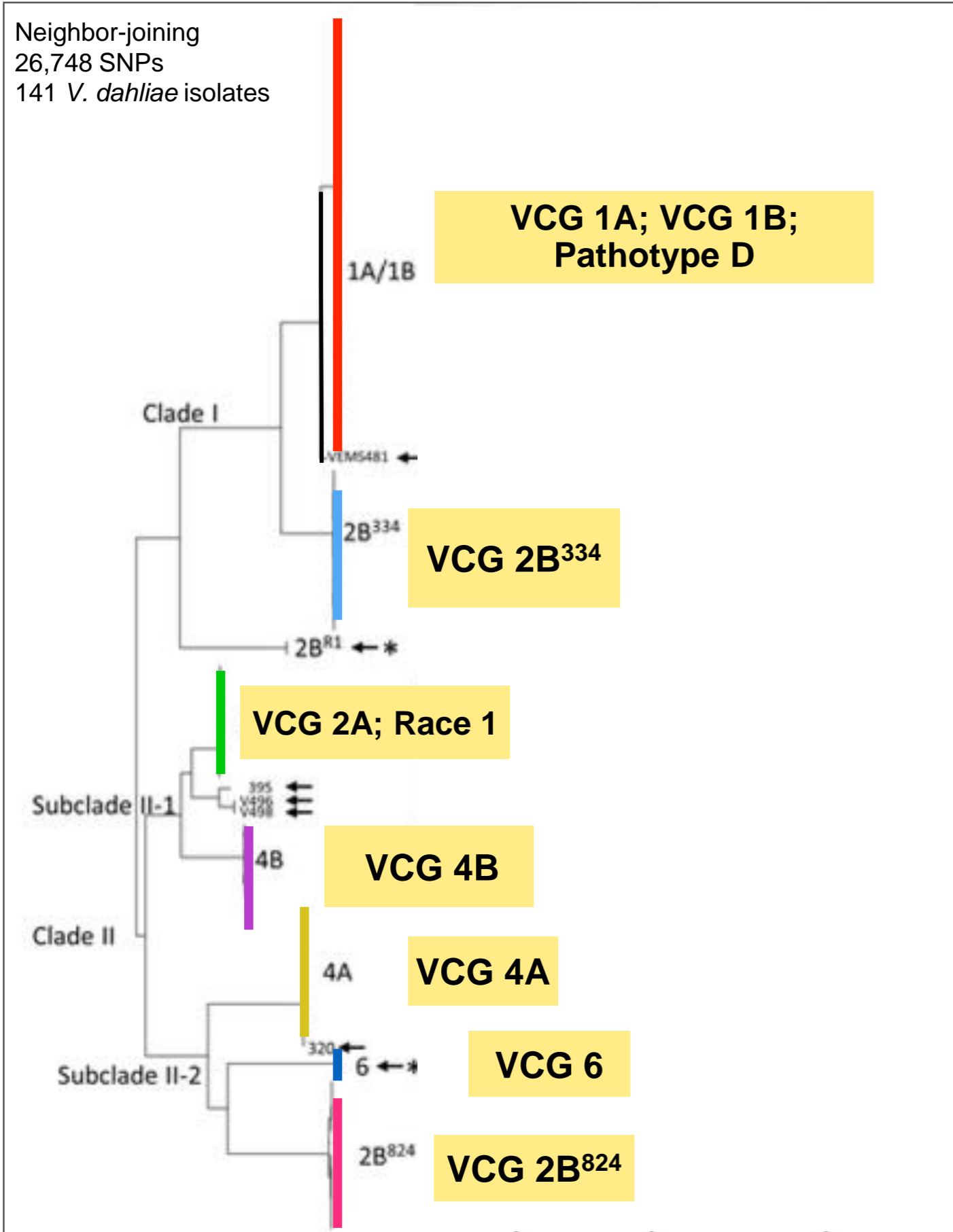


pathotype D vs. ND in cotton



Race 1 resistant tomato cv vs. susceptible cv

Variation of pathogenicity and genetic diversity of *V. dahliae* populations



- Clonal lineages correlates with VCGs
- Pathotypes and races correlates with clonal lineages

Research goal and hypothesis

Research goal

To improve long-term management of Verticillium wilt of potato by understanding the interactions of *V. dahliae* populations with plant hosts in potato agroecosystems.

Hypothesis

V. dahliae populations can infect asymptotically rotational crops and weeds. These hosts can serve as inoculum reservoirs for subsequent Verticillium wilt epidemics.

Research approach

- Chapter 1: Diversity and genetic structure of *Verticillium dahliae* populations infecting symptomatic and asymptomatic hosts in a potato field
- Chapter 2: *Verticillium dahliae* clonal lineages and their secondary hosts in PA and Israel potato fields

