

Using Genetics to Address Conservation Issues on the Kenai Peninsula



The grizzly or brown bear (Ursus arctos) population on the Kenai Peninsula in south-central Alaska is a lineage-specific (Untergesetz Russischer Bären, Bear Study Team [IBRST] 2003). Brown bears on the Kenai Peninsula exhibit plant distribution and abundance through seed dispersal as frugivores, transport name-derived nutrients into terrestrial ecosystems through salivary excretion (Hilderbrand et al. 1999), and possibly regulate ungulate populations through avoidance, predation under certain conditions (Zager and Bachman 2006). Brown bears are recognized as a source of concern by residents and visitors as a source of recreation for commercial wildlife viewing and hunting, damage, and a welfare issue (Alaska Department of Fish and Game [ADF&G] 2001).

The 16-km wide isthmus that separates the 24,300-km² Kenai Peninsula from the adjacent mainland restrict brown bear migration and interisland gene flow (Jackson et al. 2008). Using microsatellite and mitochondrial DNA (mtDNA), Jackson et al. (2008) verified that the Kenai brown bear population is isolated, reflecting lower mtDNA haplotypic diversity than some other brown bear populations on mainland Alaska but similar to other peninsular populations.

The Kenai Peninsula is also one of the fastest urbanizing areas in Alaska, with approximately 10,000 new residents added every decade since 1980 (<http://www.census.gov/population/counts/ak030011st.html>; accessed 23 Sep 2015). Over these same 5 decades, brown bears have declined in defense of $<1\%$ of property (D.L. Jones and D.L. Price 2002; Zehrtz 2012). Legal harvest of brown bears has varied with hunting regulations over this same period, ranging from 0/day during much of the past decade to 64 individuals in 2014. In 2013, the year of

Canadian Journal of Forest Research
OPEN ACCESS | Article
The dynamics of a changing Lutz spruce (*Picea × lutzii*) hybrid zone on the Kenai Peninsula, Alaska
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Abstract: We investigated the genetic makeup of Lutz spruce, a natural hybrid between white and Sitka spruce on the Kenai Peninsula, Alaska. Microsatellites indicate 73% of individuals sampled had predominantly white spruce ancestry, whereas 16% had predominantly Sitka spruce ancestry; some individuals classified as white spruce had Sitka spruce plastid genotypes. As Ptna autochthonous are maternally inherited and plastids are paternally inherited, it appears that white spruce was the ancestral seed parent of nearly all spruce on the western peninsula, whereas Sitka spruce alleles originated from pollen. Pollen records show that white spruce colonized the western peninsula ~8300 YBP from glacial refugia, whereas Sitka spruce arrived on the eastern peninsula ~4000 YBP after migrating up the Pacific coast. Our data suggest that Sitka spruce colonization west of the Kenai Mountains may have occurred *in situ* seed dispersal but by long-distance transport of wind-borne pollen and subsequent hybridization with established white spruce populations. Hybridization was an important mechanism that allowed Sitka spruce to expand the leading edge of its range in response to historical climate change. As the climate continues to warm, climate envelope modeling suggests Lutz spruce may ultimately displace white spruce on the western peninsula even as Sitka spruce is constrained to the eastern peninsula where it will continue to hybridize.
Key words: Alaska, climate change, DNA analysis, hybridization, Lutz spruce, *Picea × lutzii*
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Keywords: Alaska, brown bear, genetics, hair DNA, Kenai Peninsula, mark-recapture, population, spatial results, brown bears

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Research Article
Open Access
Toward conserving natural diversity: A biotic inventory by observations, specimens, DNA barcoding and high-throughput sequencing methods
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<https://doi.org/10.5281/zenodo.850124>

Abstract: The Kenai National Wildlife Refuge has been given a broad conservation mandate to conserve natural diversity. A prerequisite for fulfilling this purpose is to be able to identify the species and communities that make up that biodiversity. We tested a set of varied methods for inventory and monitoring of plants, birds and terrestrial invertebrates on a grid of 40 sites in a 938 ha study area in the Silkok Creek watershed, Kenai Peninsula, Alaska. We sampled plants and lichens through observation and specimen-based methods. We surveyed birds using bird call surveys on variable

