

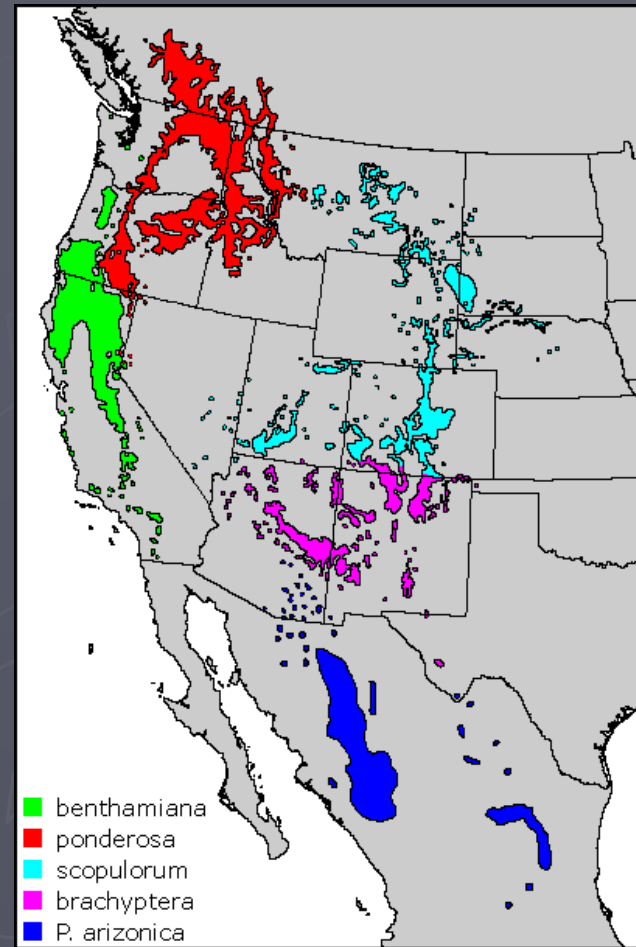
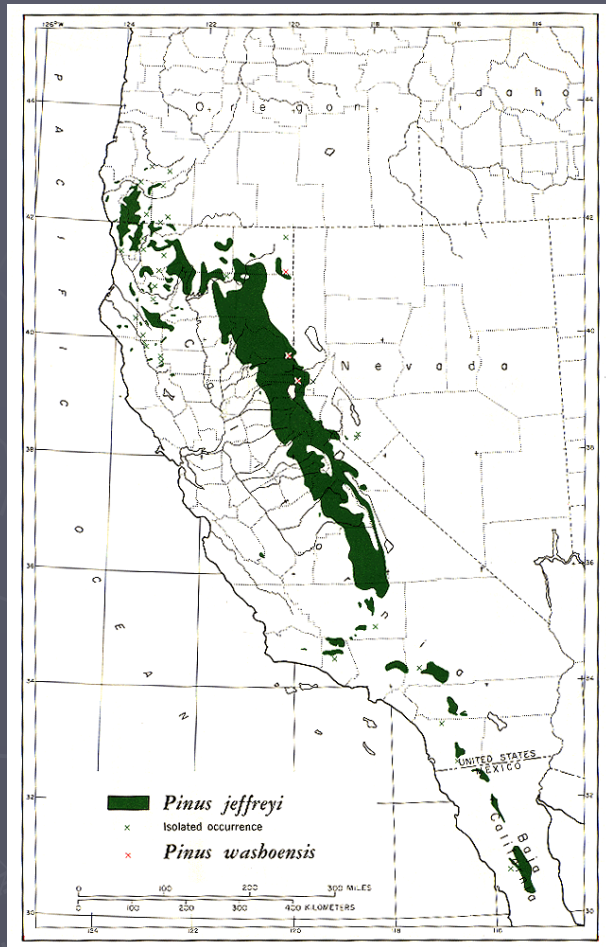
**Nesting microsatellite genotype
data within low-copy nuclear gene
trees to interpret the complex
evolutionary relationships of *Pinus
washoensis* (*Ponderosae*; *Pinaceae*)**

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Richard Cronn (USDA Forest Service PNW)**

