Population dynamics of aerial and terrestrial populations of *Phytophthora ramorum* in a California watershed under different climatic conditions

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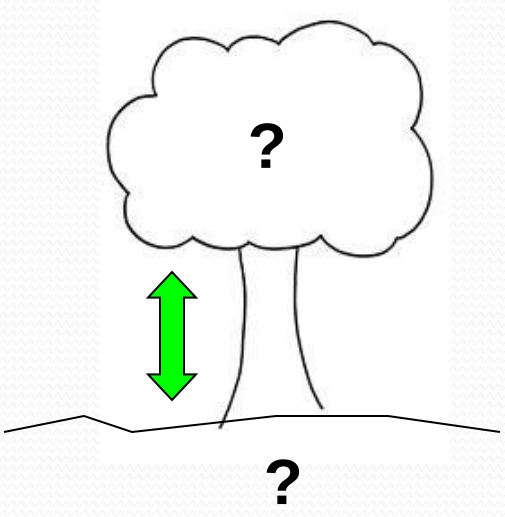
5th Sudden Oak Death Symposium

P. ramorum populations in different substrates

- Soil vs Leaves



- Are populations different?
- Does one feed the other?
- Are they both affected similarly by the climate?

- SSR/Microsatellite markers
  - Ivors *et al.* (2006), Prospero *et al.* (2007), Vercauteren *et al.* (2010)

SFPUC watershed

- Long term infestation – since 2001
- Relatively undisturbed and minimal management

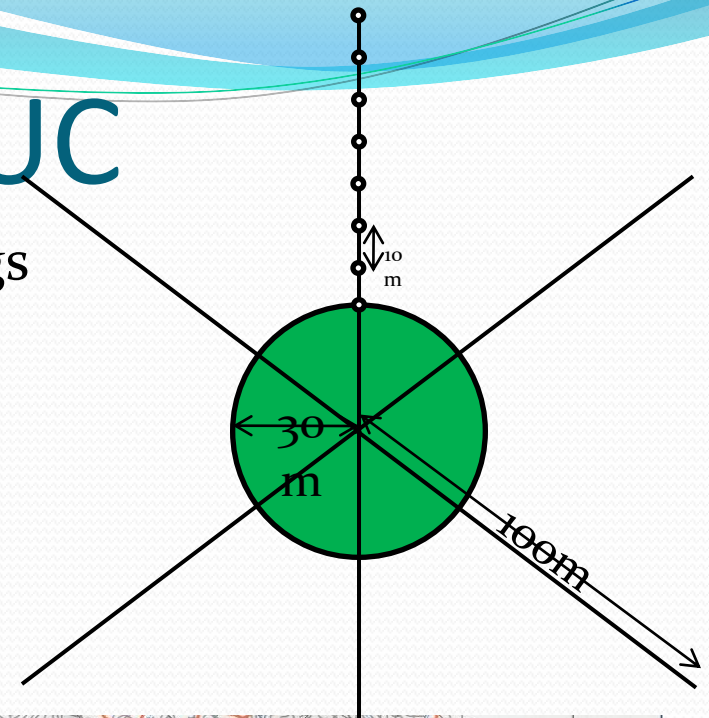
- Plots:
- Tanoaks
- Bay
- Coast Live Oak

