Northern Spotted Owl listing Genetic Considerations: Systematics and Subspecies Validity

Subcommittee:

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Jack Dumbacher
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Taxonomy and Systematics of NSO

- Evaluate the subspecies designation (as well as "phylogenetic species" or DPS or ESU or MU).
- Evaluate and interpret the degree of hybridization between subspecies; extent of genetic introgression.

Subspecies definition:

- Subspecies are considered to be interbreeding populations of the same species in different areas that are recognizably different.
- "a collection of populations occupying a distinct breeding range and diagnosably distinct from other such populations" (Mayr and Ashlock 1991).
  - Implies that there may be some interbreeding or hybridization among subspecies
- The 75% rule: 75% of a population must be distinct or diagnosably different from 75% of the individuals of the other population (Amadon 1949)

Subspecies descriptions and morphological studies:

- *Strix occidentalis* [*occidentalis*] of California described by Xantus in 1858. California Spotted Owl (CSO).

- *S. o. caurina* described by Merriam in 1898 (notes: "Comparison of the northwestern Spotted Owl with the type specimen of *S. occidentale* shows it to be a well-marked subspecies, differing, like so many birds of the same region, in darker and richer coloration" also that spots are smaller and fewer, and that the dark areas are larger and darker). Northern Spotted Owl (NSO).

- *S. o. lucida* described by Nelson in 1903 (also by Swarth in 1910 as *S. o. huachucae*, but these now considered synonymous). Mexican Spotted Owl (MSO). Much paler in color and larger white spots than CSO or NSO.

- Oberholser (1915) tried to lump CSO and NSO into single subspecies, but AOU (checklists from 1910-1957) did not agree.

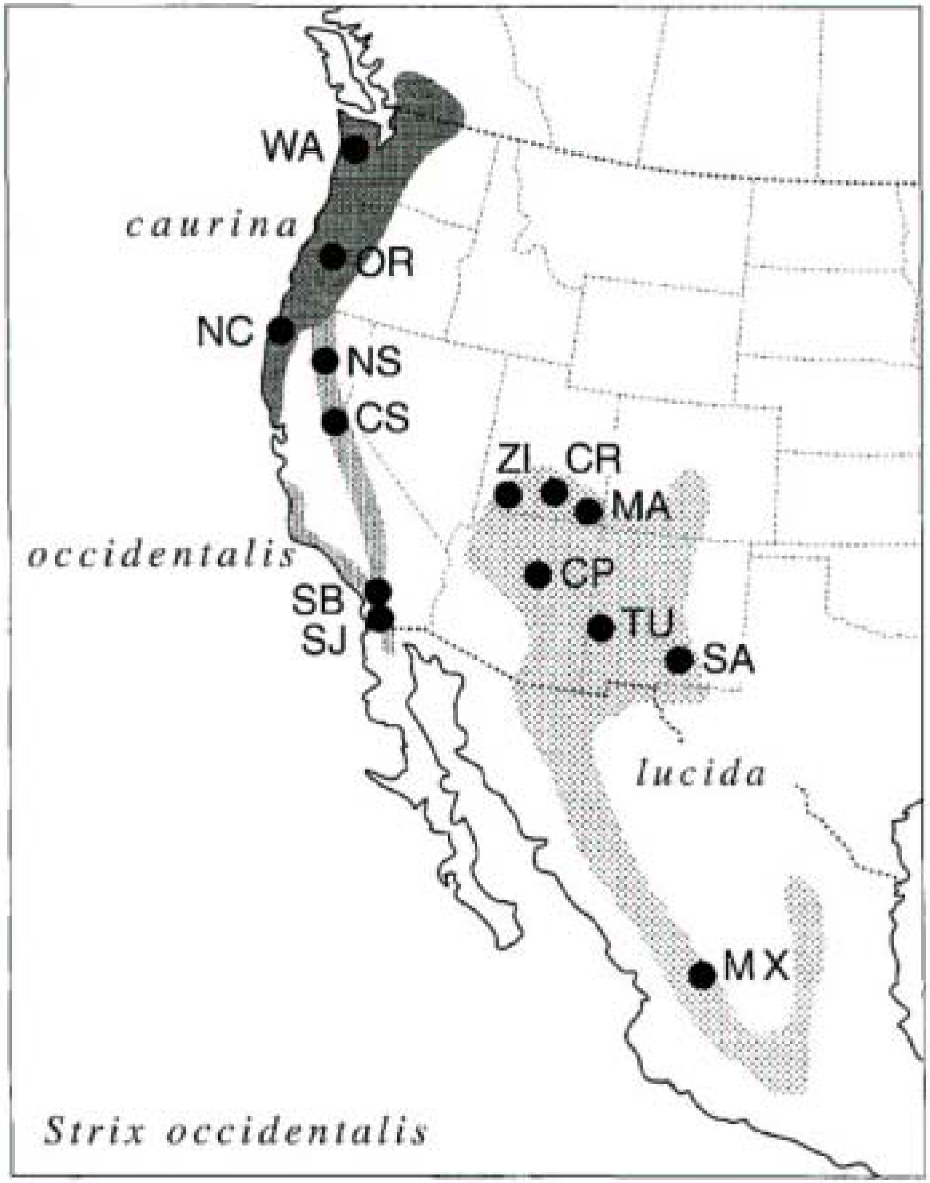


FIG. 1. Geographic ranges of the three recognized subspecies of spotted owls and the locations and acronyms of population samples analyzed in this study. *Strix occidentalis caurina, S. o. occidentalis*, and *S. o. lucida* are the northern, California, and Mexican spotted owls, respectively.

Subspecies descriptions and morphological studies (continued):

Various authors - (AOU, Ridgeway, Coues, Bent, Pyle) support 3 subspecies based on morphology. Very detailed descriptions:

- Spots on breast small in NSO, medium in CSO, large in MSO.
- Plumage medium-dark brown in NSO, medium brown in CSO, pale brown in MSO.
- Wing/tail bars medium-pale brown in NSO, pale brown in CSO, whitish in MSO.

Barrowclough - summaries in 1996 **(*Wild Mammals in Captivity* & Int. Orn. Congress XX)**: Dataset details (sample sizes, provenance, specimens) not provided. Three types of morphological data: size characters, plumage characters, and plumage color ranking.

- PC1 (size): difference between MSO and other SO's (MSO smaller). No difference between NSO and CSO in body size.
- Plumage pattern: differences between MSO and other SOs. Clinal variation within CSO and NSO.
- Plumage color shows clinal variation from Pacific NW (darker) to SW deserts (lighter) [Gloger's Rule].

Behavioral studies: Song

Van Gelder, J.J. 2004. Masters Thesis, Humboldt State University.

- Examined Spotted Owl four-note location call from all three subspecies, and multiple localities
- found that the model that best explained variation in Spotted Owl calls was a model inferring that all three subspecies are distinct
- This model had the lowest Akaike value and accounted for 83.6% of the Akaike weight, which indicates strong fit of these data to the model
- Song is often adaptive, and important in mate selection

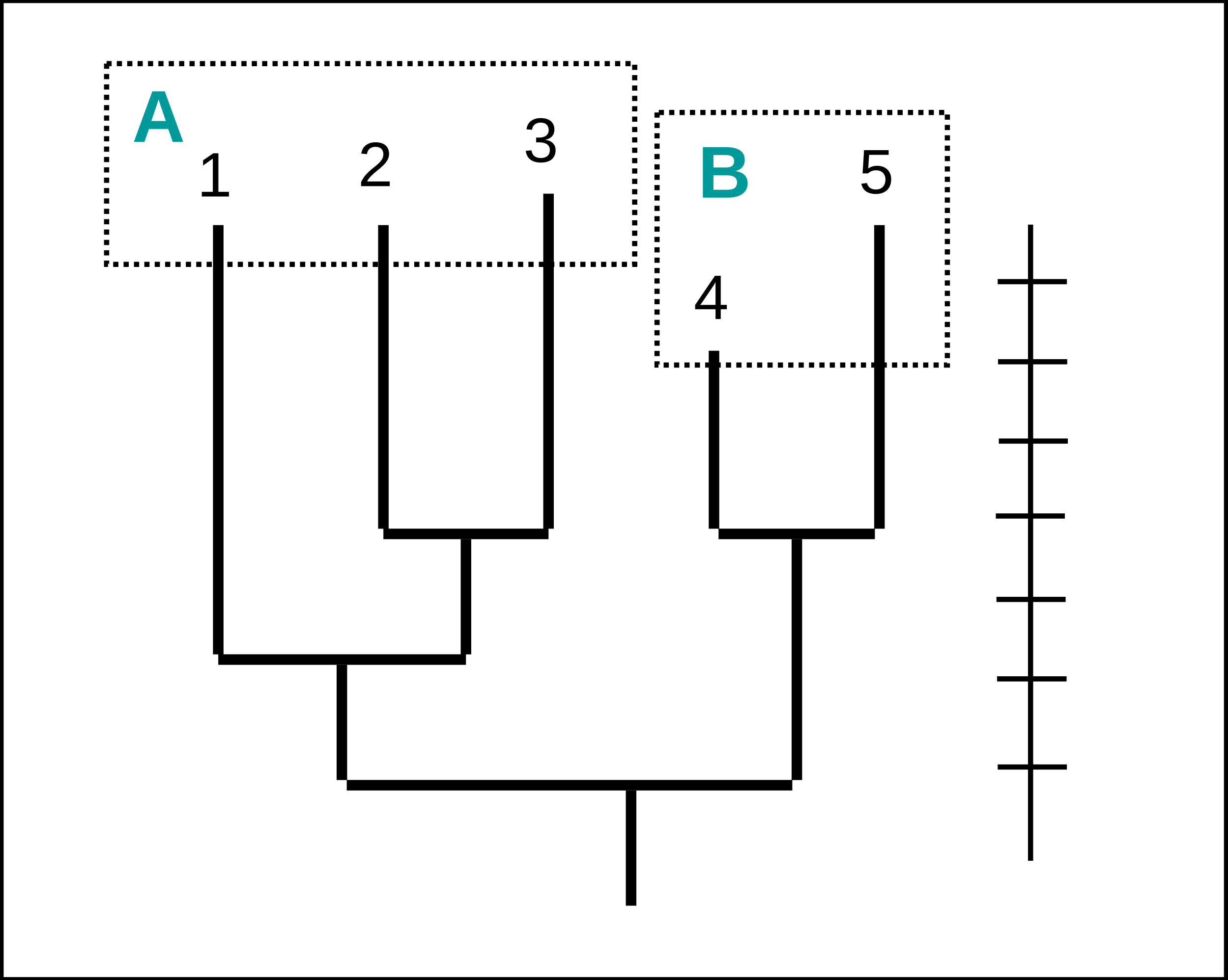
Genetic studies: Patterns supporting subspecies

Haplotypes (or alleles) found to occur in only one subspecies range (unique haplotypes, “diagnosability”)

Haplotypes of putative subspecies are reciprocally monophyletic (members of each subspecies are more closely related to each other than to other subspecies).

Haplotype of one group monophyletic, clustered within another group (young subspecies, or subspecies that derived from another)

Reciprocal monophyly between populations A and B:



Genetic studies: Allozymes

Barrowclough and Gutiérrez 1990. Auk 107:737-744.