Jeffrey

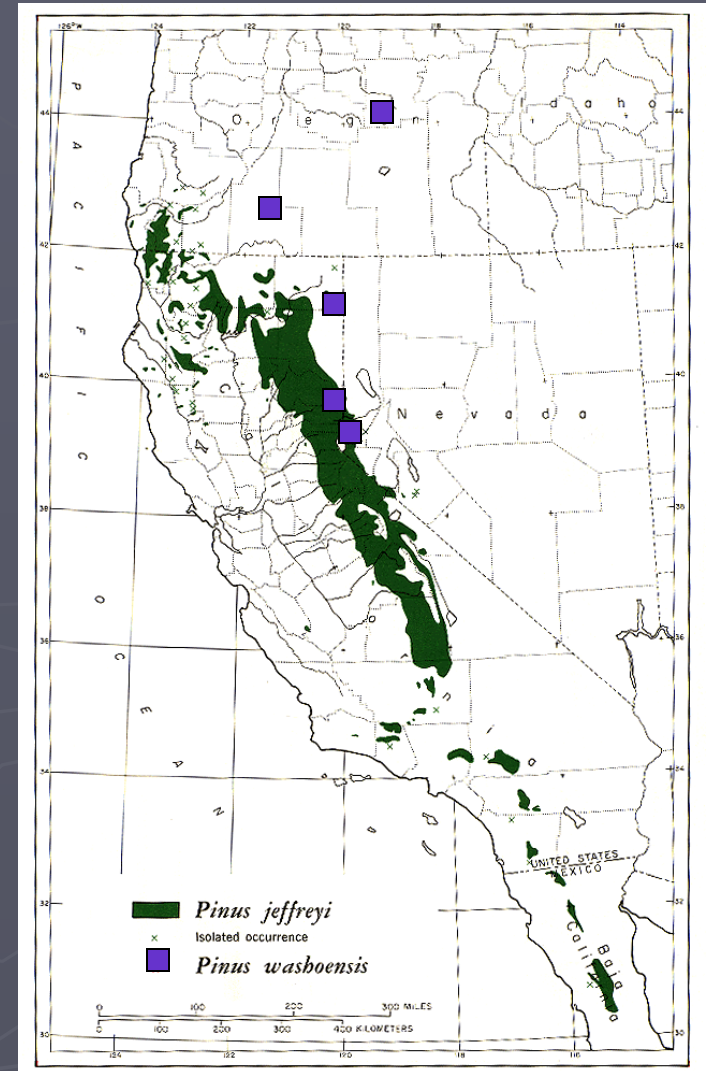
&

ponderosa



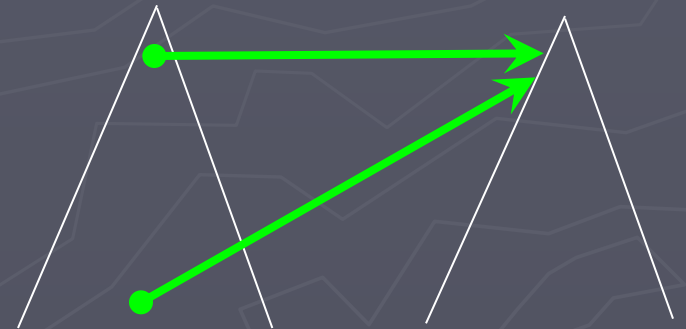
What is Washoe pine?

- Only known from three locations
- Higher elevation than Jeffrey
- Looks different than nearby Pacific race ponderosas
- Suggested that outlying populations of North Plateau race