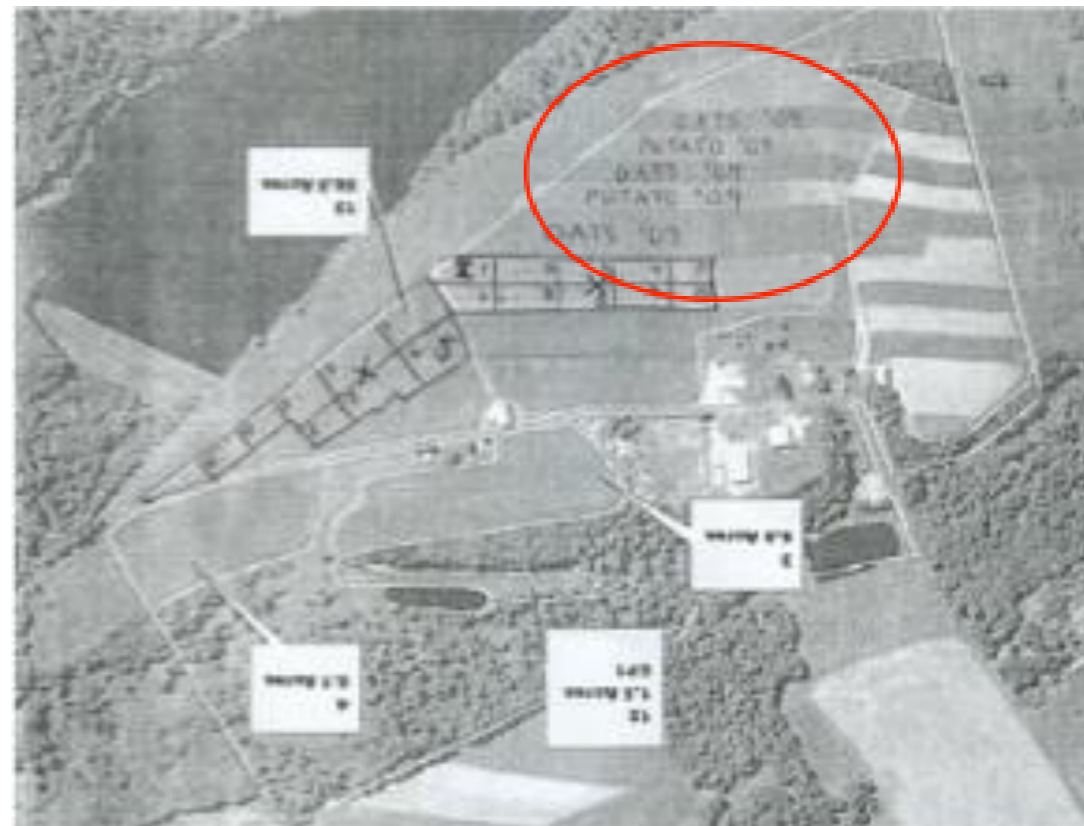
Chapter 1



Potato

Oat

Schuylkill, PA



Hypothesis

- Genetic diversities of the pathogen populations infecting the different host genotypes are different due to differential host responses to the disease.
- The asymptomatic rotational host, oat, serves as a reservoir for pathogenic lineages to potato, such as 4A and 4B.

Objectives

- To assess the genetic composition and structure of the *V. dahliae* populations isolated from three host species cultivated under rotation in the same commercial field, potato 'Snowden', potato 'Reba', and oat 'Armor'.
- To assess the pathogenicity of the *V. dahliae* isolates to potato and oat

Characterization of *V. dahliae* populations

- 10 microsatellite (SSR) markers that are polymorphic in *V. dahliae*
- Mating type: 1-1 and 1-2 idiomorphs
- VCG: nitrate non-utilizing (*nit*) mutants