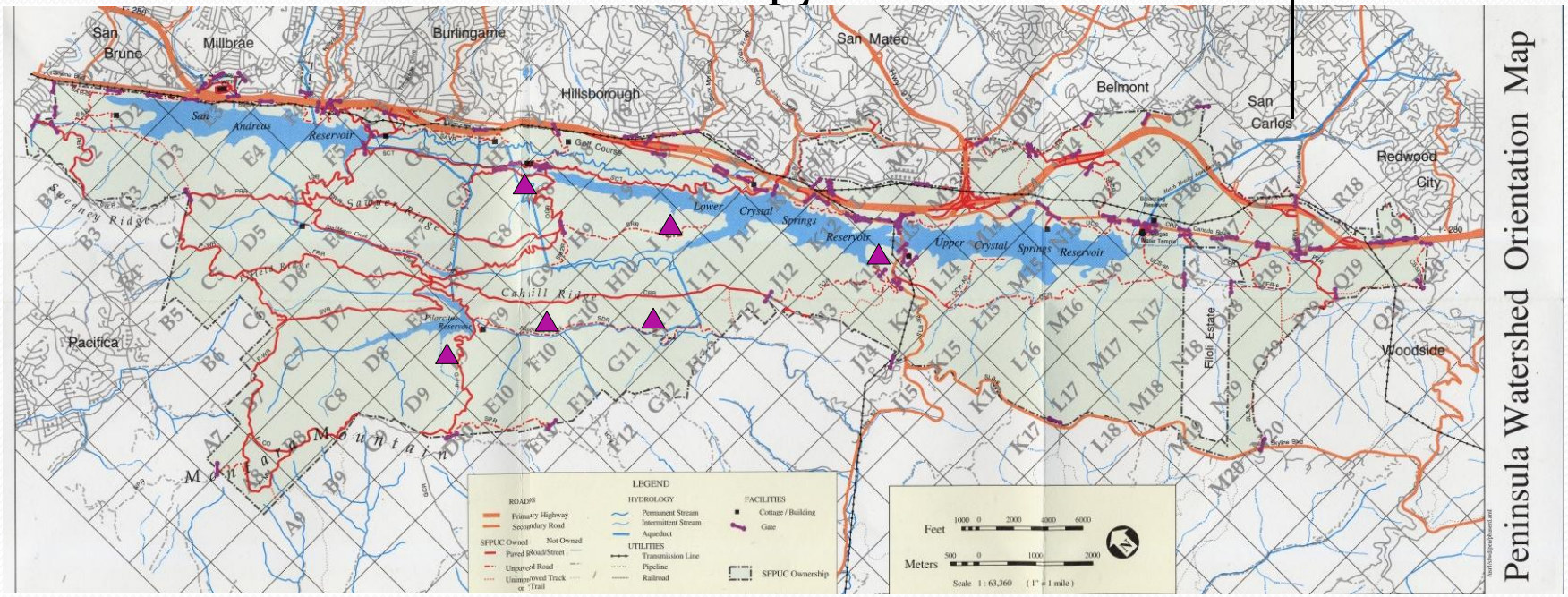
- Intensive sampling over short range distances

- 2009 - Dry
- 2010 - Wet

Soil genotyping SFPUC

- 2 drainages – Pilarcitos and Crystal Springs
- 3 plots within each drainage
- 6 transects at each plot.
- Soil collected from beneath canopy





Soil baiting

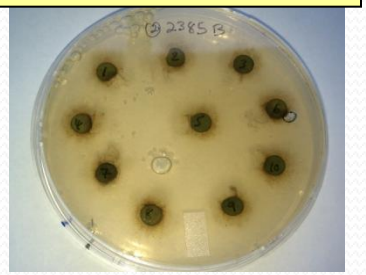
Collect soil

(300+ samples)