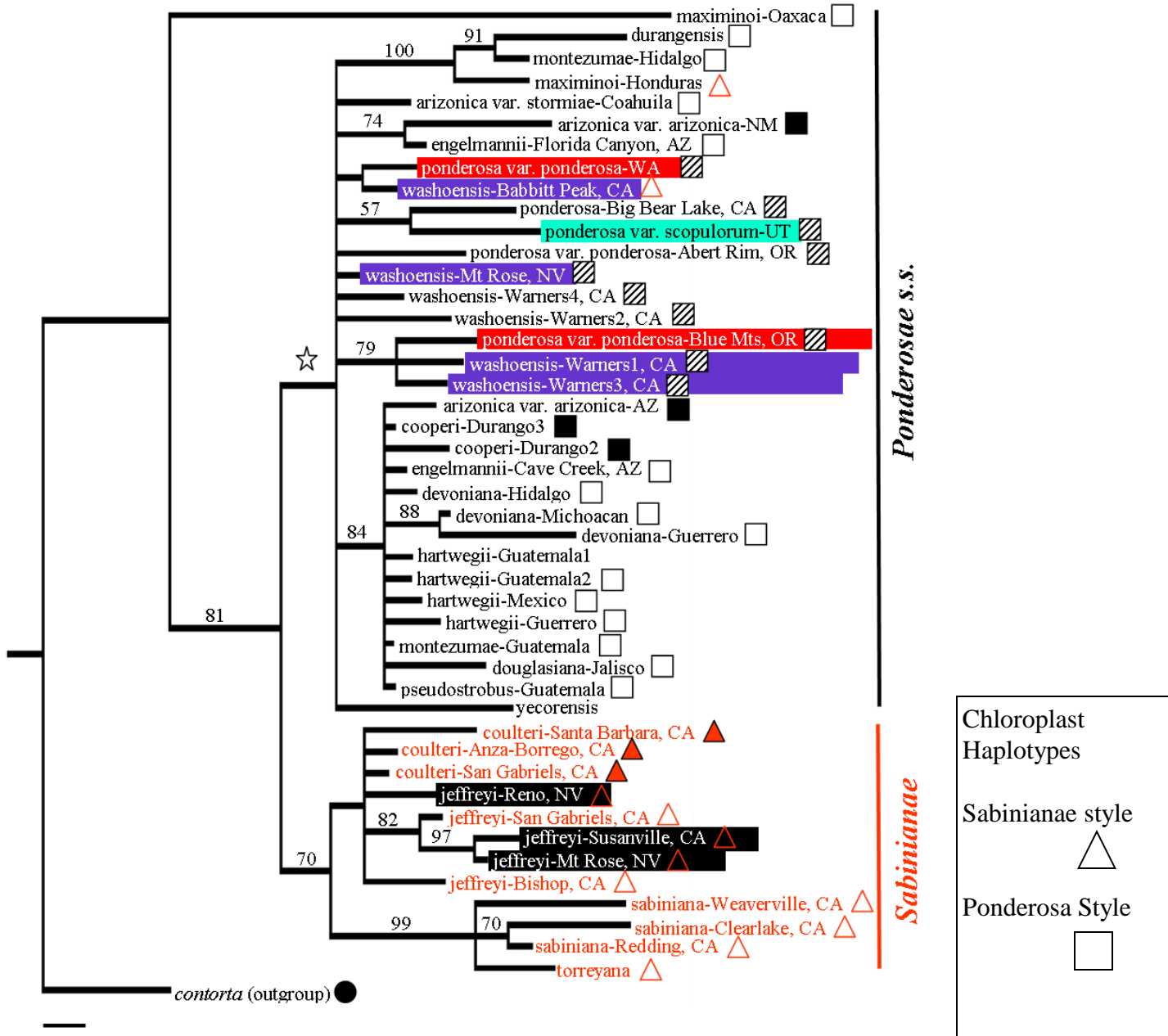
Washoe pines

- ▶ High elevation Washoe resemble other high elevation ponderosa and grow better
- ▶ Low ponderosa resemble high Washoe pops