Positive complementation between *nit* mutants from a same VCG



Potato 'Snowden'
Susceptible
N = 30

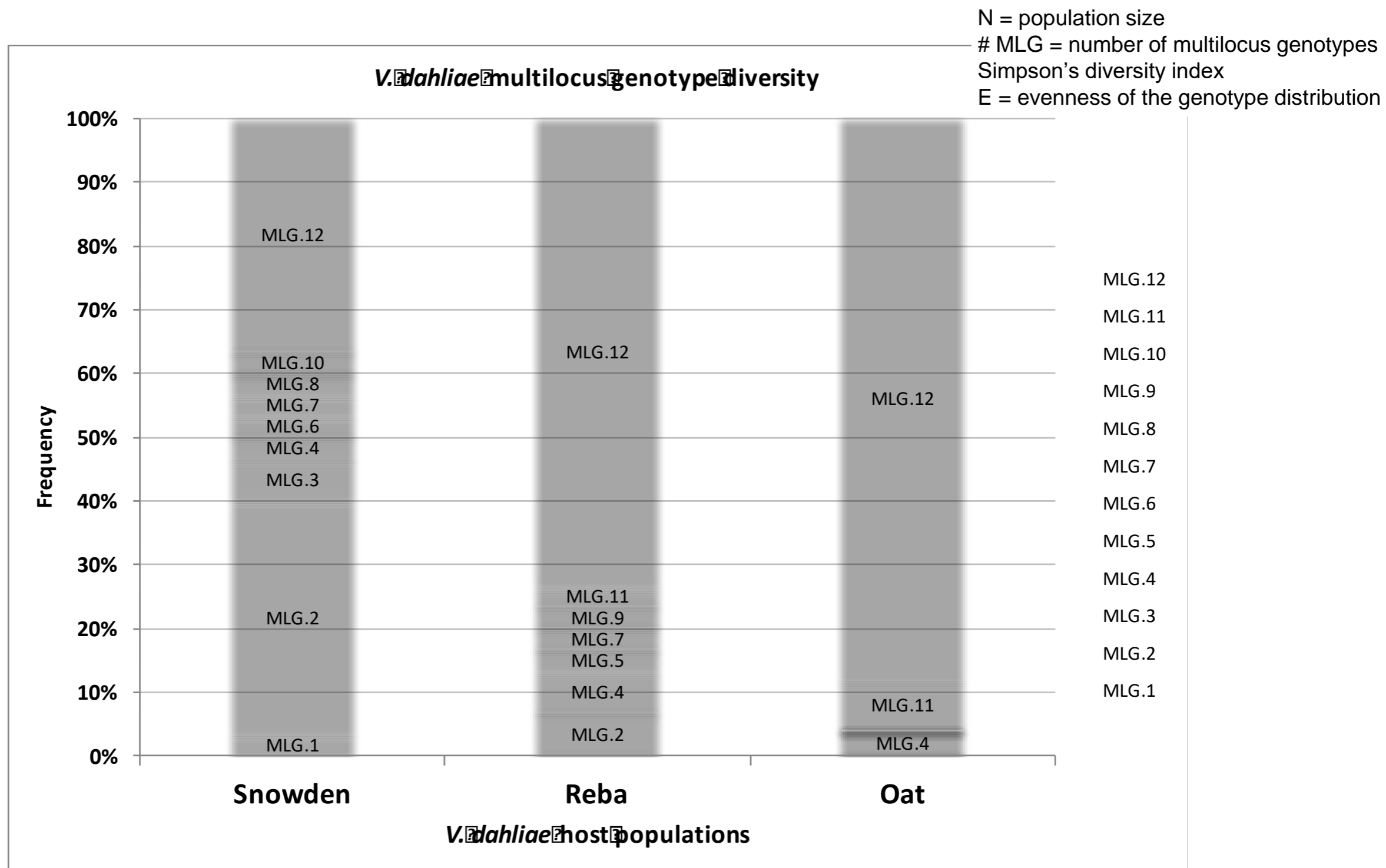


Potato 'Reba'
Moderately susceptible
N = 30



Oat 'Armor'
Asymptomatic
N = 25

Characterization of *V. dahliae* populations: SSR multilocus genotypes



Susceptible
 N = 30
 # MLG = 9
 Simpson's = 0.72
 E = 0.65

Moderately susceptible
 N = 30
 # MLG = 7
 Simpson's = 0.45
 E = 0.44

Asymptomatic
 N = 25
 # MLG = 3
 Simpson's = 0.22
 E = 0.50

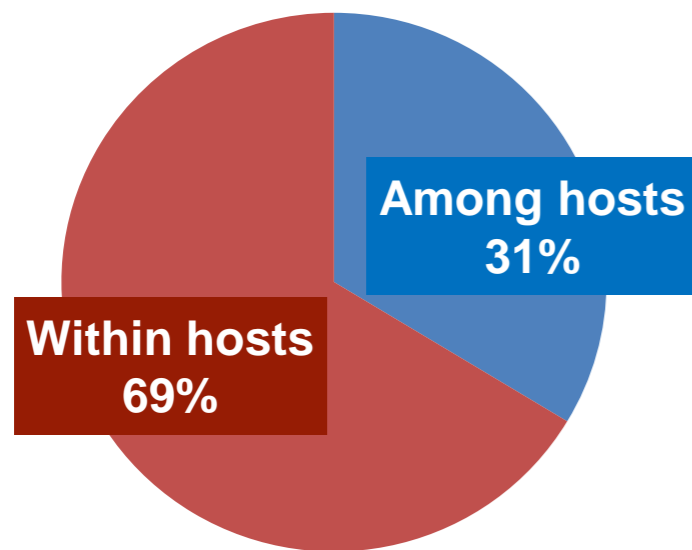
Characterization of *V. dahliae* populations: VCGs and mating type

- **Mating type1_2** (3 isolates that have unknown mating type)
- VCGs found among *V. dahliae* isolates: **2A, 4A, 4B**
- Each MLG was associated with one VCG, but one VCG has multiple MLGs

VCG	MLGs	Host
4B	12, 11, 10	Snowden, Reba, Oat
4A	2, 3, 7, 6, 8	Snowden, Reba
2A	4, 1, 5	Snowden, Reba, Oat