- All 3 subspecies assessed, 8 populations, 107 individuals.
- Examined 23 allozyme loci from whole blood.
- 22 were monomorphic, one (fluorescent esterase-D) had two alleles, fixed in NSO and CSO; at 39% and 61% in MSO.

Genetic studies: Mitochondrial DNA

Barrowclough et al. 1999. Evolution 53:919-931.

- All 3 subspecies assessed, 13 populations, 73 individuals.
- Sequenced 1105 bp of Control Region of mitochondrial DNA.
- Found 37 haplotypes, phylogenetic tree showed three clear clades -
  - corresponding almost completely to each described subspecies and geographical region (one case of a mismatched genotype = disperser? Hybrid?).
  - Lowest variation (nucleotide diversity) in CSO. CSO+MSO clade is sister to NSO.
  - Only 1 individual evidence of hybrid or dispersal (1/20 or 5% of NSO)

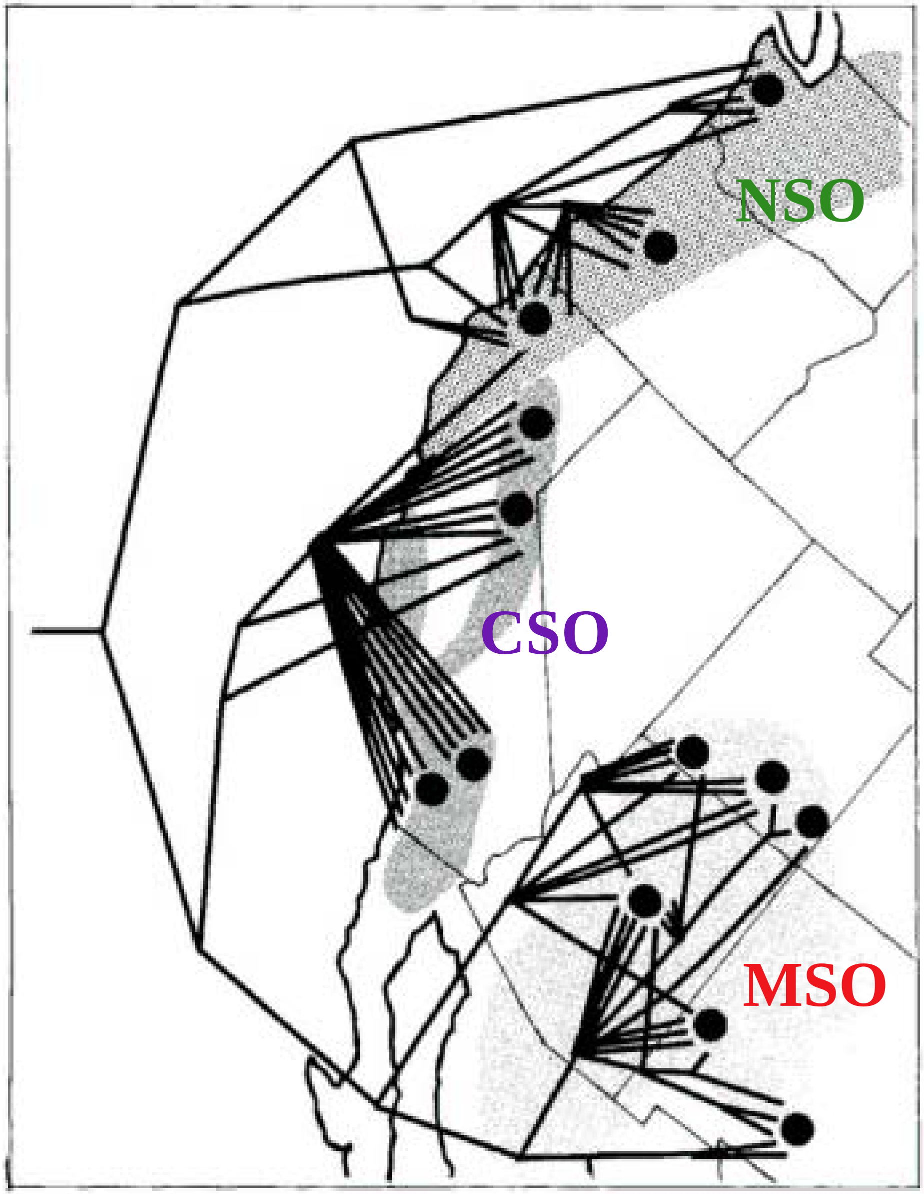


FIG. 5. A maximum-parsimony tree for 73 spotted owls based on mtDNA control region sequences. One terminal branch is directed to the geographic source of each individual owl; some of these branches have a length of zero. The tree was rooted with two barred owls.

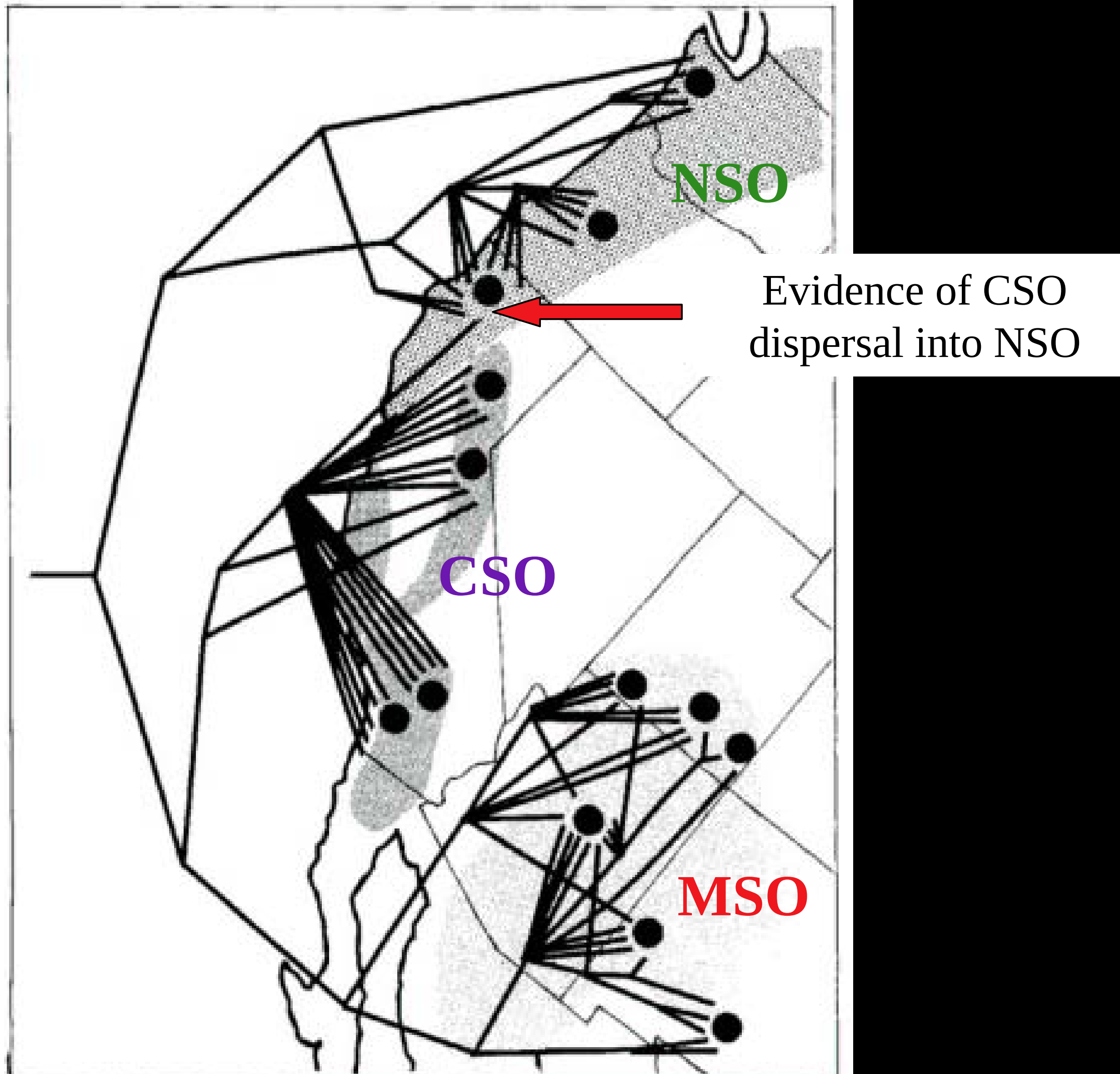
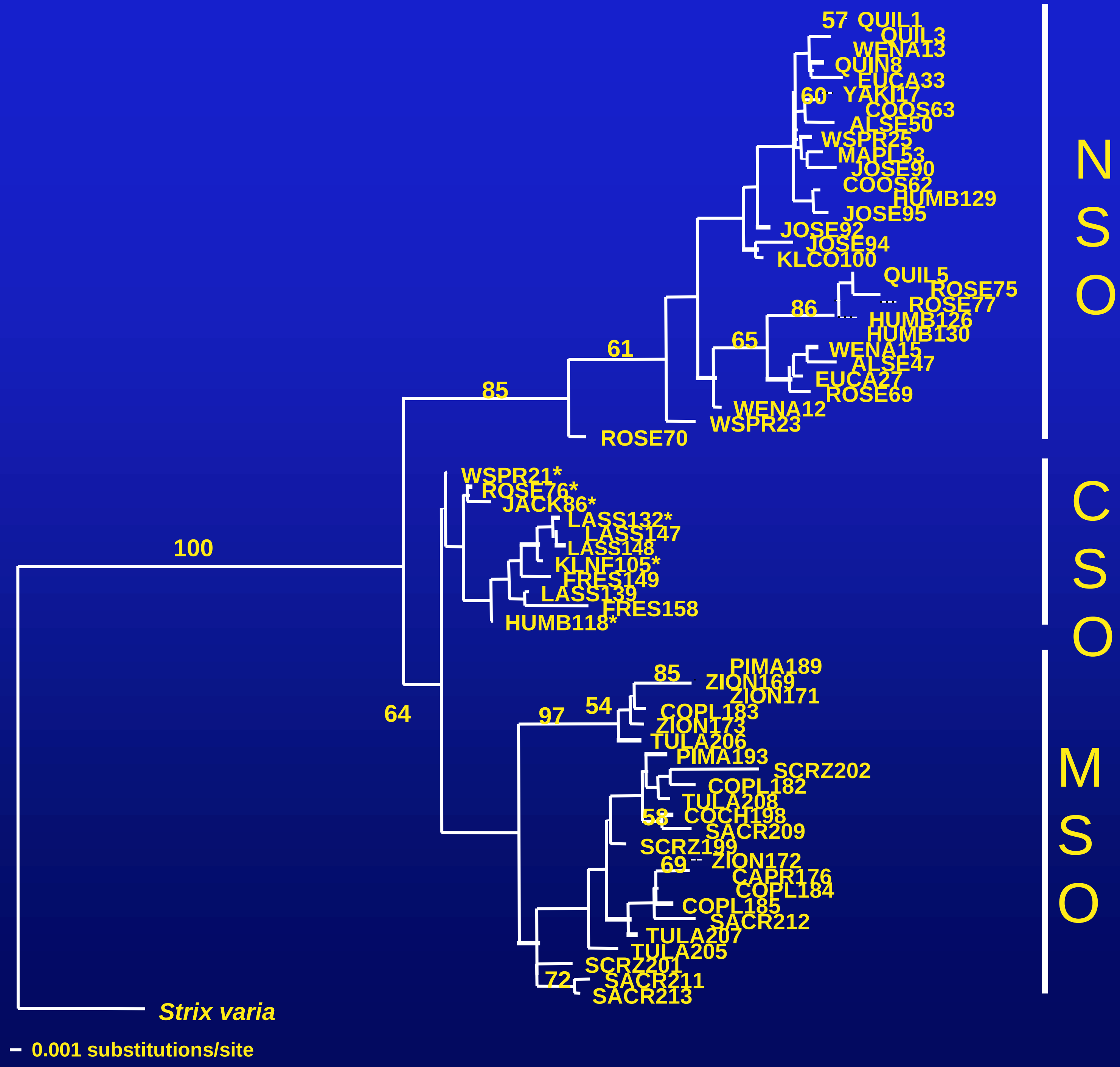


FIG. 5. A maximum-parsimony tree for 73 spotted owls based on mtDNA control region sequences. One terminal branch is directed to the geographic source of each individual owl; some of these branches have a length of zero. The tree was rooted with two barred owls.