Flood with H_2O

Bait with Rhododendron discs

Plate onto PARP+H



Subculture

Grow mycelium in pea broth

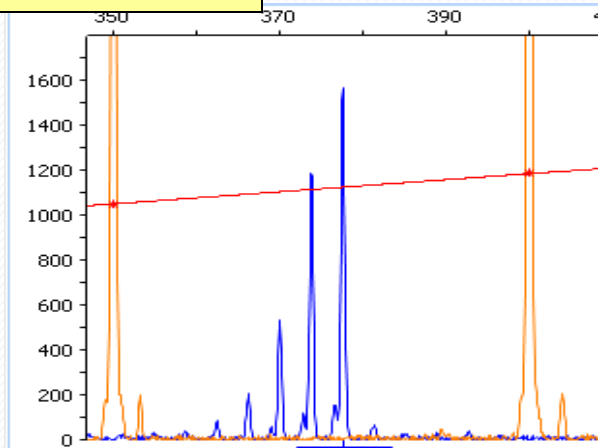
Lyophilise & grind

****NaOH DNA extraction****

Amplify DNA with microsatellite primers for 10 loci

Fragment analysis

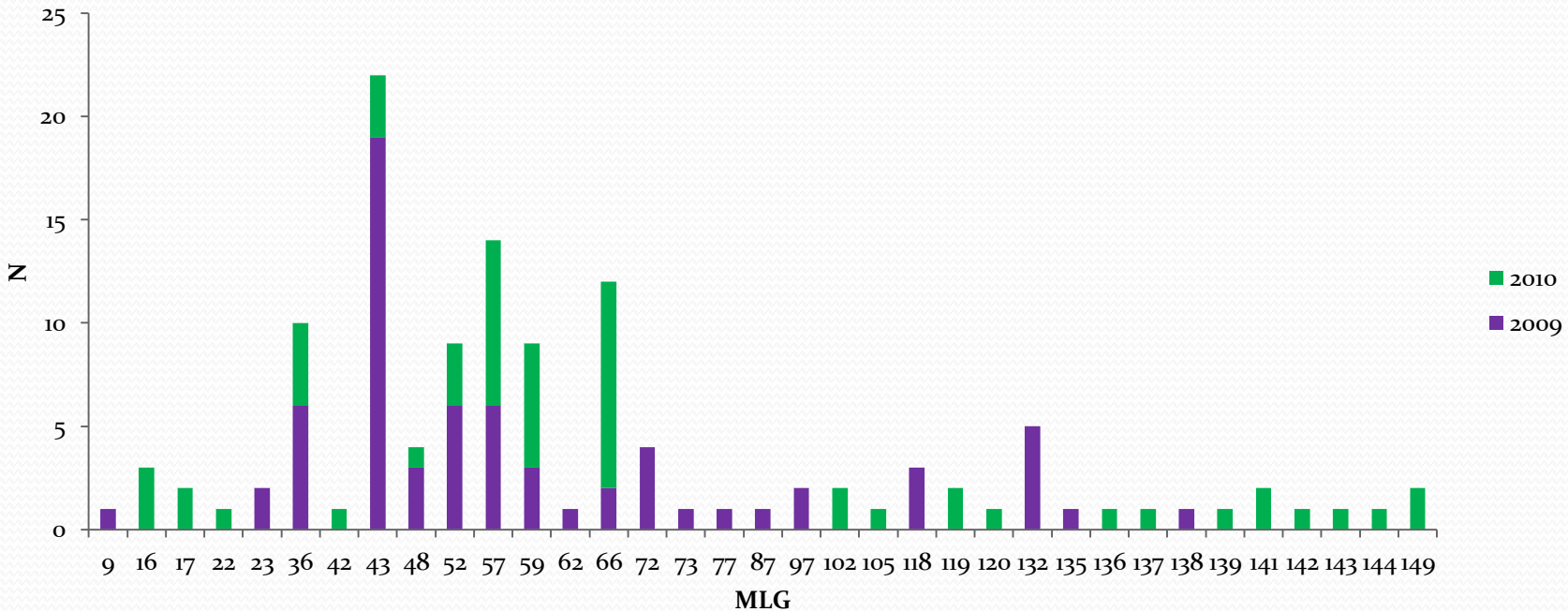
ABI 3730 sequencer/PeakScanner



Prospero, Ivors, Vercauteren

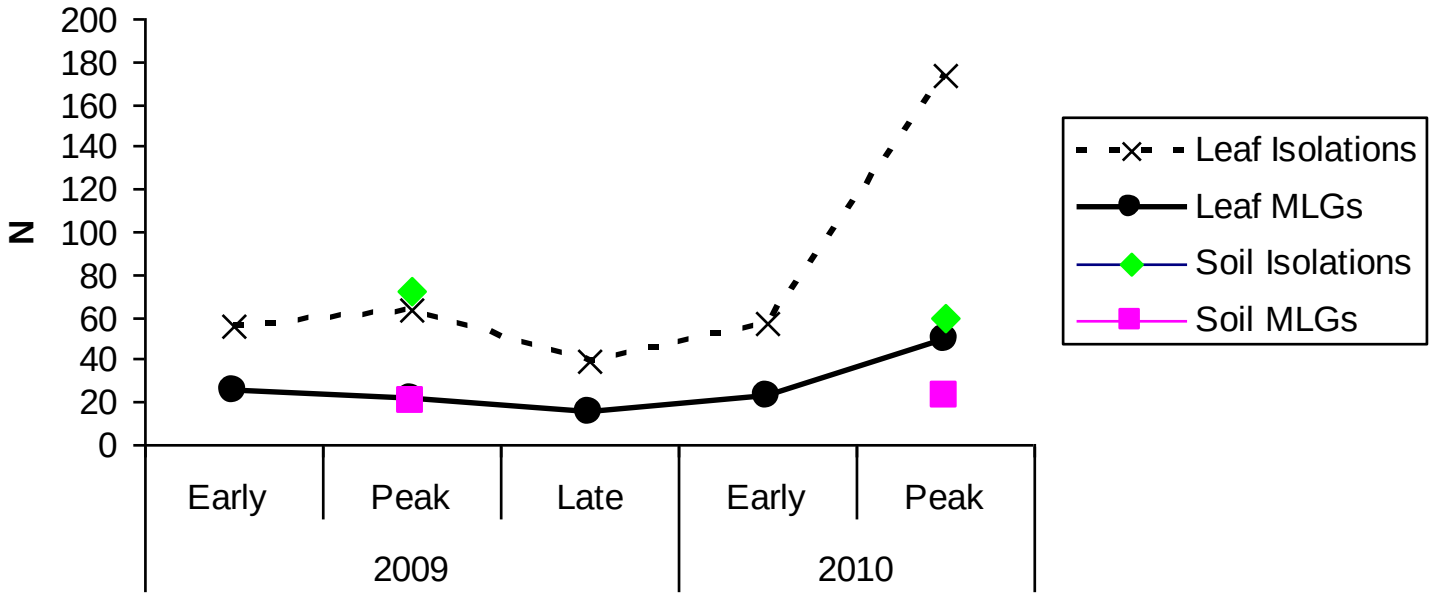