Characterization of *V. dahliae* populations: population genetics analysis

Percentages of Molecular Variance



Phi = 0.3085

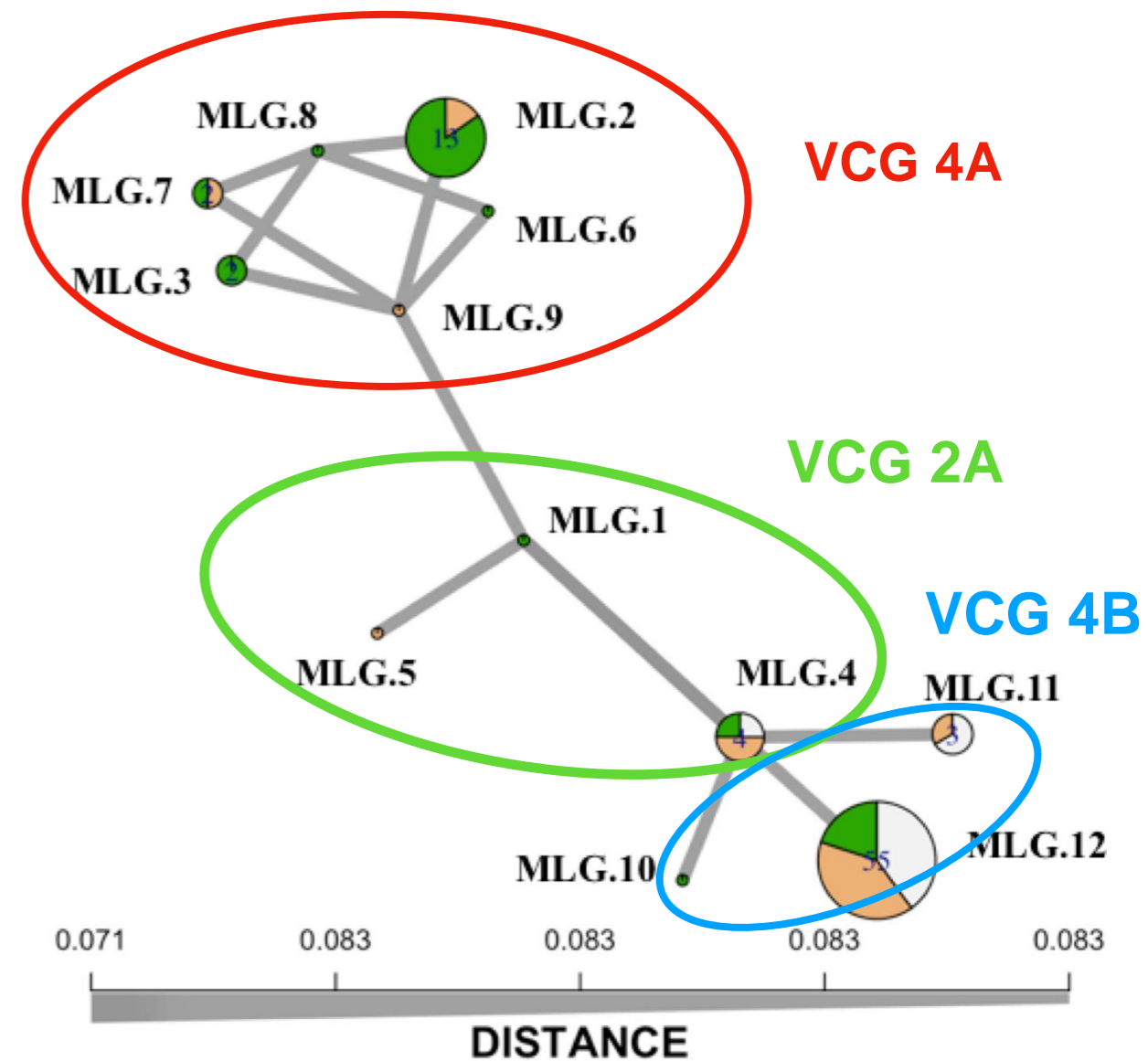
p-value = 0.001

Analysis of Molecular Variance (AMOVA)

Microsatellite allele data

85 *V. dahliae* isolates

POPULATION



Minimum Spanning Network (MSN)

Microsatellite allele data

85 *V. dahliae* isolates

Pathogenicity of *V. dahliae* populations

Hypothesis

- Oats are infected by VCG 4B but not by 4A *V. dahliae* isolates
- *V. dahliae* isolates that infect oats are pathogenic to potato
- 4A isolates are more virulent than 4B to potato

Experiment

- Oat isolate: MLG.12, VCG 4B
- Potato isolate: MLG.2, VCG 4A
- Control (+), VCG 4A isolate from collection pathogenic to potato
- Control (-), mock



Pathogenicity of *V. dahliae* populations

Potato 'Snowden'



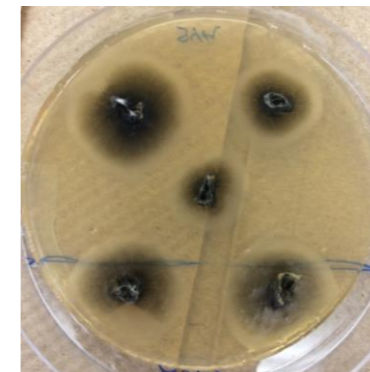
Control -



4A



4B



V. dahliae infected potato stem samples on semi-selective nutritional medium

- *V. dahliae* 4B and 4A infected potato
- *V. dahliae* 4A is more virulent than 4B

Oat 'Armor'



- *V. dahliae* 4B infected oats
~15% incidence (6-30% incidence in field)
- *V. dahliae* 4A did not infect oats

Oat stem samples infected by *V. dahliae* 4B on semi-selective nutritional medium

Chapter 1: conclusions and implications

- *V. dahliae* diversity in susceptible hosts (potato) >>> asymptomatic host (oats)
- **VCG 4A** only infects potato cultivars
- **VCG 4B** infects oats asymptotically and is pathogenic to potato
- **VCG 4A is more virulent** to potato than 4B.

Chapter 2

Hypothesis

- Some genotypes of *V. dahliae* infect rotational hosts and weed species, even though they do not express symptoms of Verticillium wilt.
- Pathogen diversity maintained by asymptomatic hosts may have the potential to impact epidemics in the subsequent susceptible crops.

Objective

To characterize the genetic diversity and structure of *V. dahliae* populations infecting asymptomatic secondary hosts (rotational crops and weed species) in Israel and PA

V. dahliae populations associated with potato agroecosystems from PA and Israel



U.S.