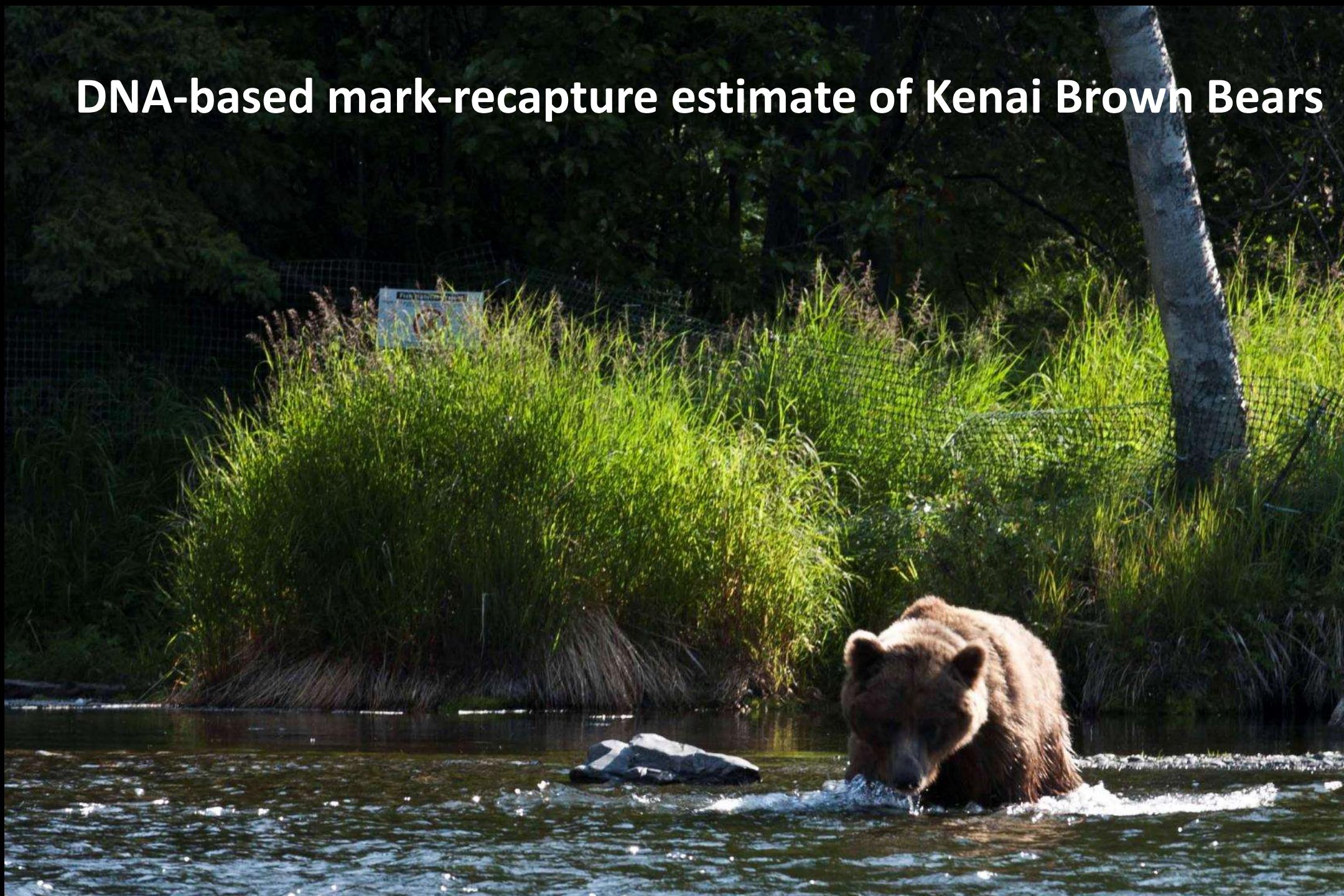
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John Morton, PhD



Brown bear population	Spruce hybridization	Species richness
Hair (DNA)	Leaves (needles)	Organism (any part)
Marker G10J (species)	12 microsatellite loci (species)	313 bp region of COI (cytochrome c oxidase) gene mitochondrial DNA (mtDNA)
Multilocus (7) microsatellite markers (individuals)	Nad5a (mitochondrial [♀] locus) Trnt-L (chloroplast [♂] locus)	
Amelogenin marker (♀♂)		

DNA-based mark-recapture estimate of Kenai Brown Bears



Why we estimated the Kenai brown bear population in 2010...