Soil genotypes 2009/2010

- Isolation success: 2009 - 24% 2010 - 19%
- 2009: 20 MLGs 2010: 23 MLGs
- 7 overlapping genotypes
- 48.6% MLGs only represented by one individual



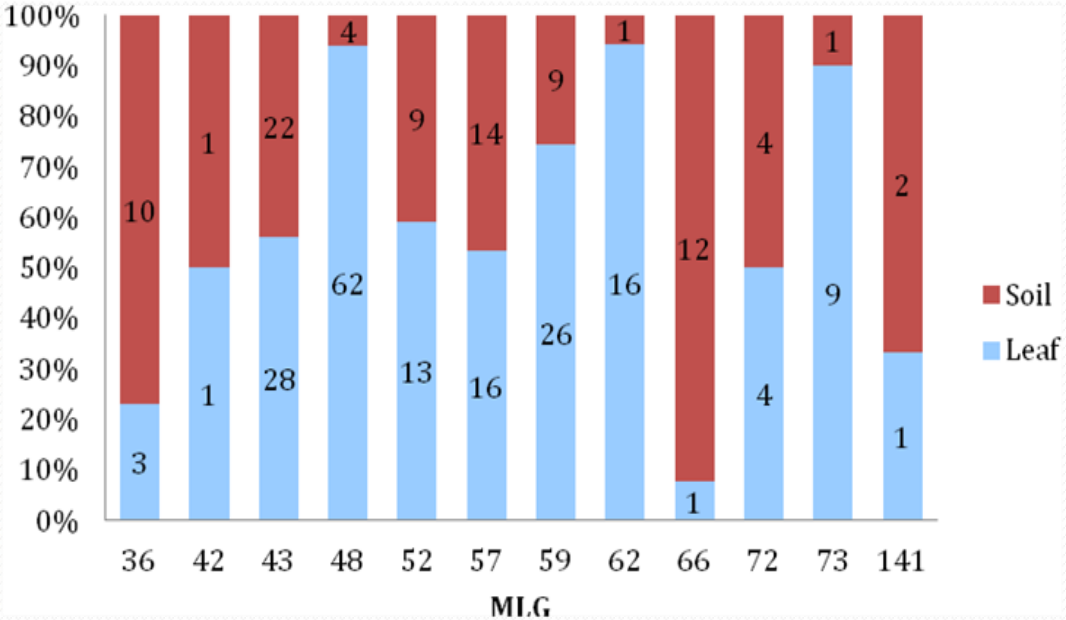
Leaf genotypes

- Peak 2009: 22 MLGs Peak 2010: 49 MLGs
- High proportion are singletons (i.e. only 1 individual)
- Leaf Isolations and MLGs increase from 2009 → 2010
- Soil Isolations and MLGs stay the same/decrease