Genetic studies: Mitochondrial DNA

Haig et al., 2004. Conservation Genetics, In press.

- sequenced 522 base pairs of control region from 213 individuals from 30 local breeding areas, representing all three Spotted Owl subspecies
- found 63 unique haplotypes; 34 of these were recovered only from NSO populations, 5 were recovered only from CSO populations, 23 were recovered only from MSO populations, and only one haplotype was found in both NSO and CSO populations.
- Based upon unique haplotypes, mixing in 2 of 168 NSO+CSO or 1.2% (1.5% of NSO)


57 QUIL1
QUIL3
WENA13
QUIN8
EUCA33
60 YAKI17
COOS63
ALSE50
WSPR25
MAPL53
JOSE90
COOS62
HUMB129
JOSE95
JOSE92
JOSE94
KLCO100
QUIL5
ROSE75
86 ROSE77
HUMB126
HUMB130
65 WENA15
ALSE47
EUCA27
ROSE69
61
WENA12
WSPR23
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ROSE70
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WSPR21*
ROSE76*
JACK86*
LASS132*
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LASS148
KLNF105*
FRES149
LASS139
FRES158
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PIMA193
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SCRZ199
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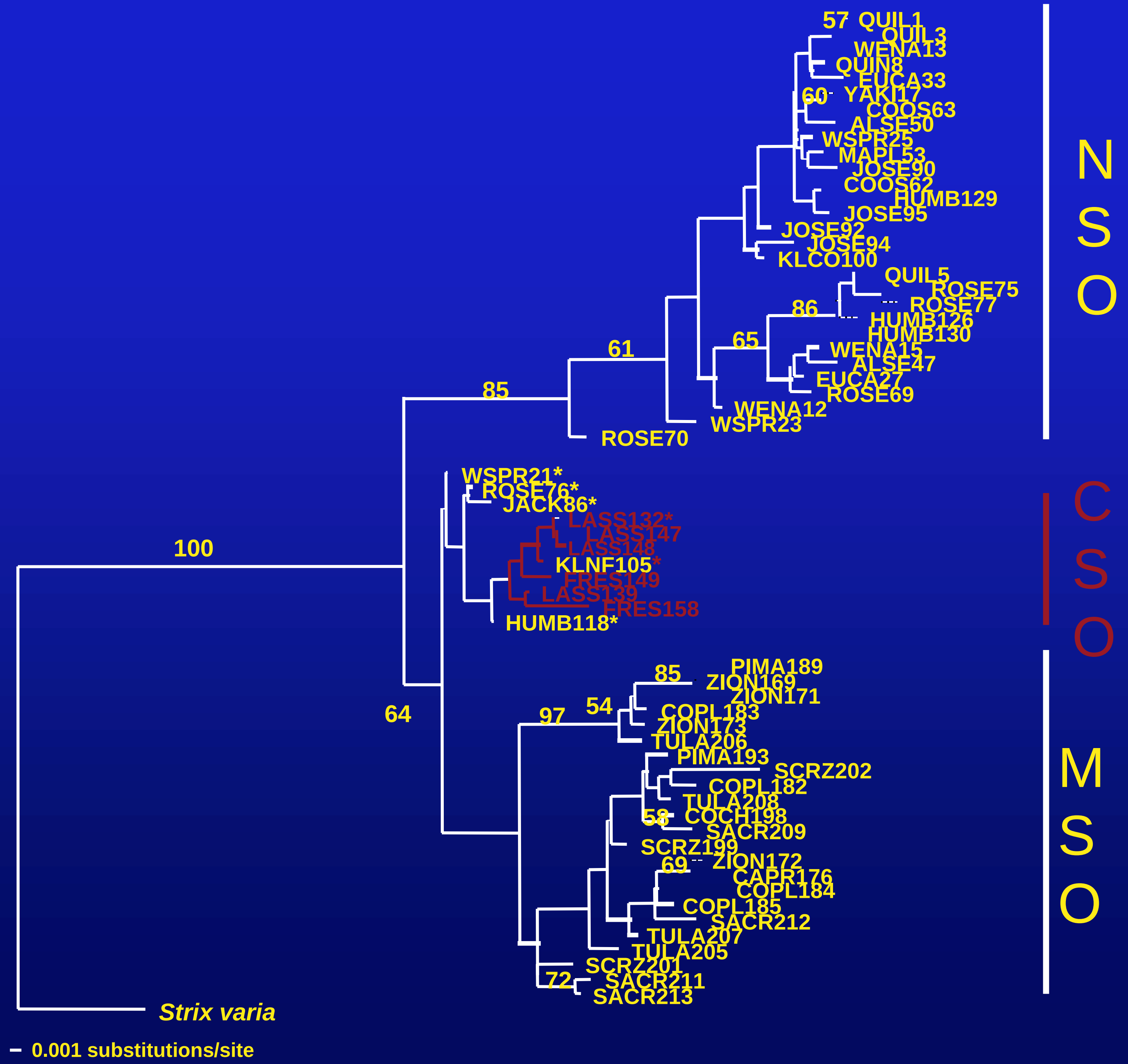
0.001 substitutions/site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ZION173
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PIMA193
SCRZ202
COPL182
TULA208
58
COCH198
SACR209
SCRZ199
69
ZION172
CAPR176
COPL184
COPL185
SACR212
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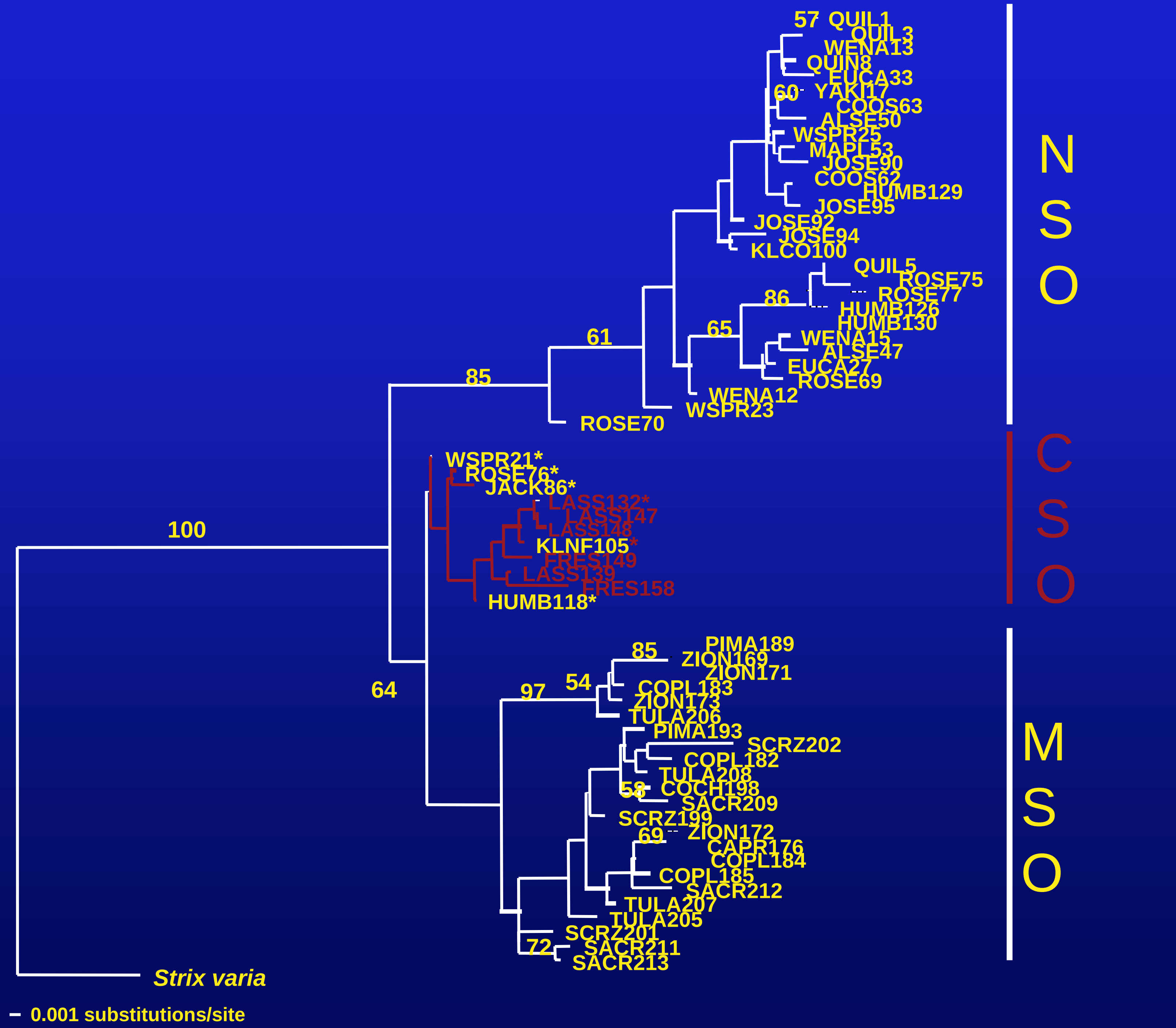
Strix varia
0.001 substitutions/site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LASS148
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ZION169
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ZION173
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PIMA193
SCRZ202
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COCH198
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SACR211
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Strix varia
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Genetic studies: Mitochondrial DNA