- ✓ Designated *Population of Special Concern* by State of Alaska in 1998 – 2010
- ✓ Genetically less diverse than (and distinct from) adjacent mainland Alaskan brown bears
- ✓ Only estimate of 250-300 brown bears based on multiplying the area of suitable habitat by mean bear density from other AK studies
- ✓ Population trend unknown ($\lambda = 0.9364 - 1.0588$)
- ✓ Low yearling survivorship and small proportion of subadult females suggested low recruitment
- ✓ Annual DLPs increased from <1 in 1960s to 5 in 1990s to >20 in 2000s

Simple Lincoln-Petersen Estimator

$$N = MC/R$$

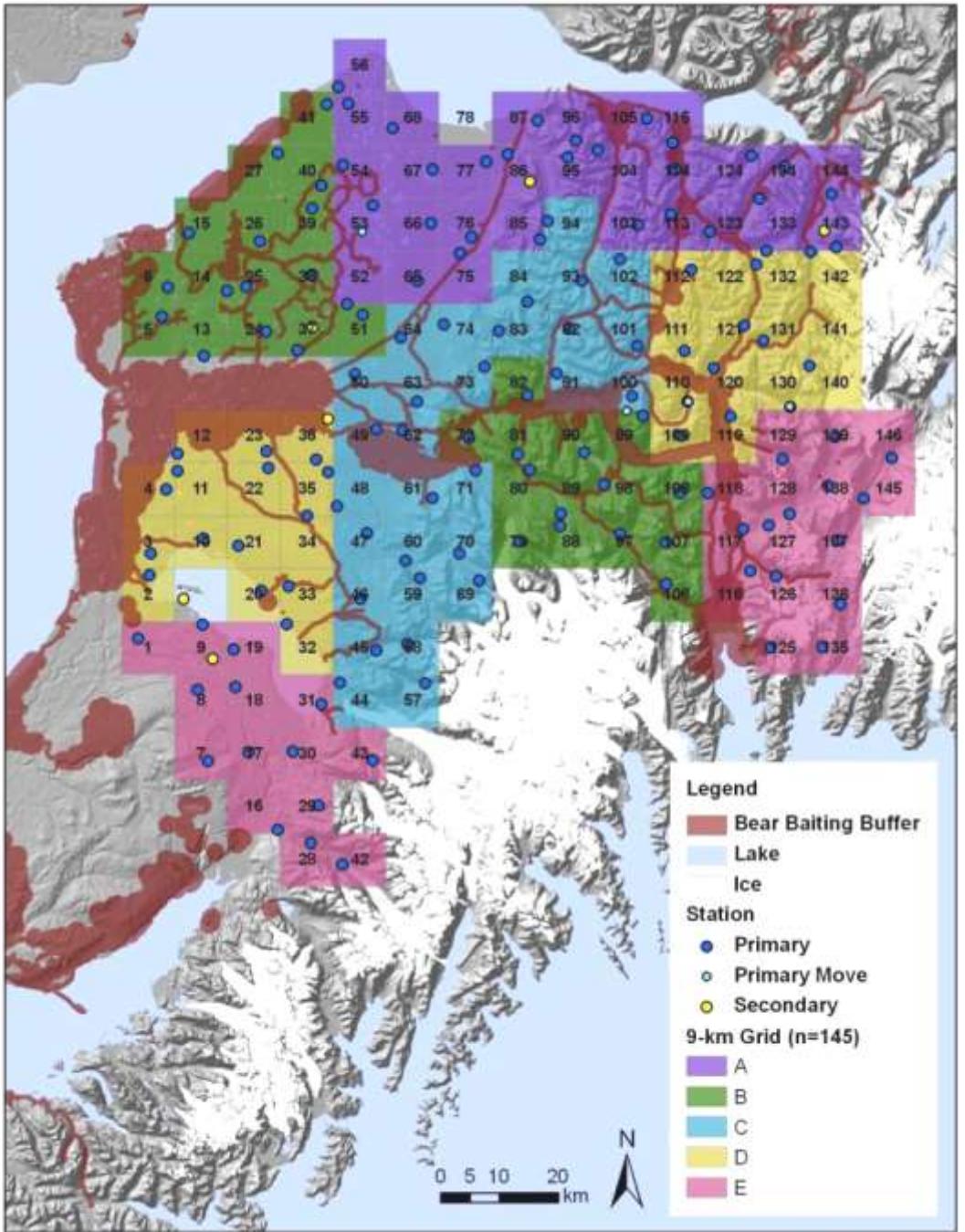
N = Estimate of total population size