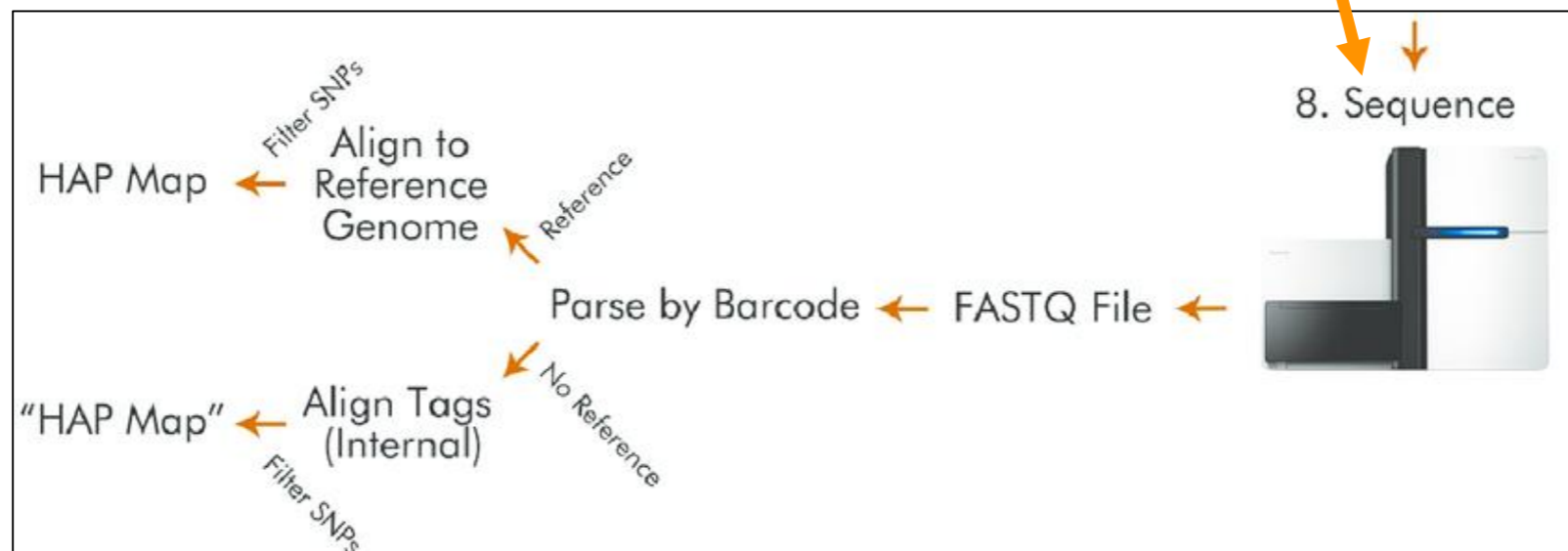
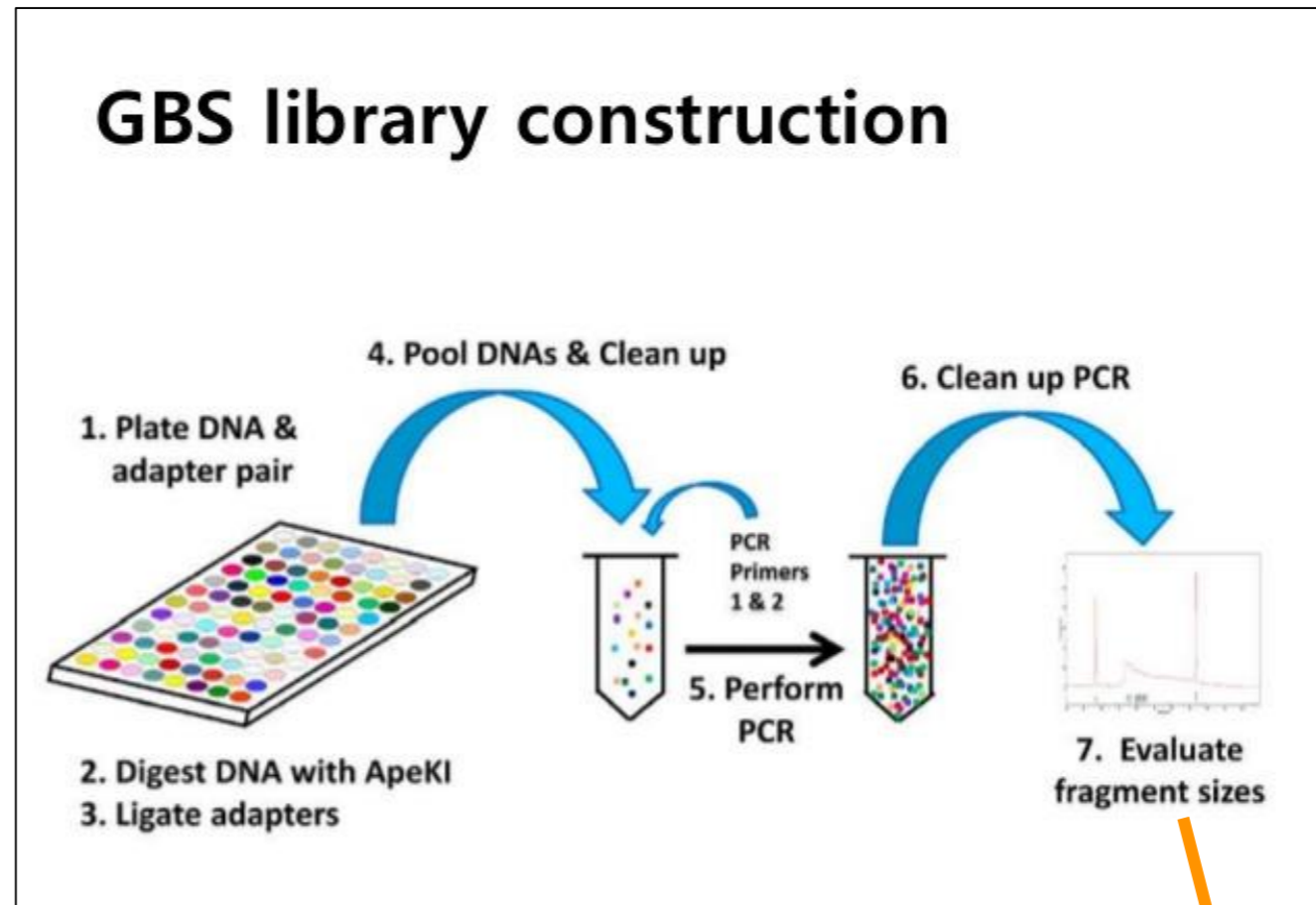
- Pennsylvania
- 65 *V. dahliae* isolates
- potato cultivars and oat