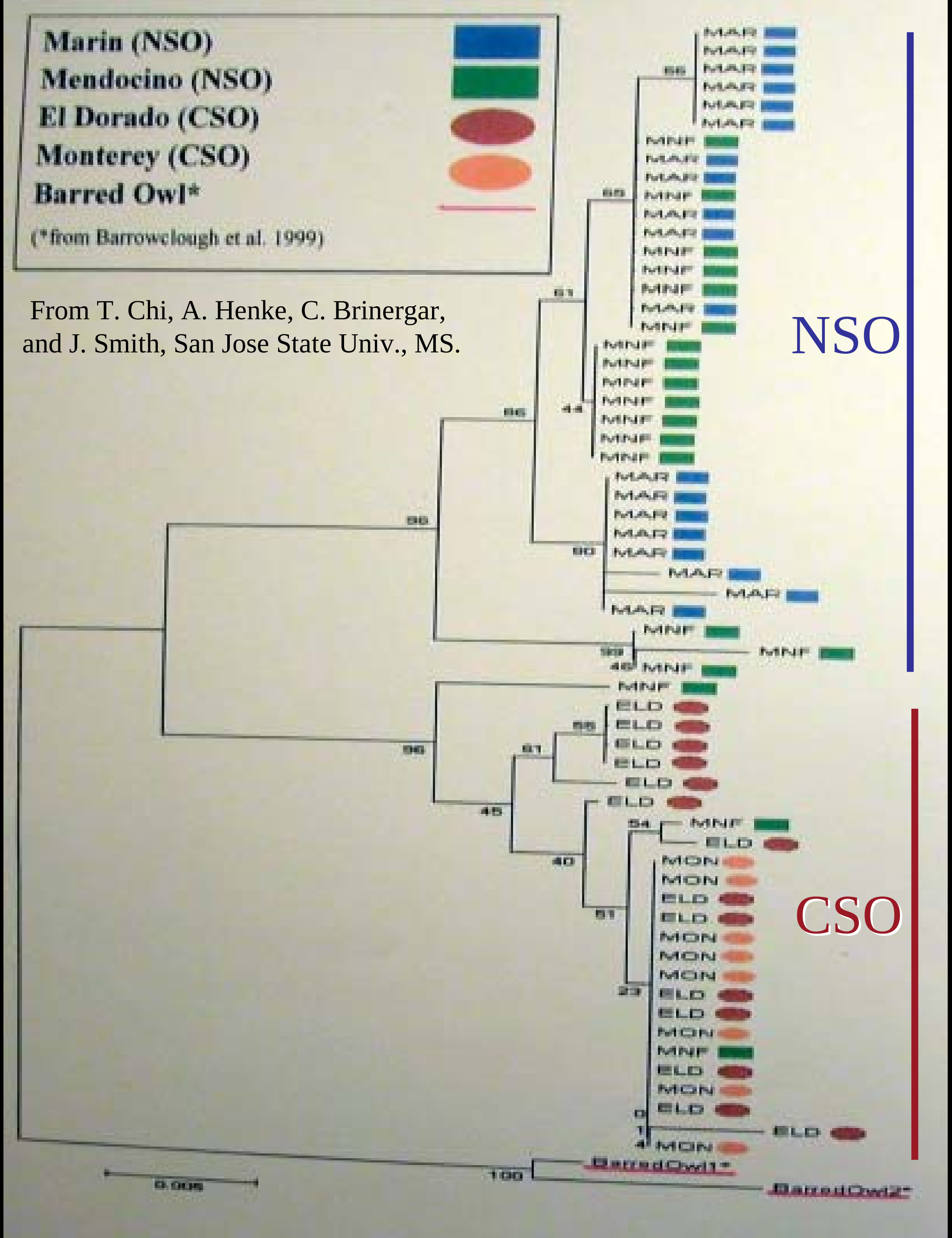
Haig et al., 2004. Conservation Genetics, In press.

- Based upon CSO and NSO clades (which are poorly resolved), 15 of 168 traditional NSO+CSO are or 8.9% (11.4% of NSO only)
- Even by these very inclusive criteria, the NSO is more than 75% "pure" NSO clade, and nearly 99% unique NSO haplotypes

Genetic studies: Mitochondrial DNA

Chi et al., MS (San Jose State University)

- Four populations examined; two in the traditional NSO range (Marin n = 19 individuals; Mendocino n = 19); and two in the CSO range (El Dorado n = 14; Monterey n = 8)
- 788 bp of Control Region sequenced and analysed
- The designation of separate subspecies based on the 75% rule is upheld, with only 1 of 38 birds collected in the NSO range having CSO haplotypes (2.6%) or 3 of 38 birds collected in NSO range with haplotypes belonging to the CSO clade (7.9%).
- An alternative way to look at this is that NSO populations are at least 92.1% pure.

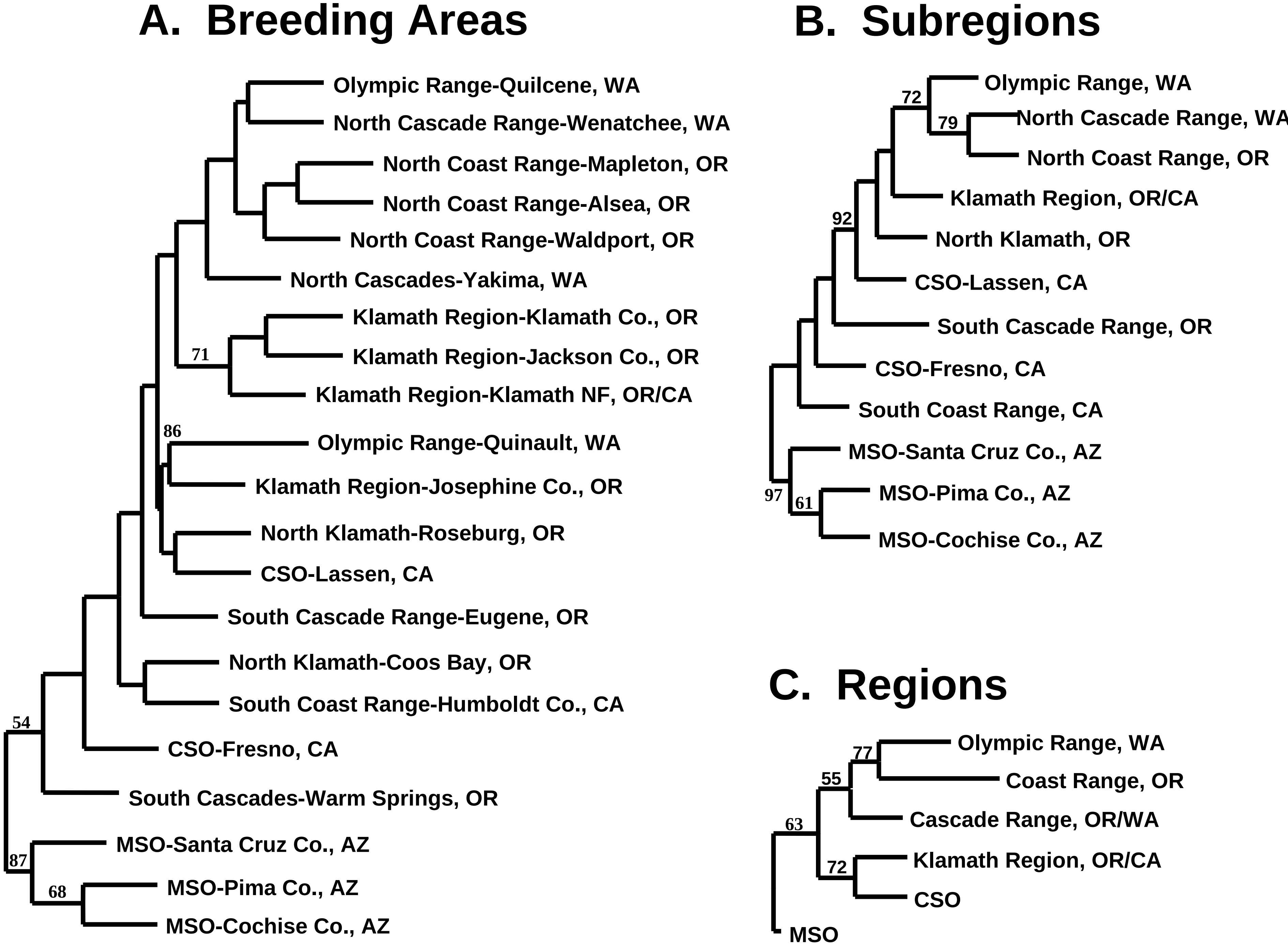


From T. Chi, A. Henke, C. Brinergar, and J. Smith, San Jose State Univ., MS.

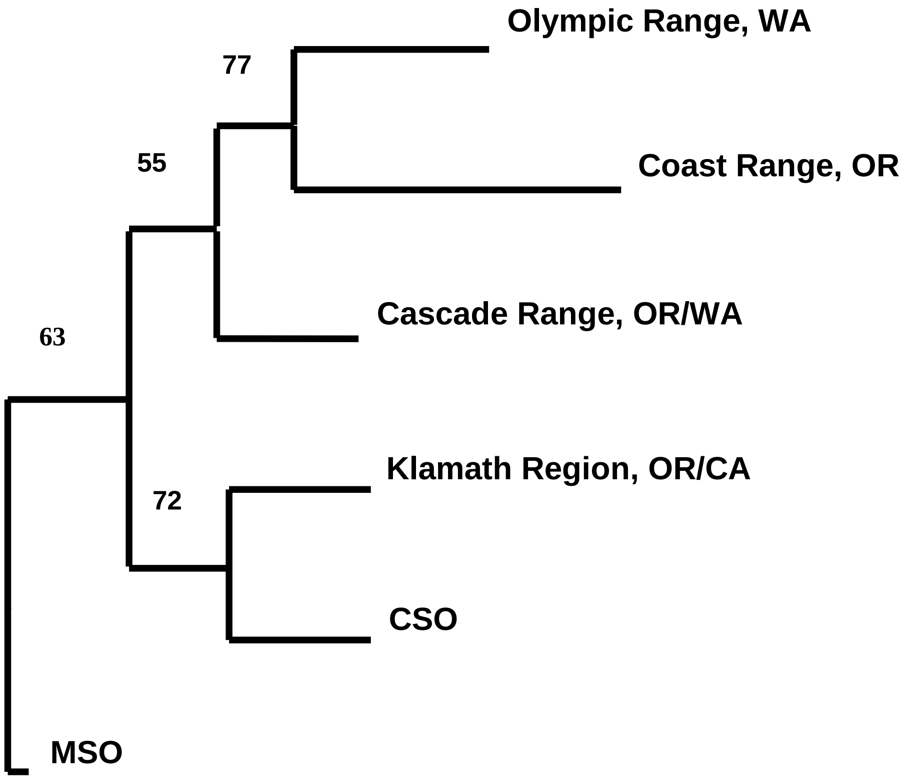
Genetic studies: Nuclear RAPD Markers

Haig et al. 2001. Conservation Genetics 2:25-40.

- All 3 subspecies assessed, 21 “populations”, 276 individuals.
- Used RAPDs - amplified with 400 primer sets.
- Found only 11 variable bands, extremely low variation, but significant structure. MSO differentiated (ESU), but no clear monophyly of CSO and NSO.