Shared genotypes in soil/leaves

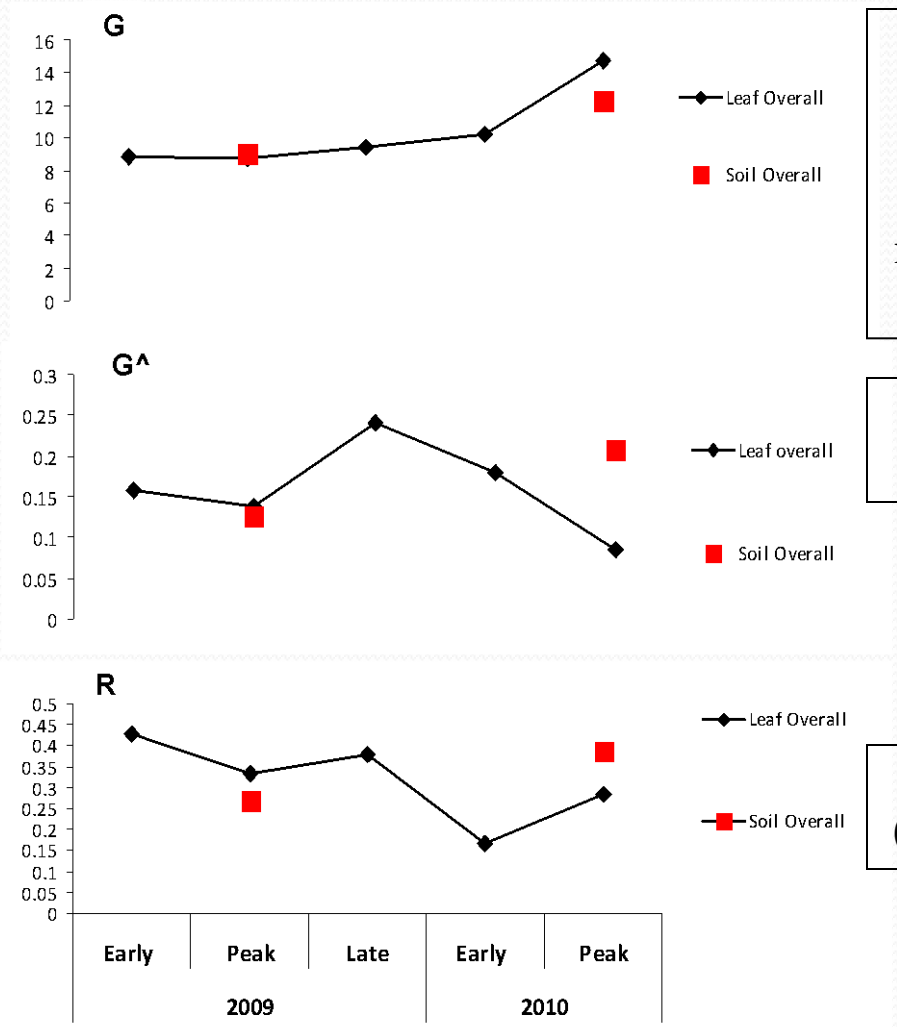
- Very few overlapping MLGs between substrates
- Rank of most abundant MLGs significantly different between years in both soil and leaves



- Relative abundance of overlapping MLGs is significantly different ($p<0.0001$)

Genetic diversity

- *G* increases
- *G^, R – similar pattern*
- Decrease at Peak
- 2010: More genotypes found than in 2009
- But.. Diversity decreases in leaves
- Overabundance of a few genotypes
- **2009 DRY > 2010 WET**
- **2010: Soil > Leaf**



Stoddart & Taylor's *G*
$G = 1/\Sigma p_i^2$
Where pi is the frequency of the *i*th MLG

*G^* = *G*/sample size

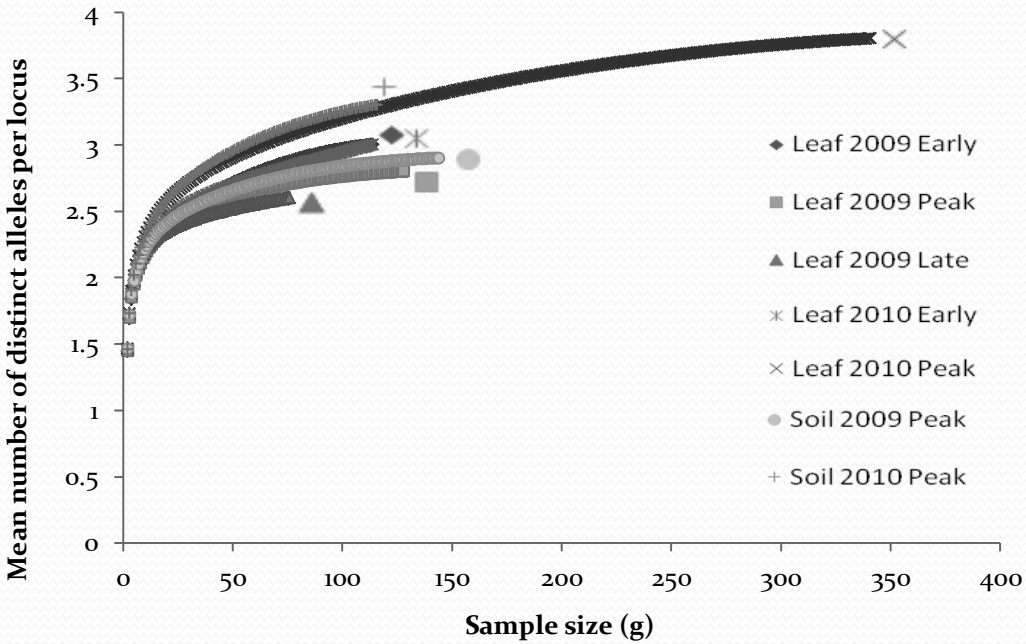
R = (#MLGs-1)/(N-1)