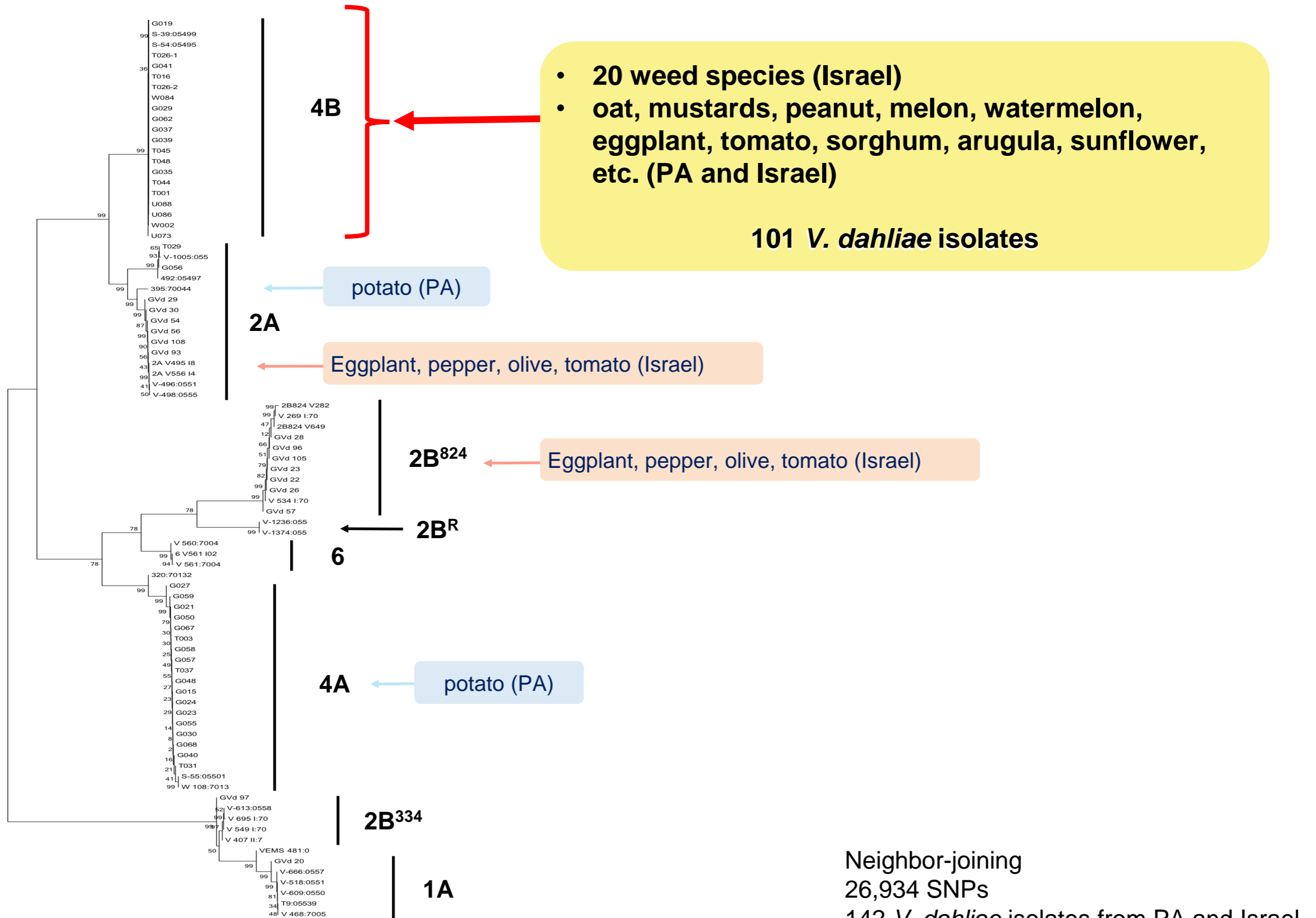
Israel

- Centre and South
- 77 *V. dahliae* isolates
- Rotational crops (sorghum, peanut, sunflower, mustards, melon, eggplant, arugula, etc.) and 20 weeds.