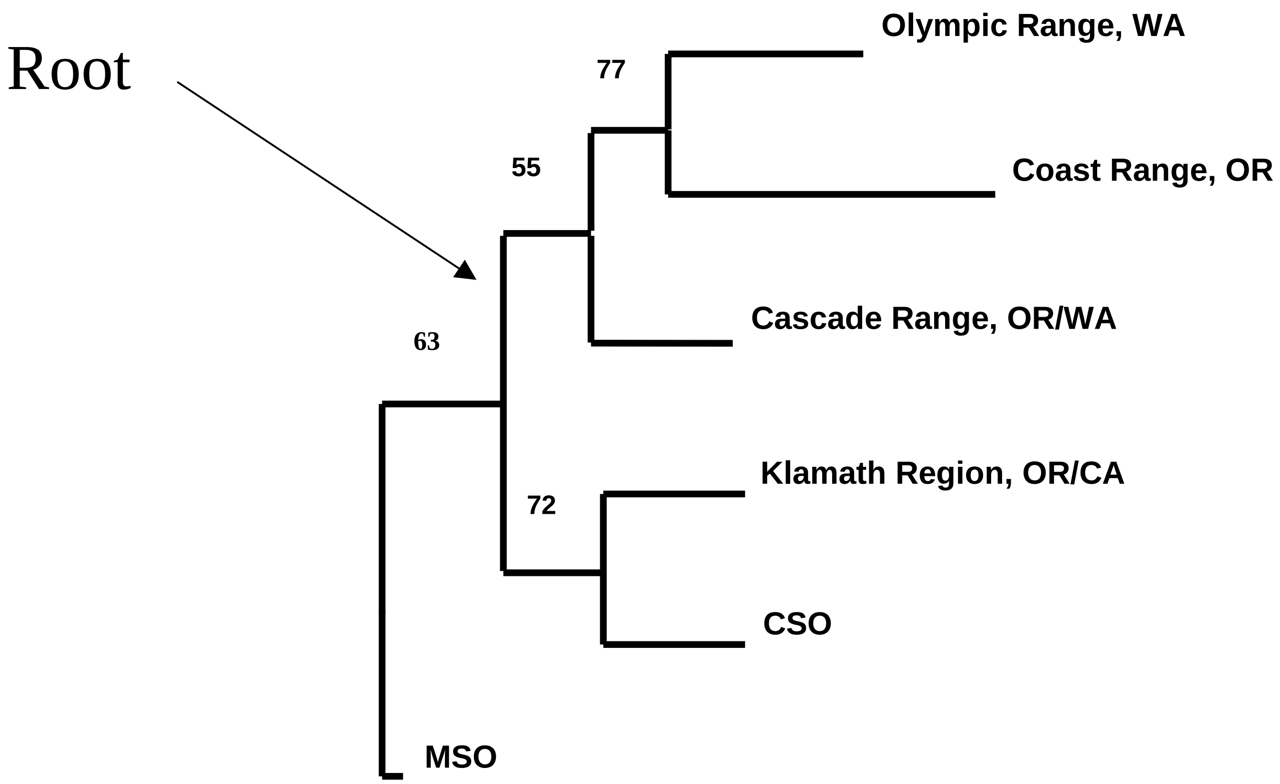

A. Breeding Areas
Olympic Range-Quilcene, WA
North Cascade Range-Wenatchee, WA
North Coast Range-Mapleton, OR
North Coast Range-Alsea, OR
North Coast Range-Waldport, OR
North Cascades-Yakima, WA
Klamath Region-Klamath Co., OR
71
Klamath Region-Jackson Co., OR
Klamath Region-Klamath NF, OR/CA
86
Olympic Range-Quinault, WA
Klamath Region-Josephine Co., OR
North Klamath-Roseburg, OR
CSO-Lassen, CA
South Cascade Range-Eugene, OR
North Klamath-Coos Bay, OR
South Coast Range-Humboldt Co., CA
54
CSO-Fresno, CA
South Cascades-Warm Springs, OR
MSO-Santa Cruz Co., AZ
87
68
MSO-Pima Co., AZ
MSO-Cochise Co., AZ
B. Subregions
Olympic Range, WA
72
79
North Cascade Range, WA
North Coast Range, OR
Klamath Region, OR/CA
92
North Klamath, OR
CSO-Lassen, CA
South Cascade Range, OR
CSO-Fresno, CA
South Coast Range, CA
MSO-Santa Cruz Co., AZ
97
61
MSO-Pima Co., AZ
MSO-Cochise Co., AZ
C. Regions
77
Olympic Range, WA
Coast Range, OR
55
63
Cascade Range, OR/WA
72
Klamath Region, OR/CA
CSO
MSO

C. Regions

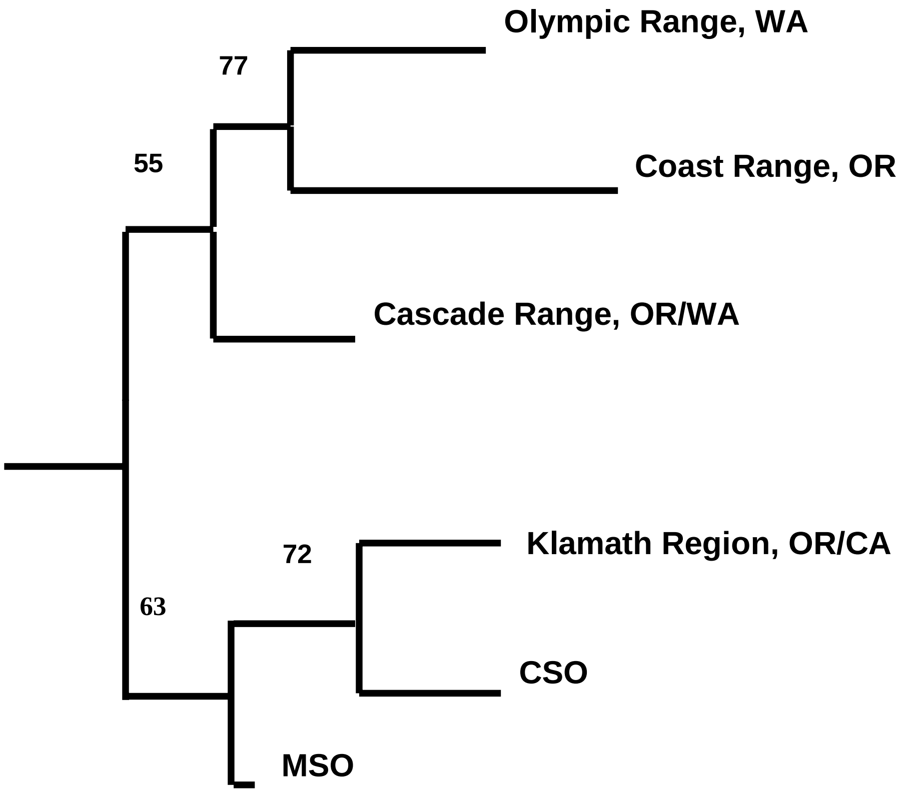


Olympic Range, WA
77
Coast Range, OR
55
Cascade Range, OR/WA
63
Klamath Region, OR/CA
72
CSO
MSO

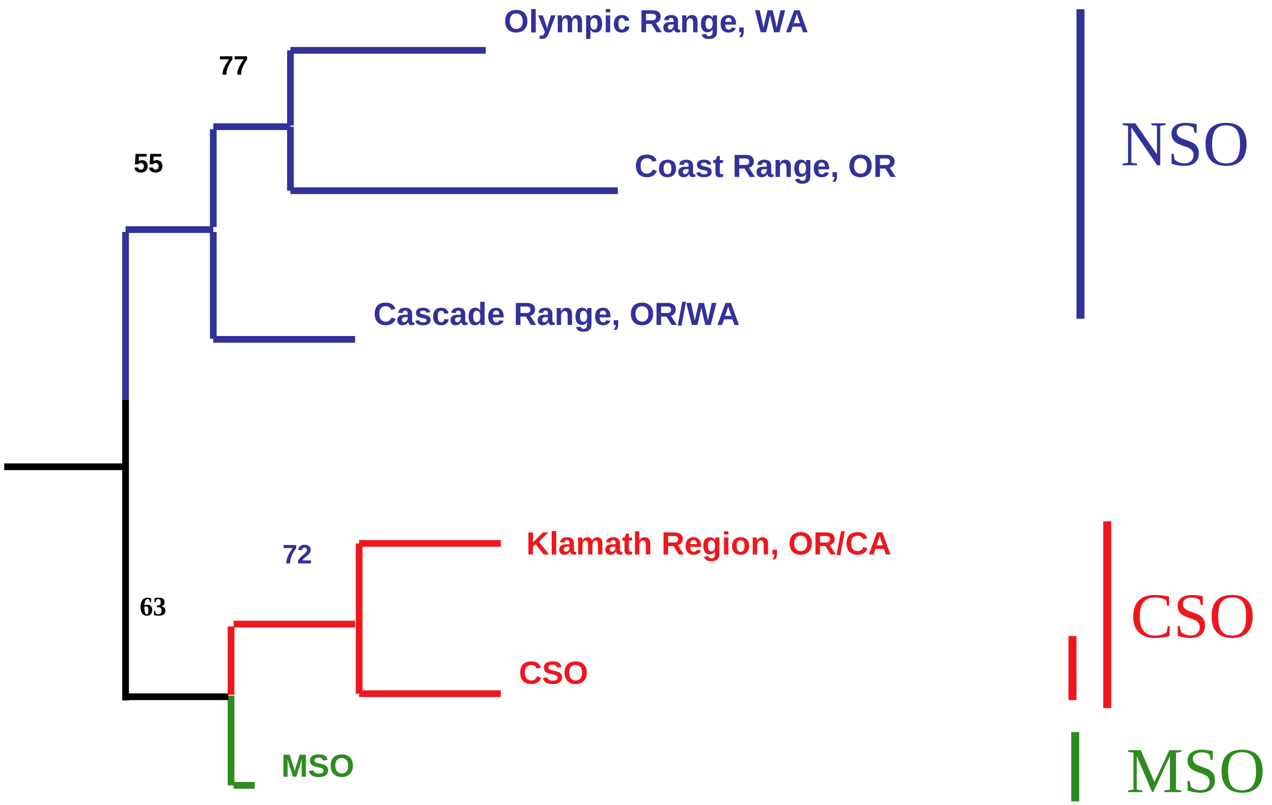
C. Regions


Root
Olympic Range, WA
77
Coast Range, OR
55
Cascade Range, OR/WA
63
Klamath Region, OR/CA
72
CSO
MSO

C. Regions


Olympic Range, WA
77
Coast Range, OR
55
Cascade Range, OR/WA
Klamath Region, OR/CA
72
63
CSO
MSO

C. Regions



Olympic Range, WA
77
55
Coast Range, OR
NSO
Cascade Range, OR/WA
Klamath Region, OR/CA
72
63
CSO
CSO
MSO
MSO

Genetic studies: Nuclear Markers; microsatellites

Henke et al., unpublished report (part of Masters Thesis work at San Jose State University).

- Four populations examined; two in the traditional NSO range (Marin n = 38 individuals; Mendocino n = 36); and two in the CSO range (El Dorado n = 41; Monterey n = 38); Assessed microsatellite variation (six loci)

- G_{ST} estimated within each subspecies is 0.055 for NSO (between Marin and Mendocino) and 0.152 for CSO (between El Dorado and Monterey). G_{ST} between subspecies averages 0.351 across the pairwise comparisons among subspecies, and ranges between 0.172 and 0.535. All values were significantly different than 0, and some were quite large for within species comparisons.