Genetic structuring

- Analysis of Molecular Variance (AMOVA) - Arlequin
- **Time** Leaf: minimal structure in leaves over time

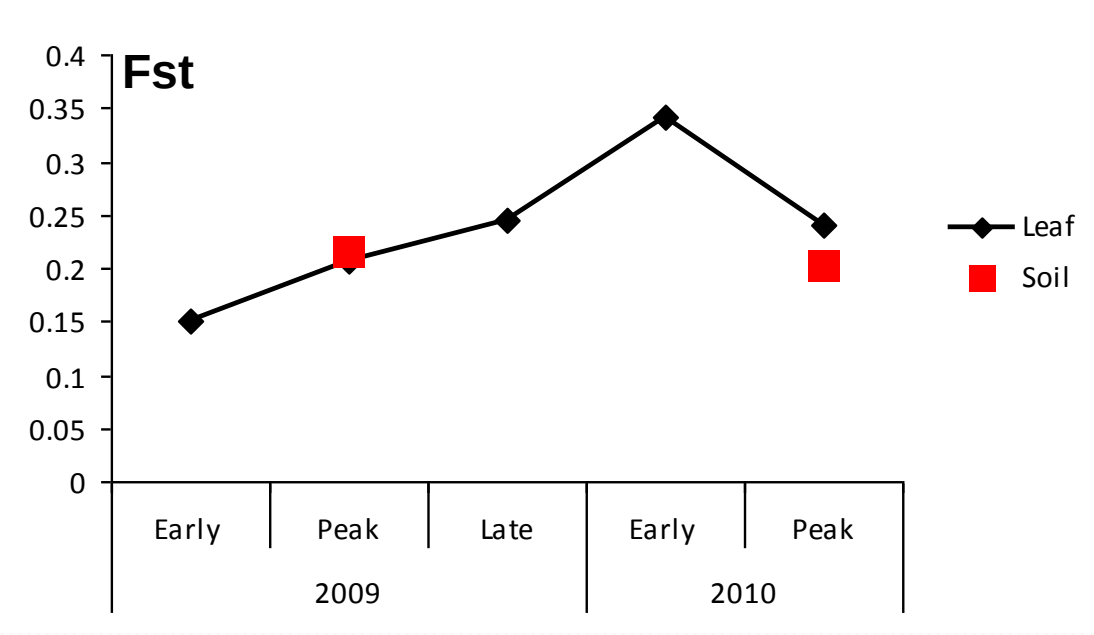
  Soil: significant structure in 2009 vs 2010 – *turnover?*
- Different ability to survive/persist in each substrate
- Carry over of MLGs in leaf but not soil populations

- Same sampling effort
- Varied sample sizes due to different isolation success
- Rarefaction – are we sampling the diversity present effectively?