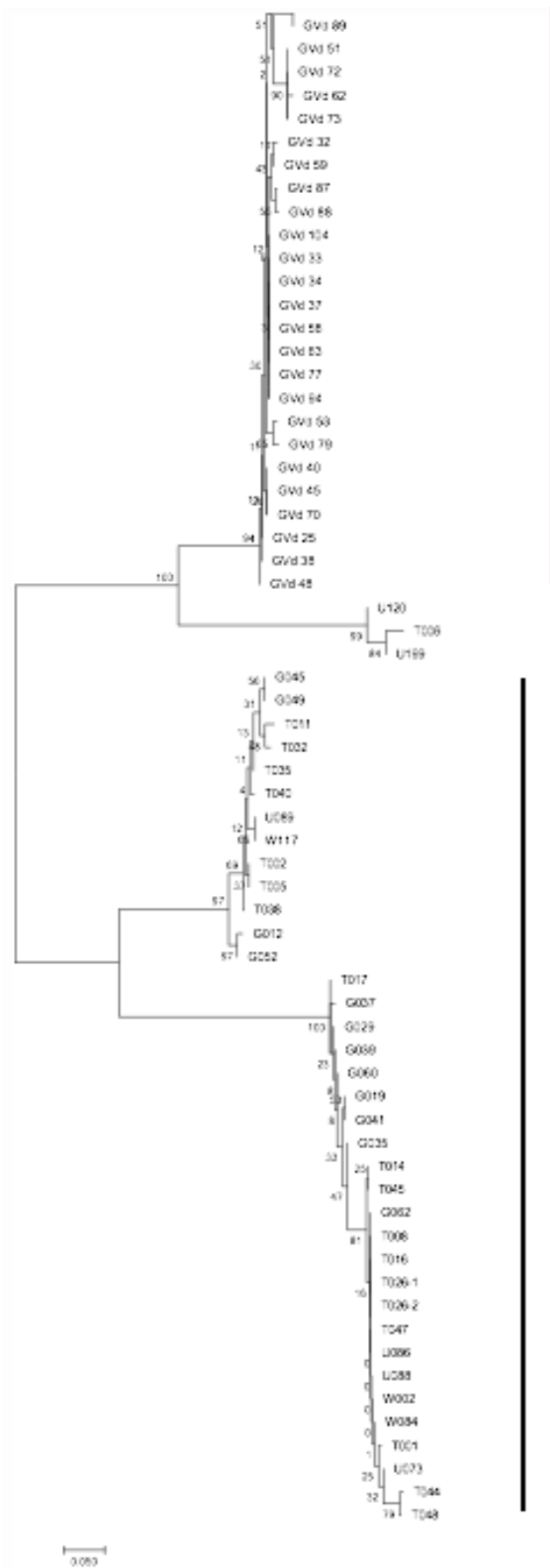
Genotyping-by-sequencing, GBS



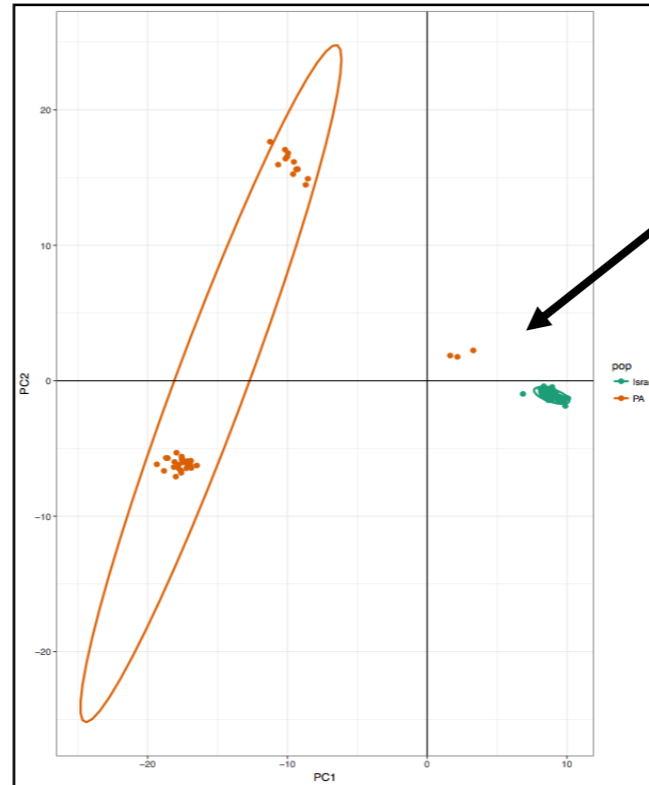
V. dahliae populations associated with potato agroecosystems from PA and Israel



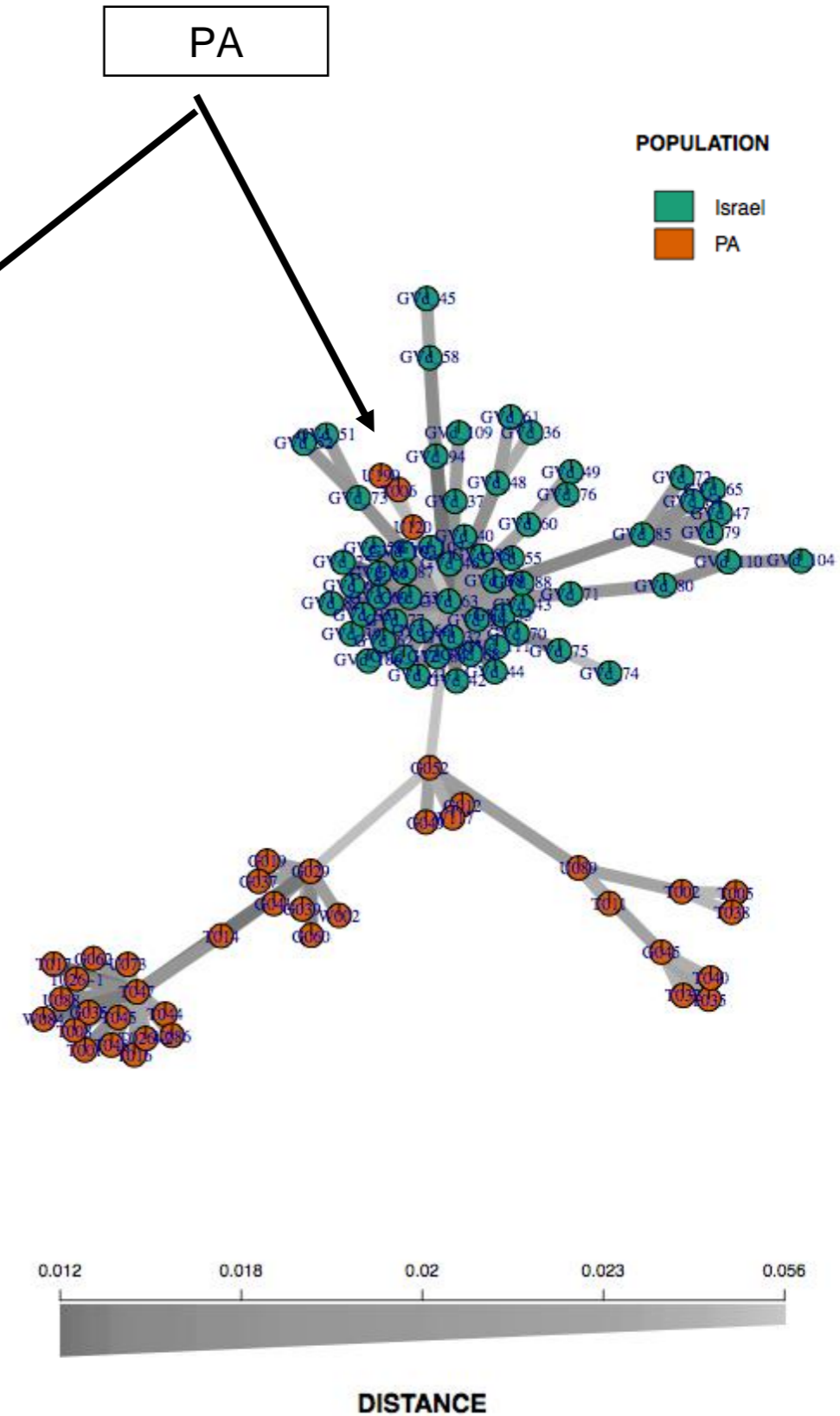
4B lineage - populations from PA and Israel