- 150 of 153 birds clustered with others from their own subspecies, again supporting the 75% rule

Summary

- Morphology - subspecific differences have been noted, but are not carefully tested or they appear to be clinal, and no clear geographic boundary or diagnostic characters are available
- Behavior - suggests there are significant differences in location call among NSO, CSO, and MSO
- mtDNA - ALL THREE studies agree that…
    - majority of haplotypes are found in only one subspecies geographical range (ie. are diagnostic)
    - deep clade differences between common subspecies haplotypes
    - Evidence of gene introgression (dispersal or hybridization) somewhere between 1% to 11%, depending upon criteria and sampling (ie. meets the 75% rule by any standards)
- Nuclear genes
    - General corroboration of mtDNA results,
    - but more ambiguous due to relatively less power (RAPDs) and smaller sampling scope (microsatellites), and expectations that coalescence takes longer

Conclusions:

- SO’s have better evidence for and delineation of subspecies than the vast majority of North American polytypic bird species

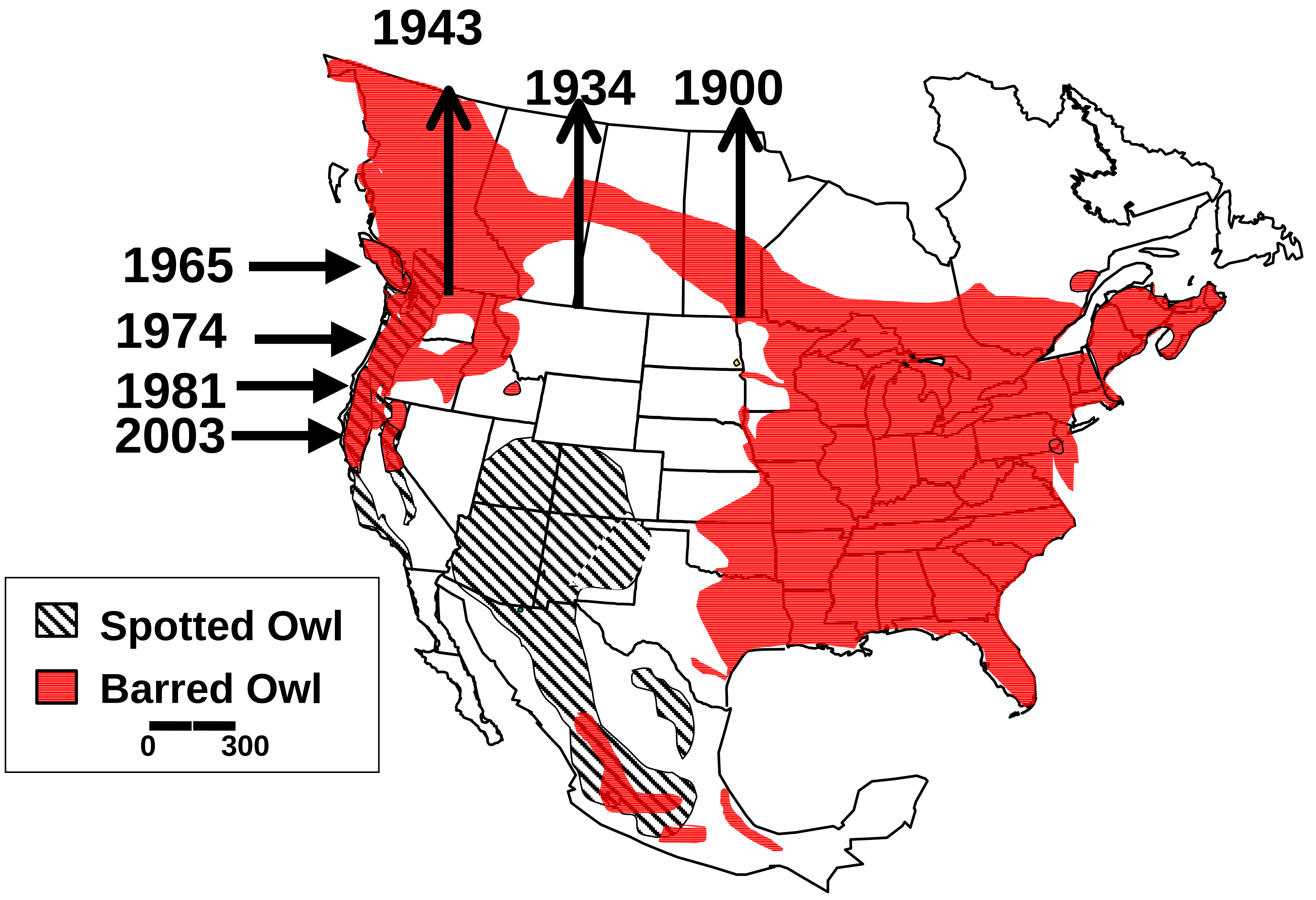
- Subspecies are well supported, and also meet requirements for ESUs, Management Units, and probably even Phylogenetic Species

- Subspecies show similar levels of hybridization as many currently-classified Biological Species

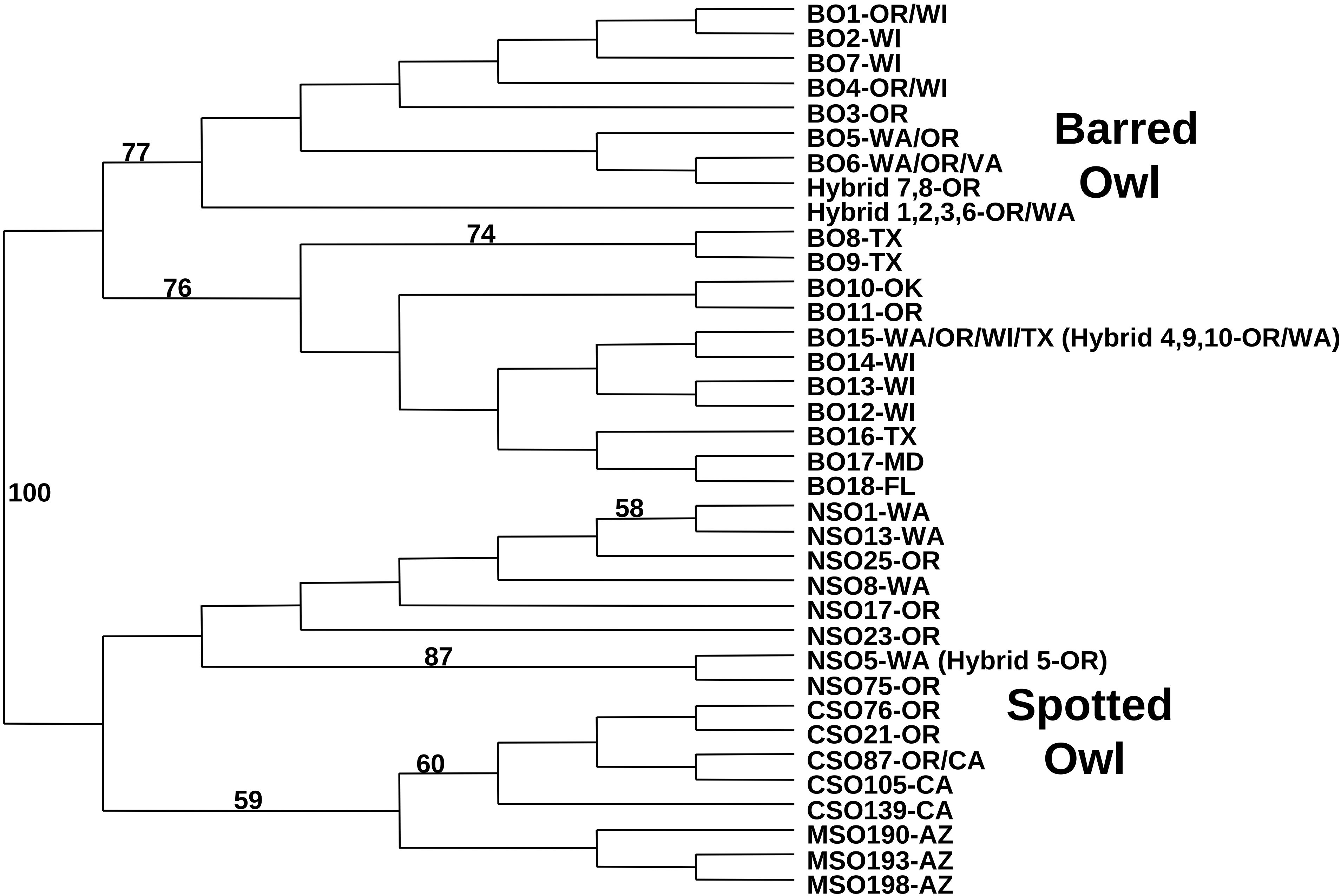
- By all measurements, these fit the 75% rule

Information needs

- Information about hybrid area
  - Studies of morphological characters that are diagnostic
  - Understanding of the fitness and directionality of introgression
  - Additional measurement of gene flow
    - Among subspecies (especially within hybrid zones)
    - Within subspecies among distant or fragmented forests
  - Genetic loss or dilution along hybrid zones?