Structure between plots

- AMOVA
- No structure between 2 drainages
- For individual plots within each timepoint
- Leaf FST: 2009 < 2010
- Soil FST: 2009 ≈ 2010
- Significant structure between the 6 plots
- Limited migration of the pathogen – small founding populations



Soil vs Leaf within plots

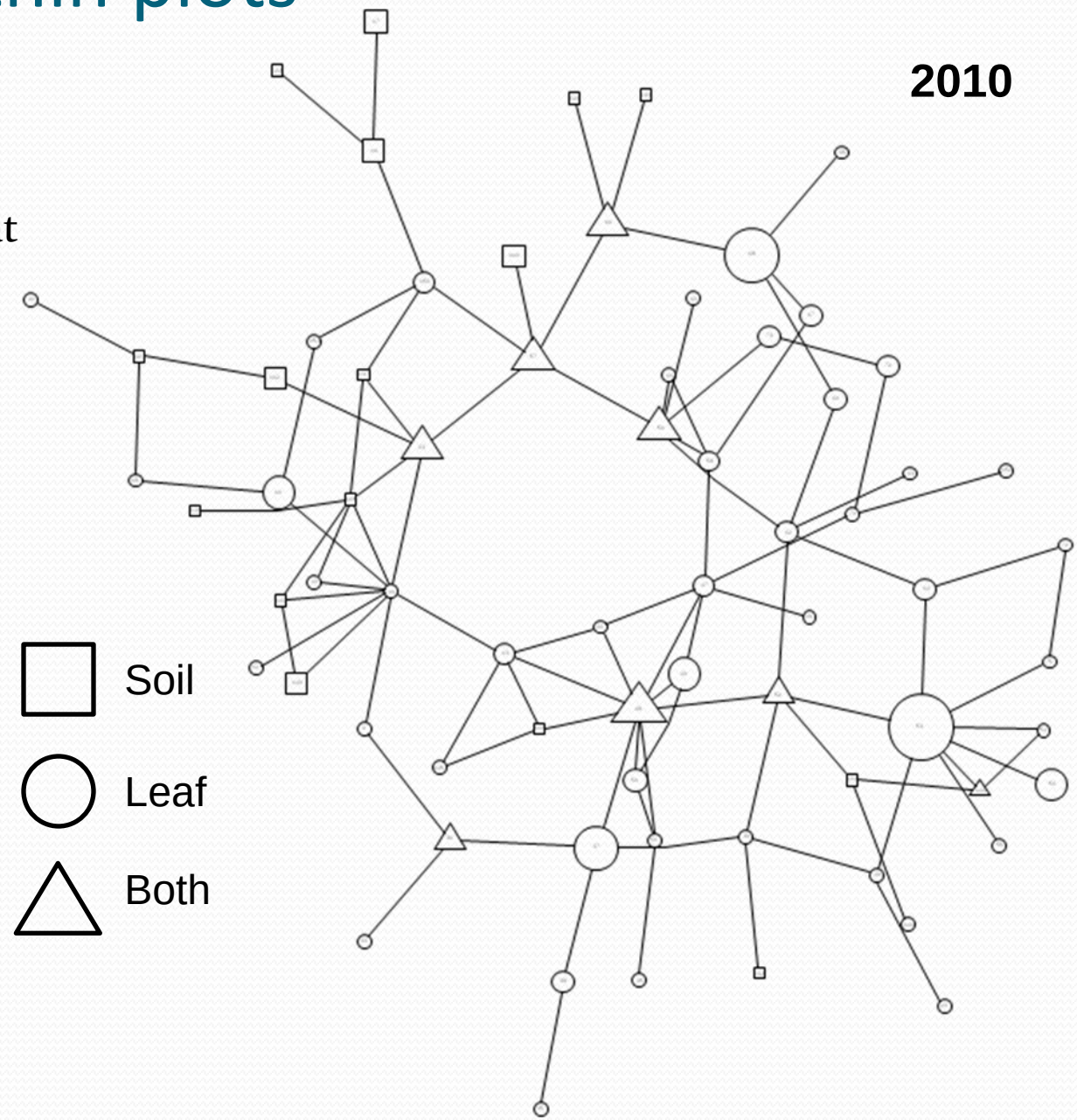
Found genetically distinct populations in soil and leaves within plots, i.e. there was structure with AMOVA even at the same timepoint

Why?
-Little migration between the two pops? seems unlikely

-Different selection pressures in the to substrates

-MLGs may be adapted to different substrates

MSN: soil and leaf genotypes very closely linked and mixed but soil MLGs arising from both leaf and soil progenitors