Neighbor-joining
5,335 SNPs
101 *V. dahliae* isolates



Principal Component Analysis (PCA)
5,335 SNPs
101 *V. dahliae* isolates



Minimum Spanning Network (MSN)
5,335 SNPs
101 *V. dahliae* isolates

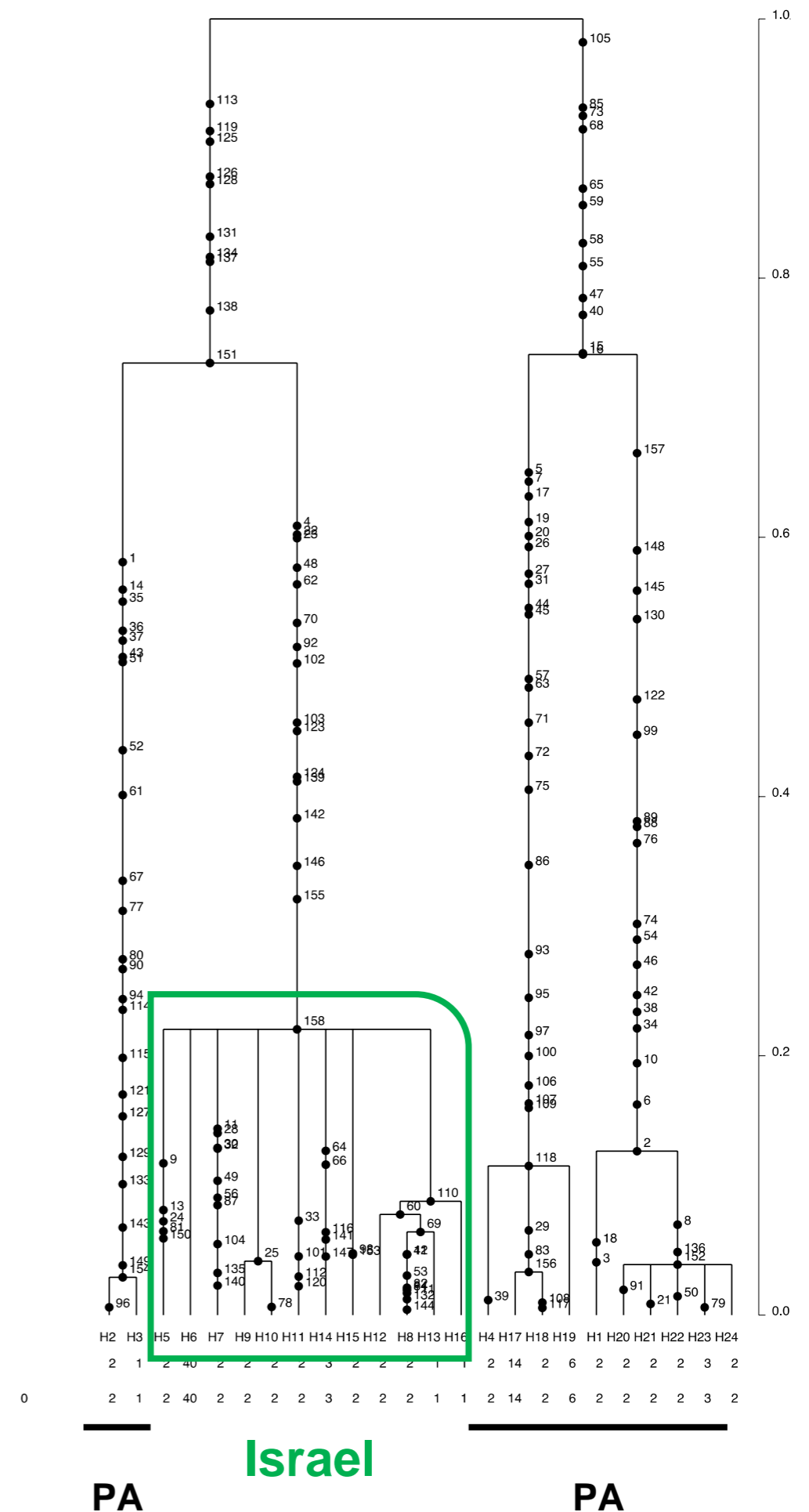
4B lineage - populations from PA and Israel

Coalescent genealogy:

- **174 SNPs** with no missing data
- **101 individuals**

Results:

- Multiple origins of PA isolates
- Common ancestor for Israel and a PA group
- Evidence for one introduction of 4B population in Israel



Chapter 2: conclusions and implications

- *V. dahliae* isolates infecting **asymptomatic secondary hosts** (rotational crops and weeds) are **lineage 4B (=VCG 4B)**.
- **4B lineage** is known to contain isolates **pathogenic to potato**.
- 4B isolates are genetically different based on their geographical origin.
- Israel 4B population has little genetic diversity and coalescent genealogy suggests at least one introduction in the country, probably from a source common to one PA population.

Next step: current and future work

- Explore more potential asymptomatic hosts: development of a PCR-based protocol to detect and quantify *V. dahliae* 4A and 4B lineages in environmental samples
- Dynamics of *V. dahliae* 4A and 4B inoculum in a potato-oat agroecosystem



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