1943
1934
1900
1965
1974
1981
2003
Spotted Owl
Barred Owl
0
300


BO1-OR/WI
BO2-WI
BO7-WI
BO4-OR/WI
BO3-OR
BO5-WA/OR
BO6-WA/OR/VA
Hybrid 7,8-OR
Hybrid 1,2,3,6-OR/WA
BO8-TX
BO9-TX
BO10-OK
BO11-OR
BO15-WA/OR/WI/TX (Hybrid 4,9,10-OR/WA)
BO14-WI
BO13-WI
BO12-WI
BO16-TX
BO17-MD
BO18-FL
NSO1-WA
NSO13-WA
NSO25-OR
NSO8-WA
NSO17-OR
NSO23-OR
NSO5-WA (Hybrid 5-OR)
NSO75-OR
CSO76-OR
CSO21-OR
CSO87-OR/CA
CSO105-CA
CSO139-CA
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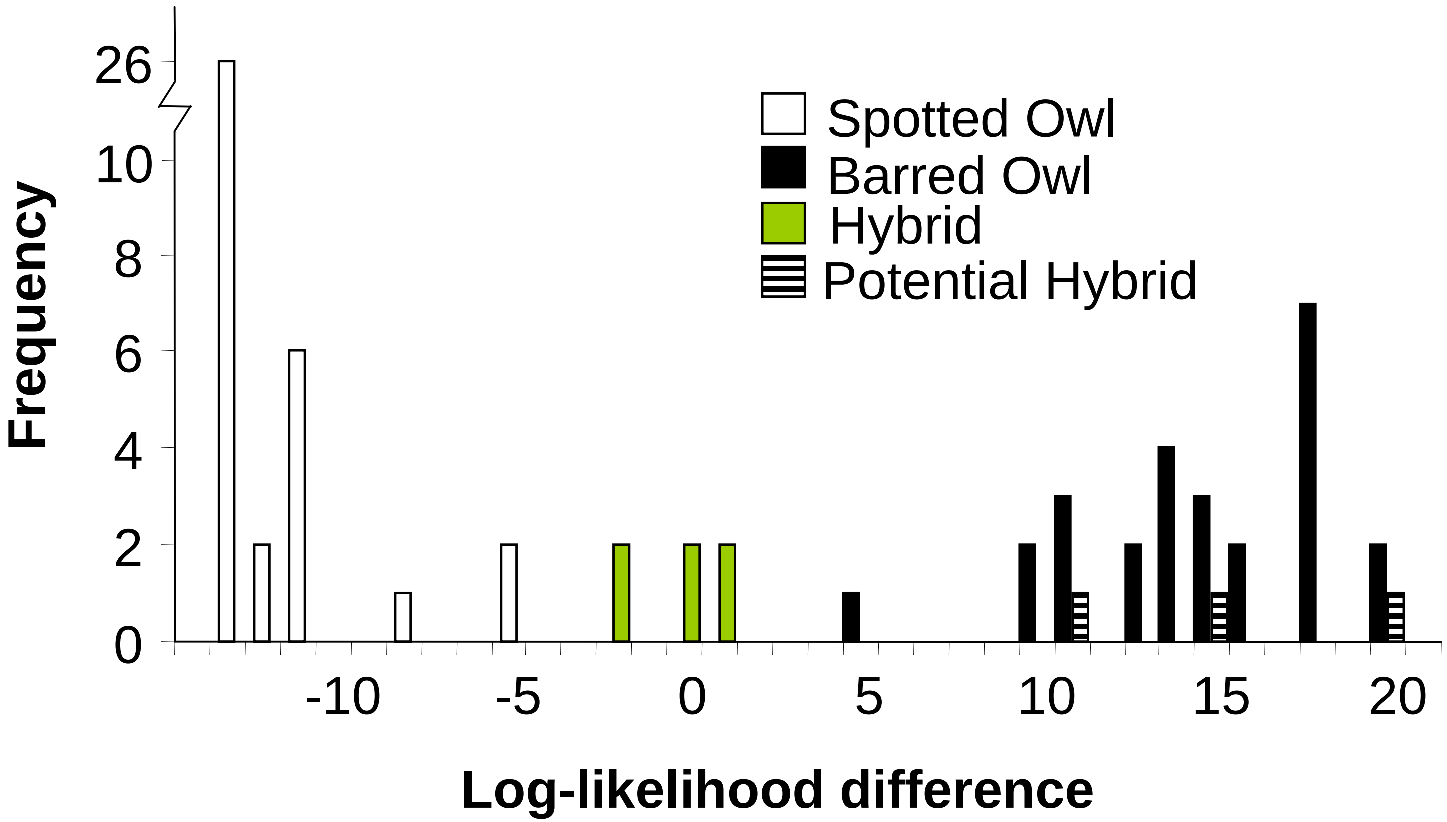
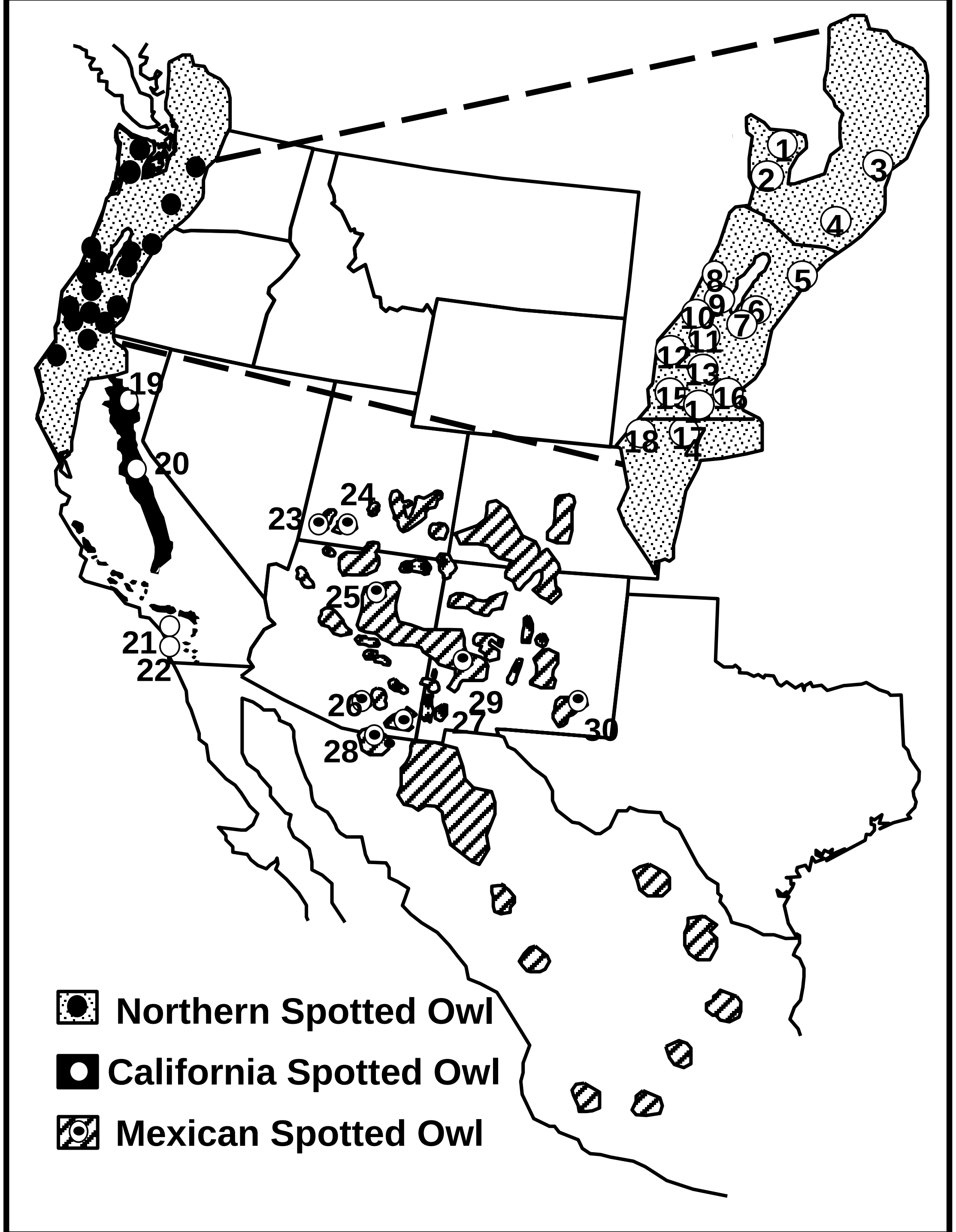
MSO193-AZ
MSO198-AZ
Barred Owl
Spotted Owl
77
74
76
100
58
87
60
59



Spotted Owl
Barred Owl
Hybrid
Potential Hybrid
Frequency
26
10
8
6
4
2
0
-10
-5
0
5
10
15
20
Log-likelihood difference



1
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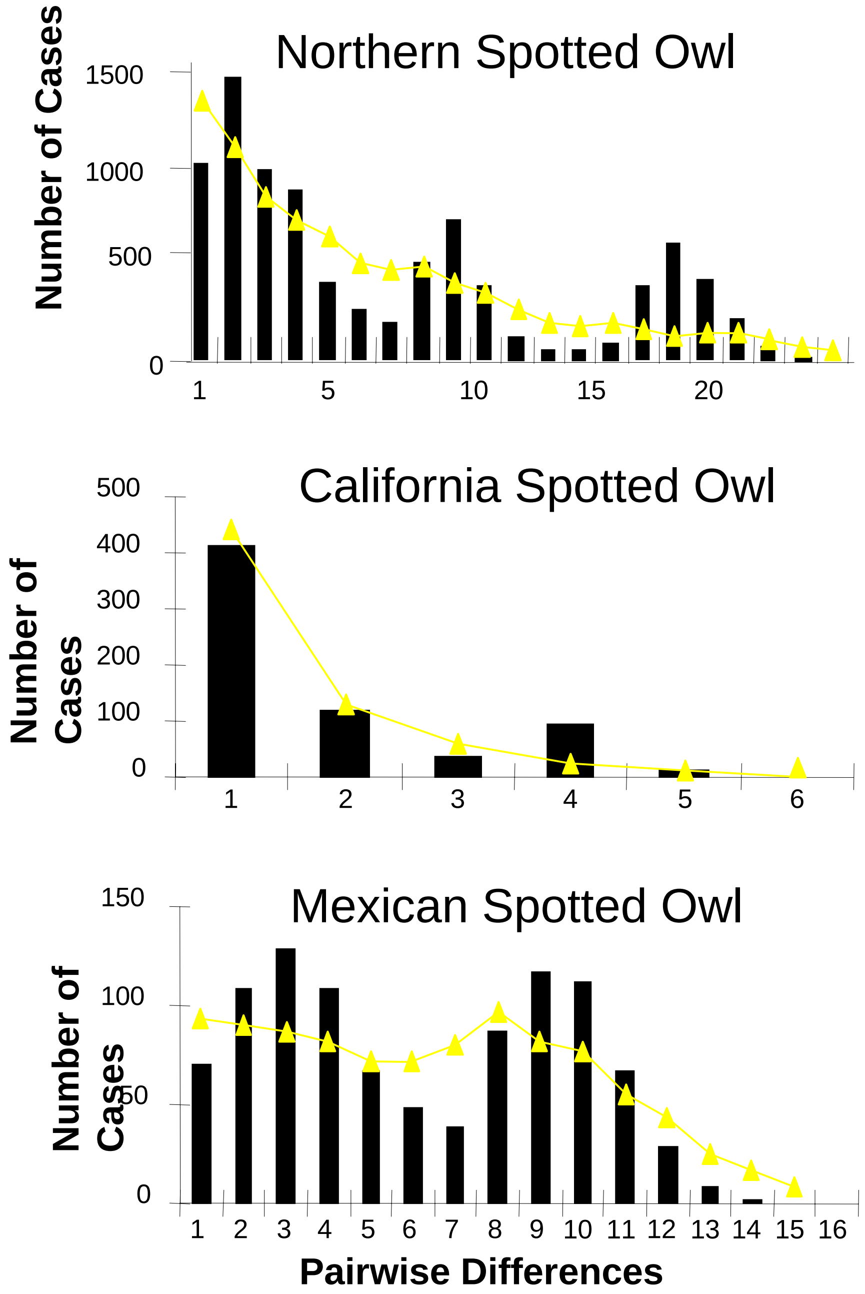
30
Northern Spotted Owl
California Spotted Owl
Mexican Spotted Owl