Low-copy
nuclear gene
tree

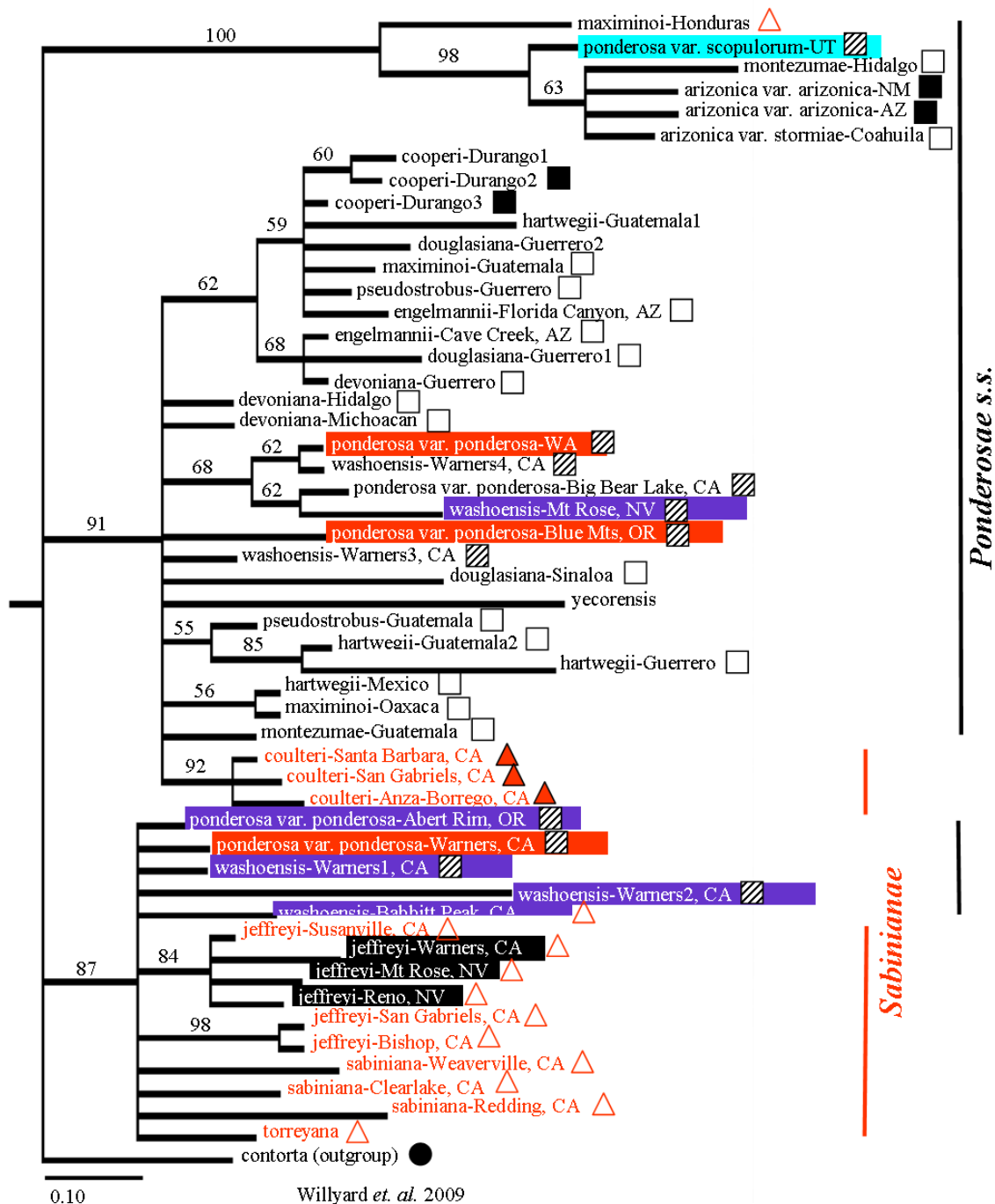
Lea-like



0.10

Low-copy
nuclear gene
tree

WD40



Chloroplast Haplotypes

Sabinianae style \triangle

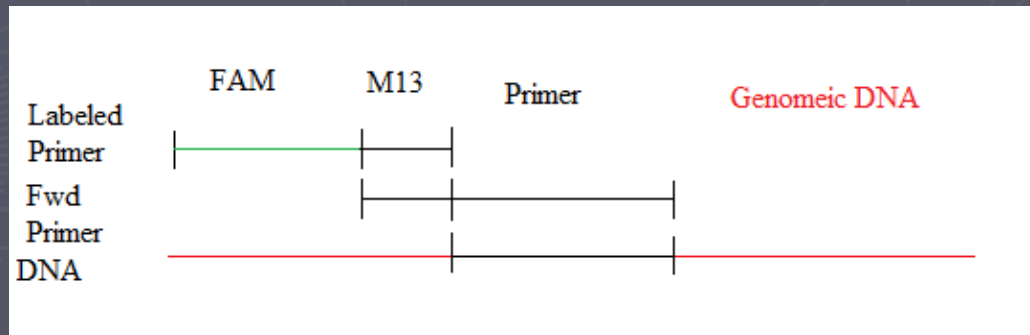
Ponderosa Style \square

Microsats/SSRs/VNTRs

- ▶ Repetitive sequences
- ▶ Non-coding
- ▶ High mutation rate
 - Usefull in phylogenetics and systematics

Amplification & Genotyping

- ▶ PCR
 - Amplify specific region
- ▶ 3 primer system



Hypothesis

Is *P. washoensis* unique or part of the wide-ranging *P. ponderosa*?



Three Alternatives:

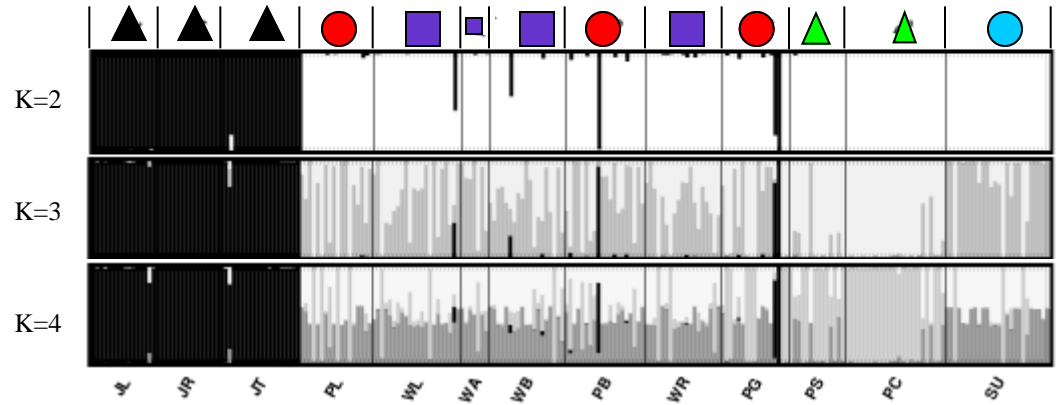
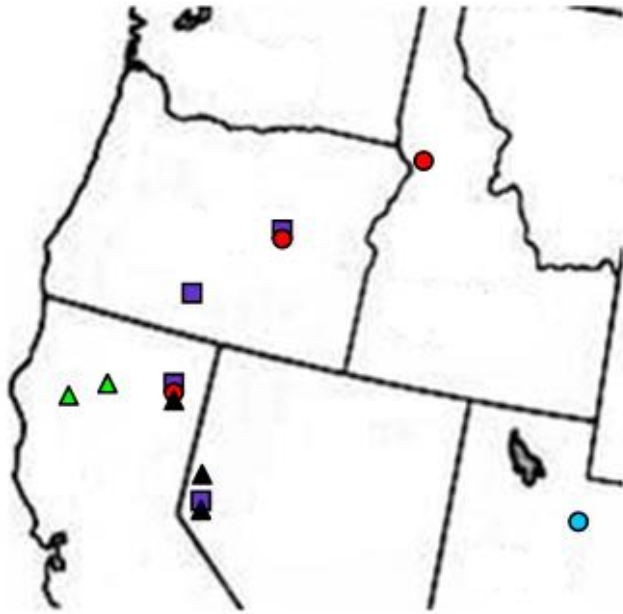
- A. *P. washoensis* distinct: high-elevation Oregon population + traditional locations
- B. *P. washoensis* distinct but confined to traditional locations
- C. *P. washoensis* encompassed within the diversity of ponderosa pine

Lab Methods

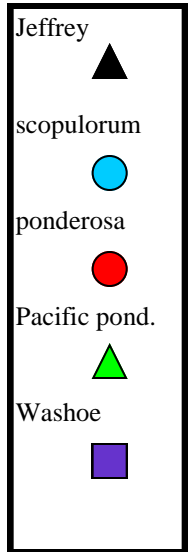
- ▶ Using 17 nSSRs from *P. taeda* (subg. *Pinus*)
- ▶ Selected 9 single-locus and useable amplifications
- ▶ From 13 populations and 249 individuals (mean=19.2 per population)
- ▶ Amplified 6 polymorphic loci
- ▶ Genotyped on ABI 3130xe
- ▶ 239 individuals with no missing data
- ▶ 70 alleles from 6 loci
- ▶ STRUCTURE (Pritchard)
- ▶ Principal Component Analysis