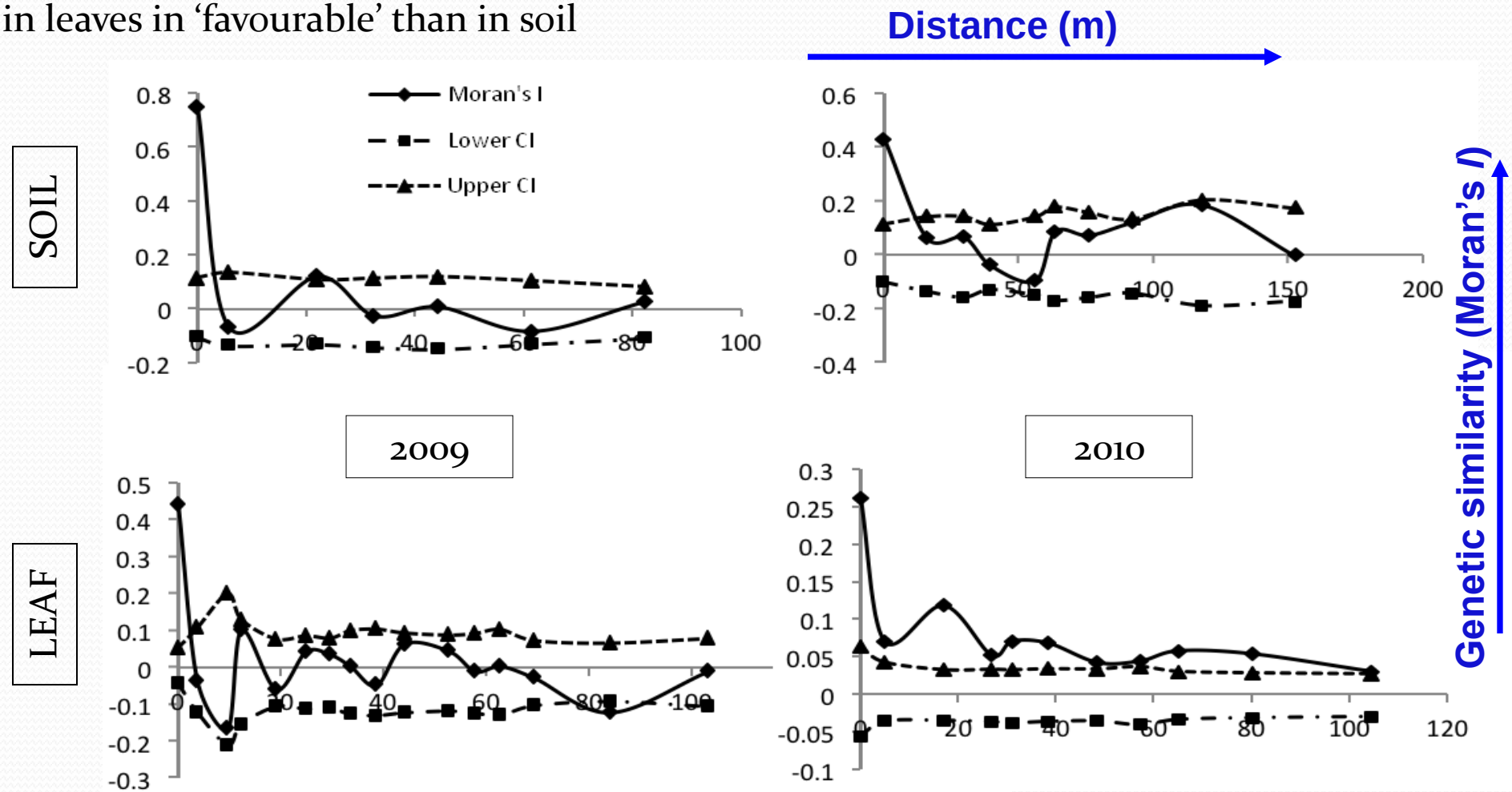


Spatial autocorrelation

Genetic similarity vs distance between individuals and the 6 plots – indication of dispersal

Soil 2009 and 2010 both relatively short distances

Leaf 2009 similar to soil, but in 2010 i.e. WET year changes a lot – indicates wider dispersal in leaves in 'favourable' than in soil



Conclusions

- Found a lot of genotypes in a small area
- Climate conditions affect diversity
  - WET → favourable for *P. ramorum* → overall diversity decreases in leaves
  - A few genotypes dominate
- Sympatric soil and leaves populations are genetically distinct
  - Some individuals better suited to survival in each substrate
- Inoculum travels further in wet conditions in leaf, but soil is relatively stable

Thank you!

- Garbelotto Lab
- SFPUC, Ellen Natesan
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- All my (un)willing field volunteers!