Northern Spotted Owl
Number of Cases
1500
1000
500
0
1
5
10
15
20
California Spotted Owl
Number of Cases
500
400
300
200
100
0
1
2
3
4
5
6
Mexican Spotted Owl
Number of Cases
150
100
50
0
1
2
3
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11
12
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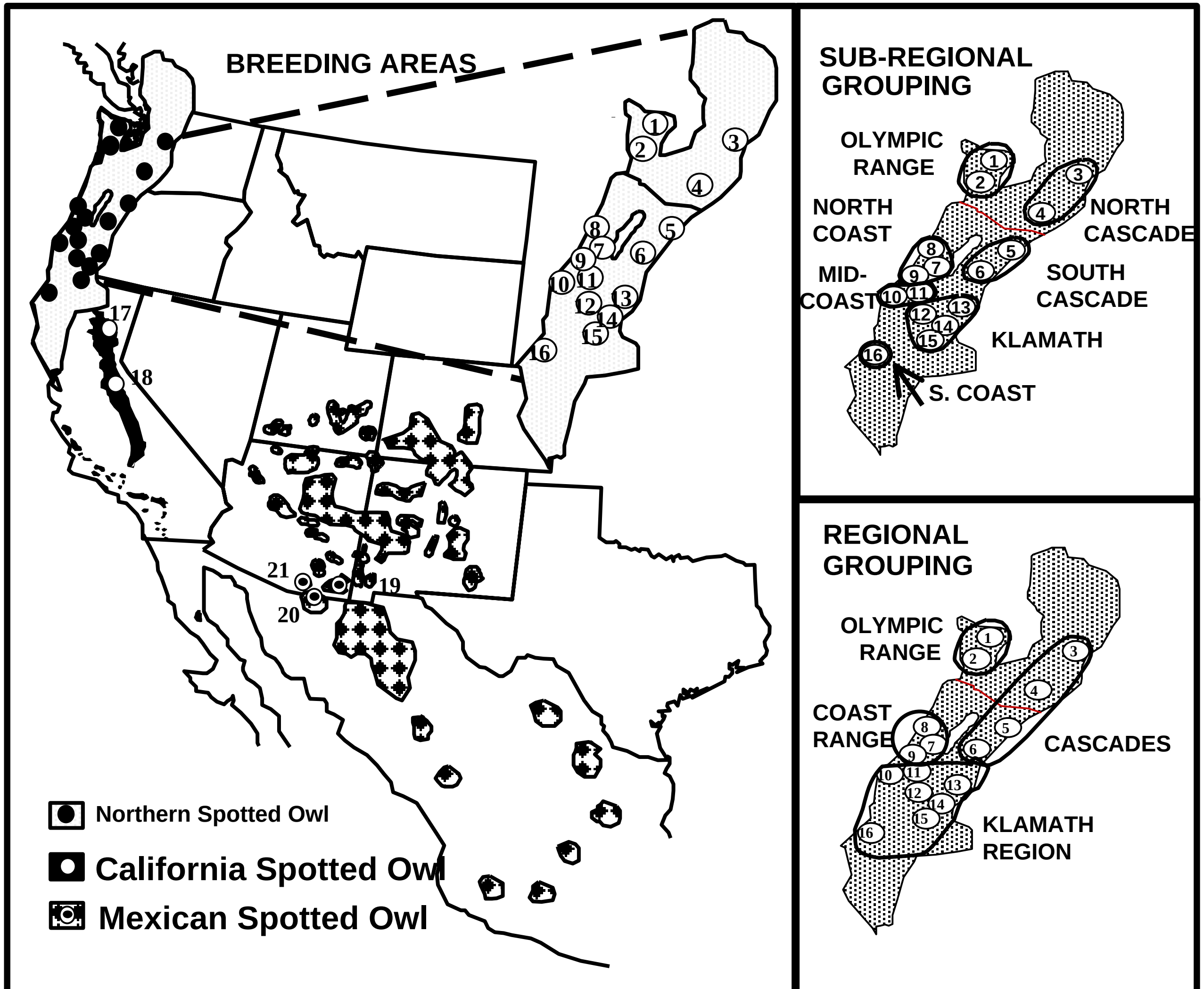
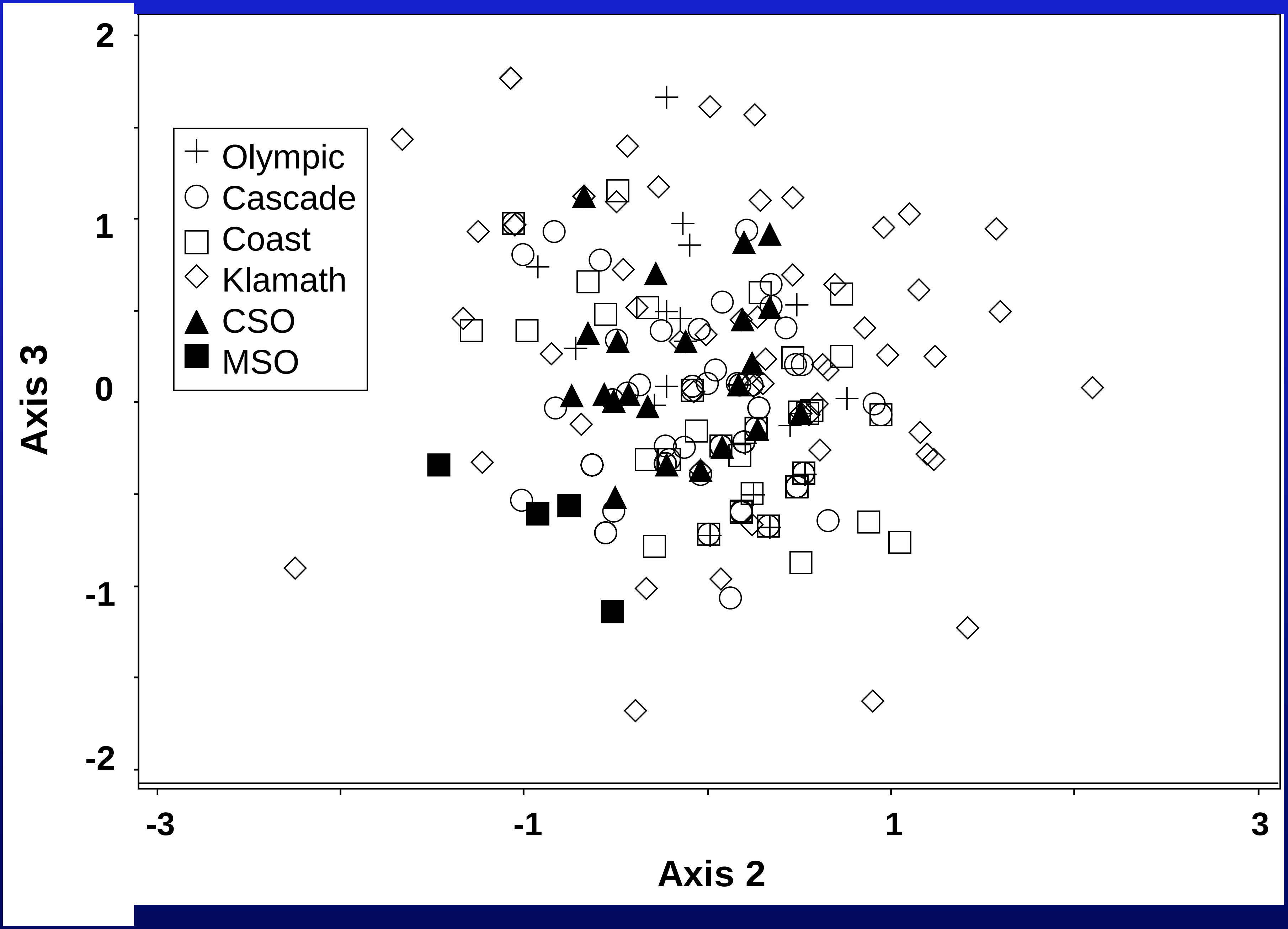
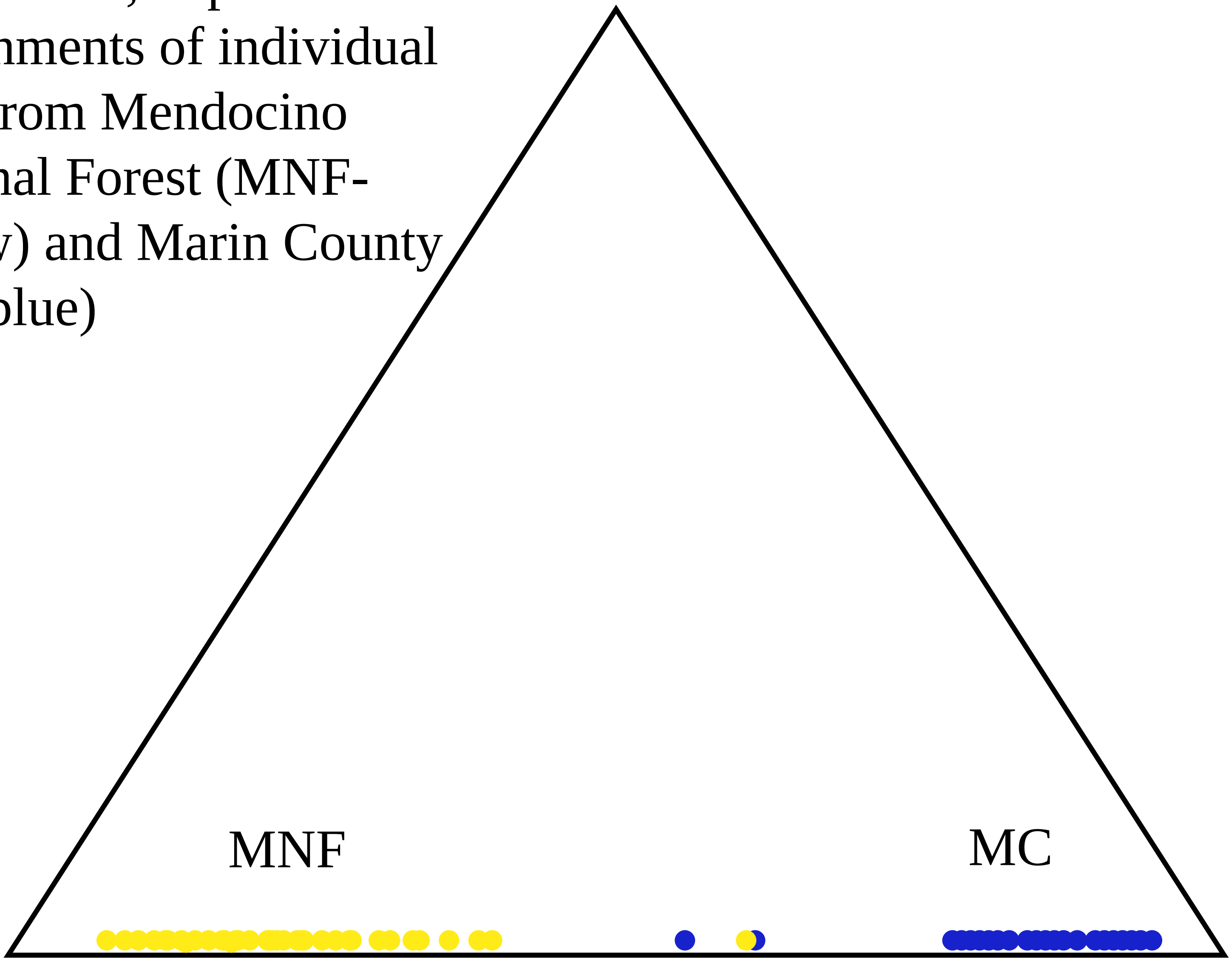
14
15
16
Pairwise Differences


BREEDING AREAS
Northern Spotted Owl
California Spotted Owl
Mexican Spotted Owl
SUB-REGIONAL GROUPING
OLYMPIC RANGE
NORTH COAST
NORTH CASCADE
MID-COAST
SOUTH CASCADE
KLAMATH
S. COAST
REGIONAL GROUPING
OLYMPIC RANGE
COAST RANGE
CASCADES
KLAMATH REGION



Olympic
Cascade
Coast
Klamath
CSO
MSO
Axis 3
Axis 2
2
1
0
-1
-2
-3
-1
1
3

Henke et al., unpublished. Assignments of individual owls from Mendocino National Forest (MNF-yellow) and Marin County (MC-blue)



Summary and Future Work

Summary and Future Work

Genetic factors:

- Status of the subspecies (or "phylogenetic species" or DPS or ESU or MU).
- extent of introgression (and hybrid zone) between NSO and CSO
- Extent of introgression and hybridization between Barred and Spotted Owls.
- Historic processes reflected in genetic data (long-term isolation, Pleistocene refugia).
  - Historical population size, gene flow, adaptive evolution
  - Effects of fragmentation, isolation, inbreeding, small N_e and low genetic variation.
- Current management practices (Forest Plan) and prediction of impacts on genetic structure.