Locus	No. of Alleles
LOP5	23
PtTX2123	7
PtTX2128	8
PtTX3025	7
PtTX3030	19
PtTX3098	6
Total	70

13 Populations: Jeffrey, ponderosa, Washoe and scopulorum

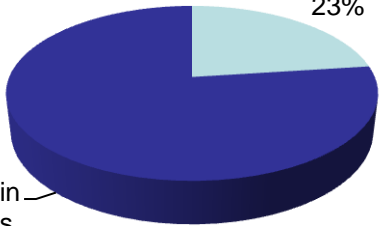


Structure

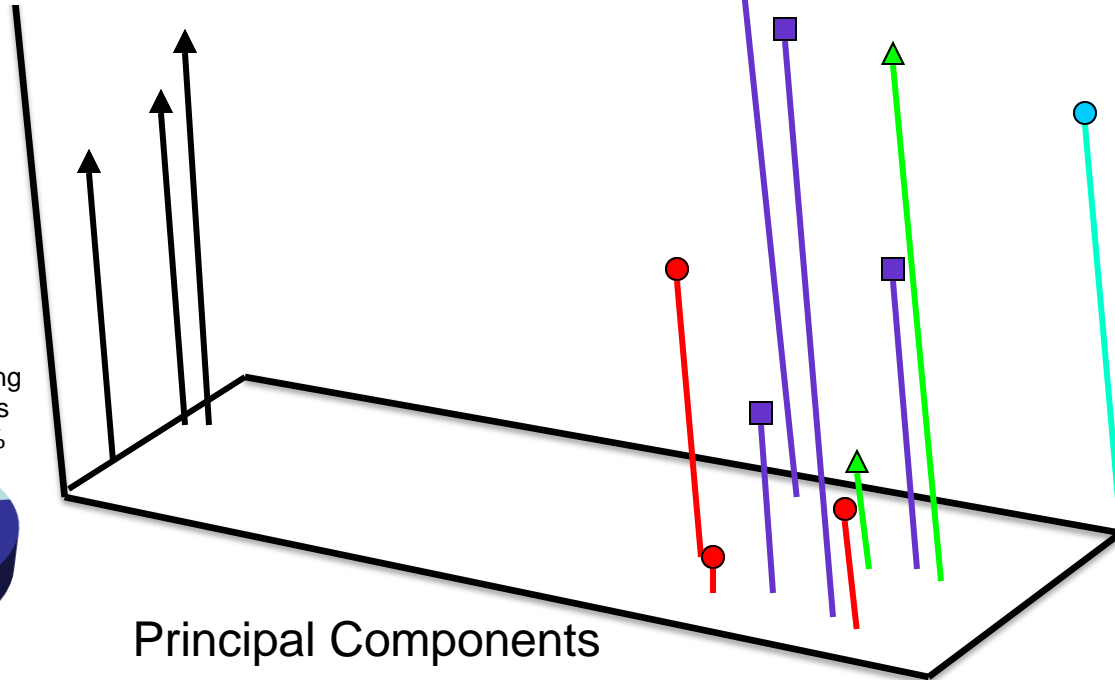


AMOVA

Among Pops 23%

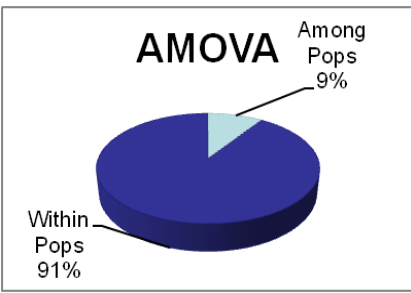
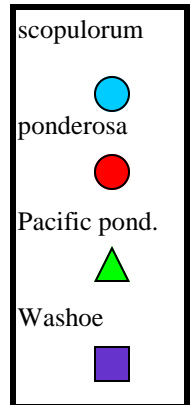
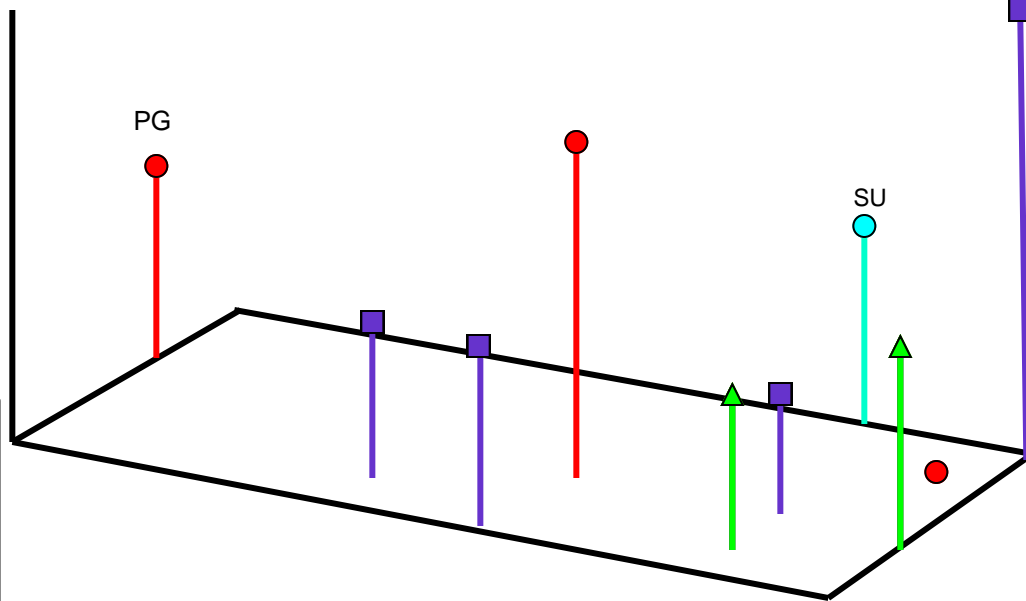
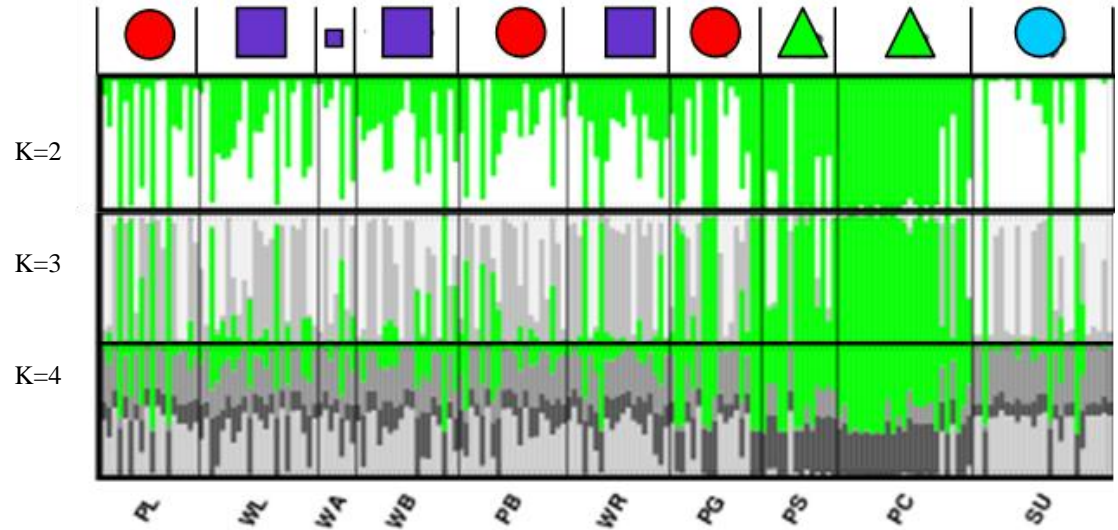
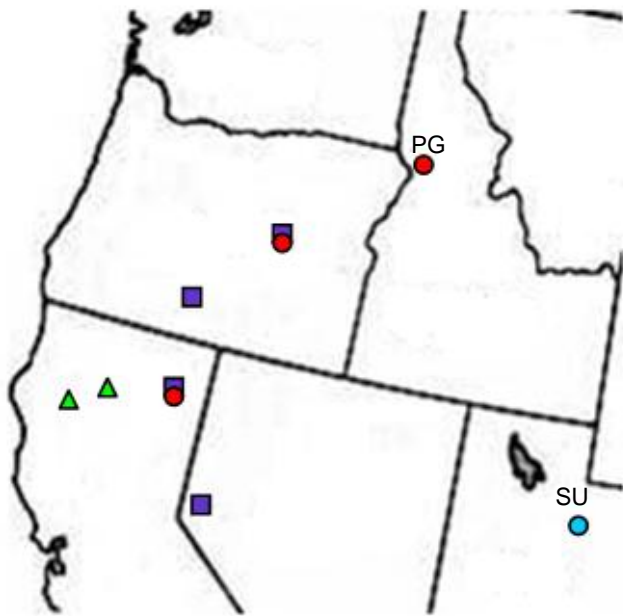


Within Pops 77%

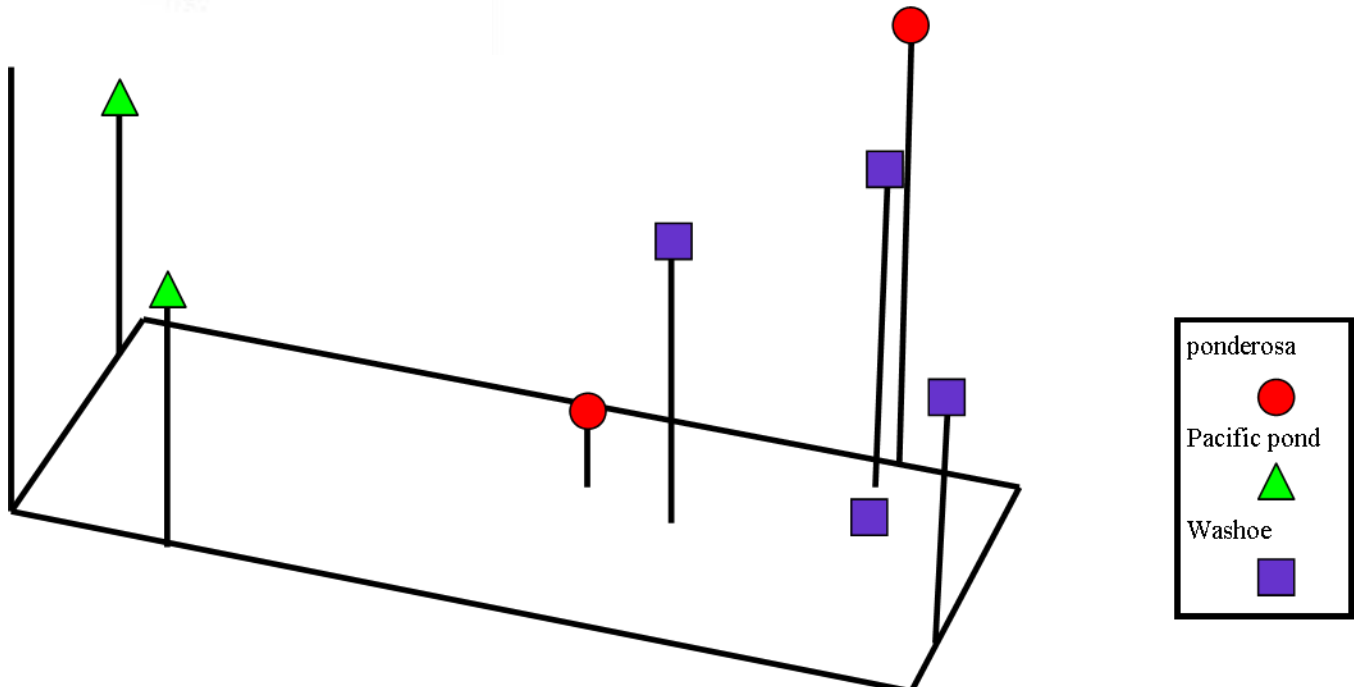
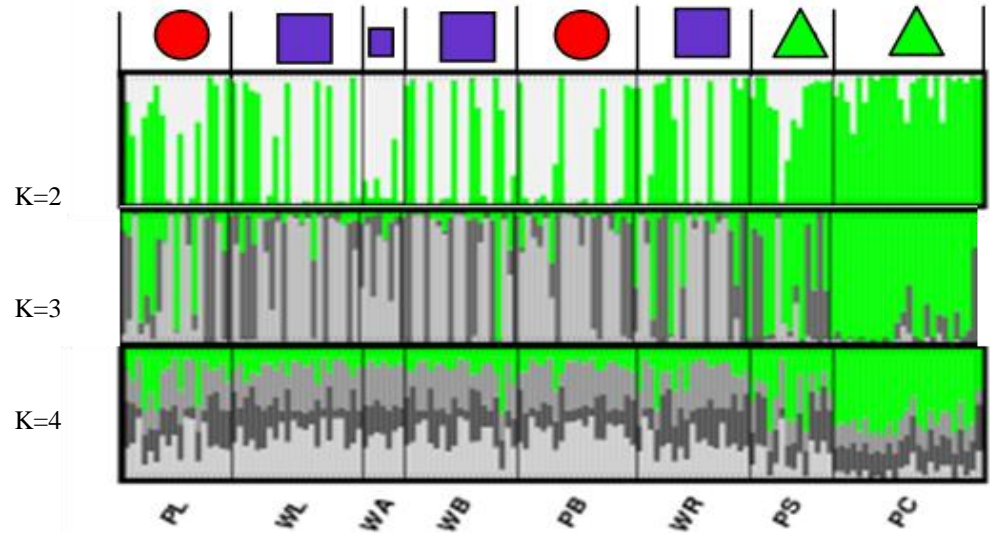
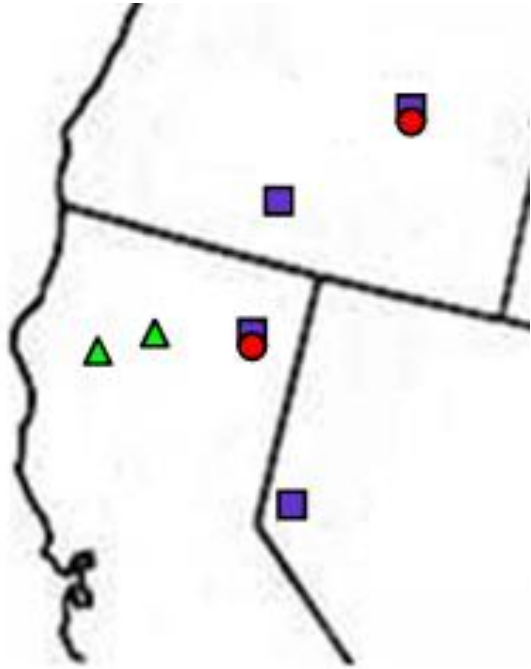


Principal Components

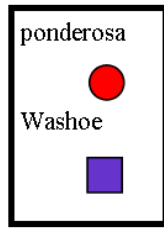
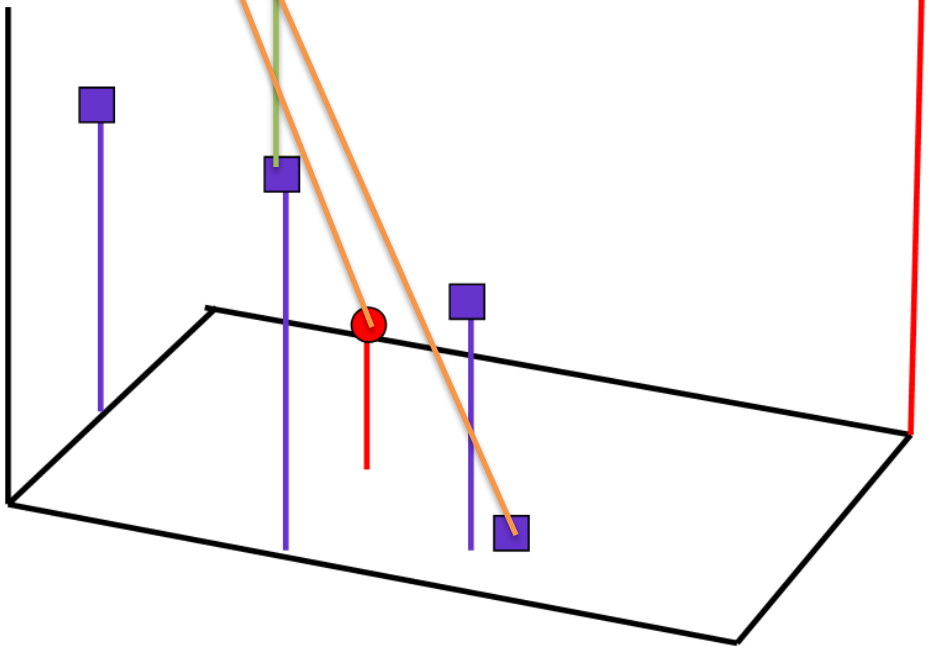
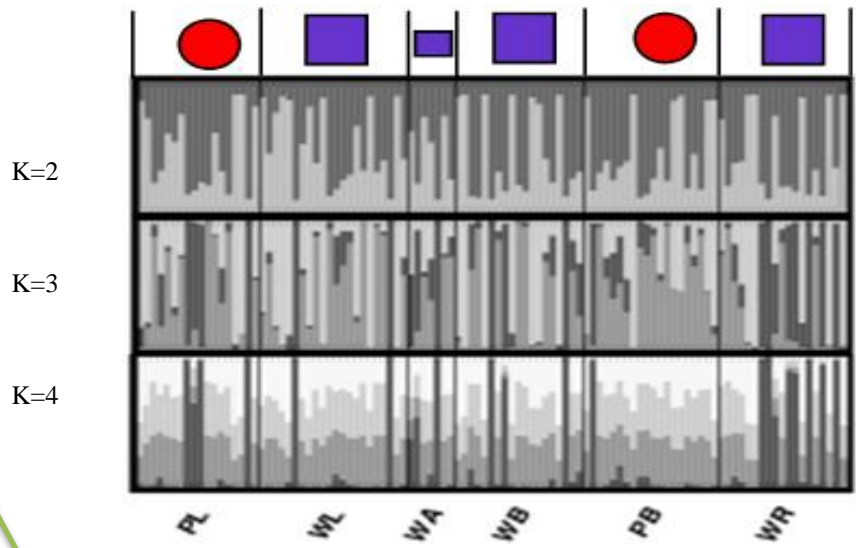
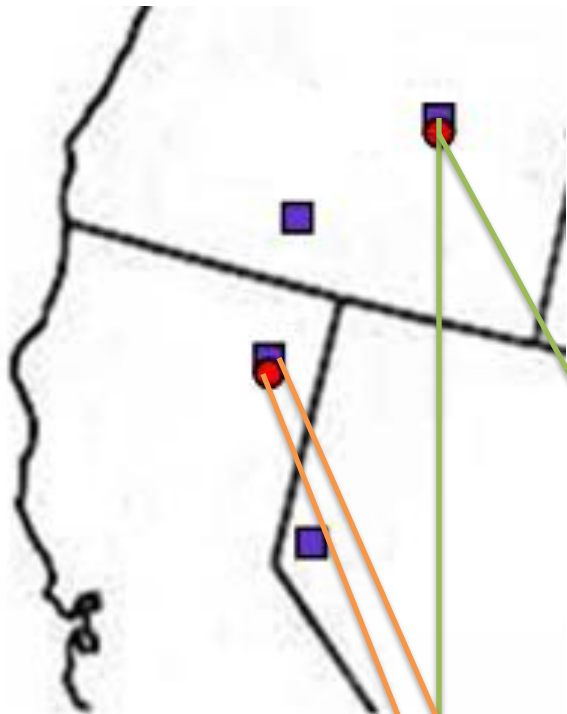
10 Populations: ponderosa, Washoe and scopulorum



8 Populations: ponderosa and Washoe



6 Populations: ponderosa and Washoe



Conclusions

- ▶ Reject hypothesis that *P. washoensis* is distinct from *P. ponderosa* var. *ponderosa*
- ▶ Support that Washoe fits within North Plateau race





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