M = Total number of animals captured and marked on the first visit

C = Total number of animals captured on the second visit

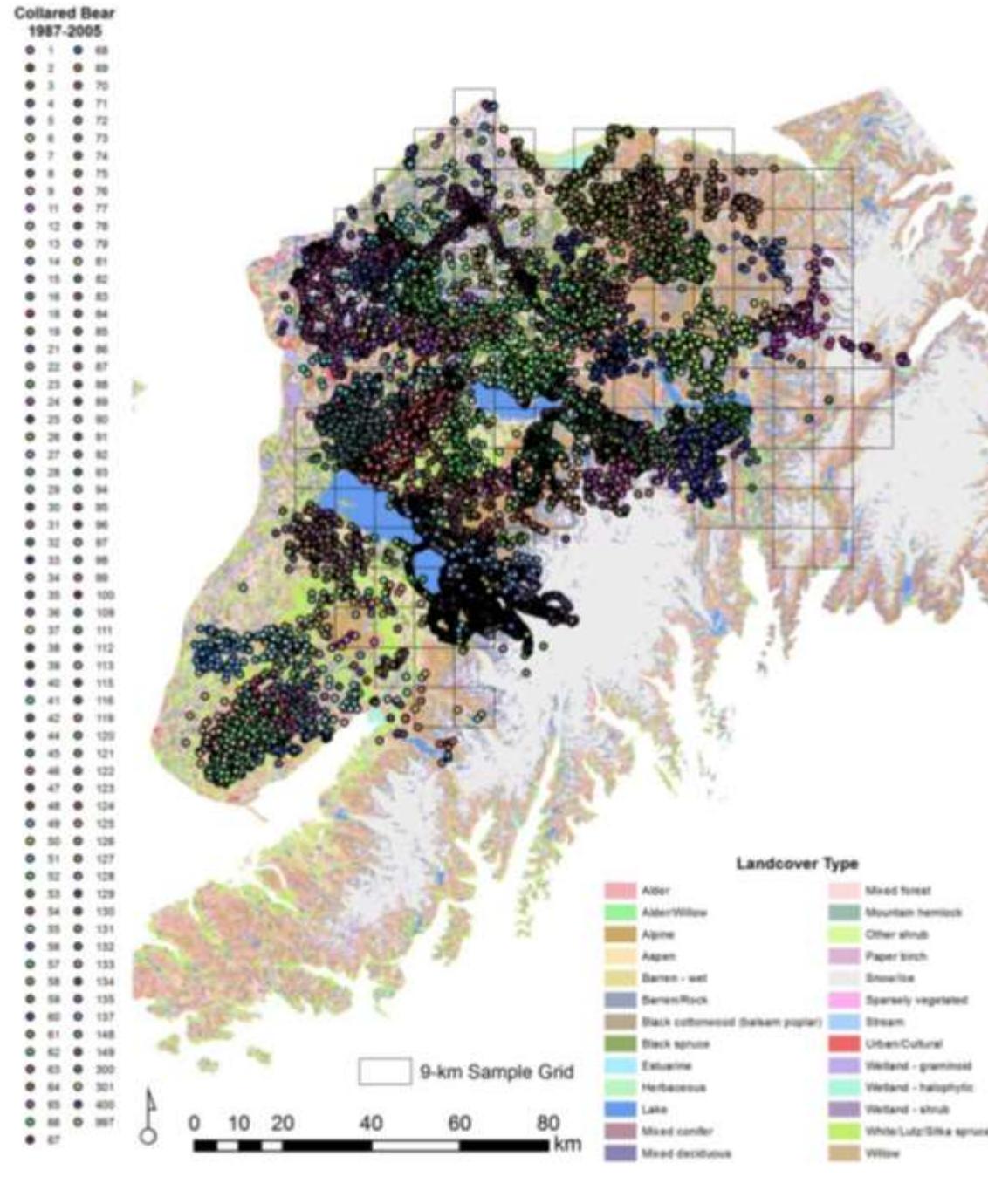
R = Number of animals captured on the first visit that were then recaptured on the second visit

**...estimate is not based on the number of individuals marked (i.e., genotypes)
but on their recapture rates**



**145 primary hair stations
subjectively placed within 81-km²
cells systematically distributed over
11,700 km² study area**

**29 stations sampled daily over five
5-day trap sessions using rotating
panel design**



**Distribution of 144,024
telemetry locations from 125
female brown bears with GPS
and VHF collars (1987-2005)**

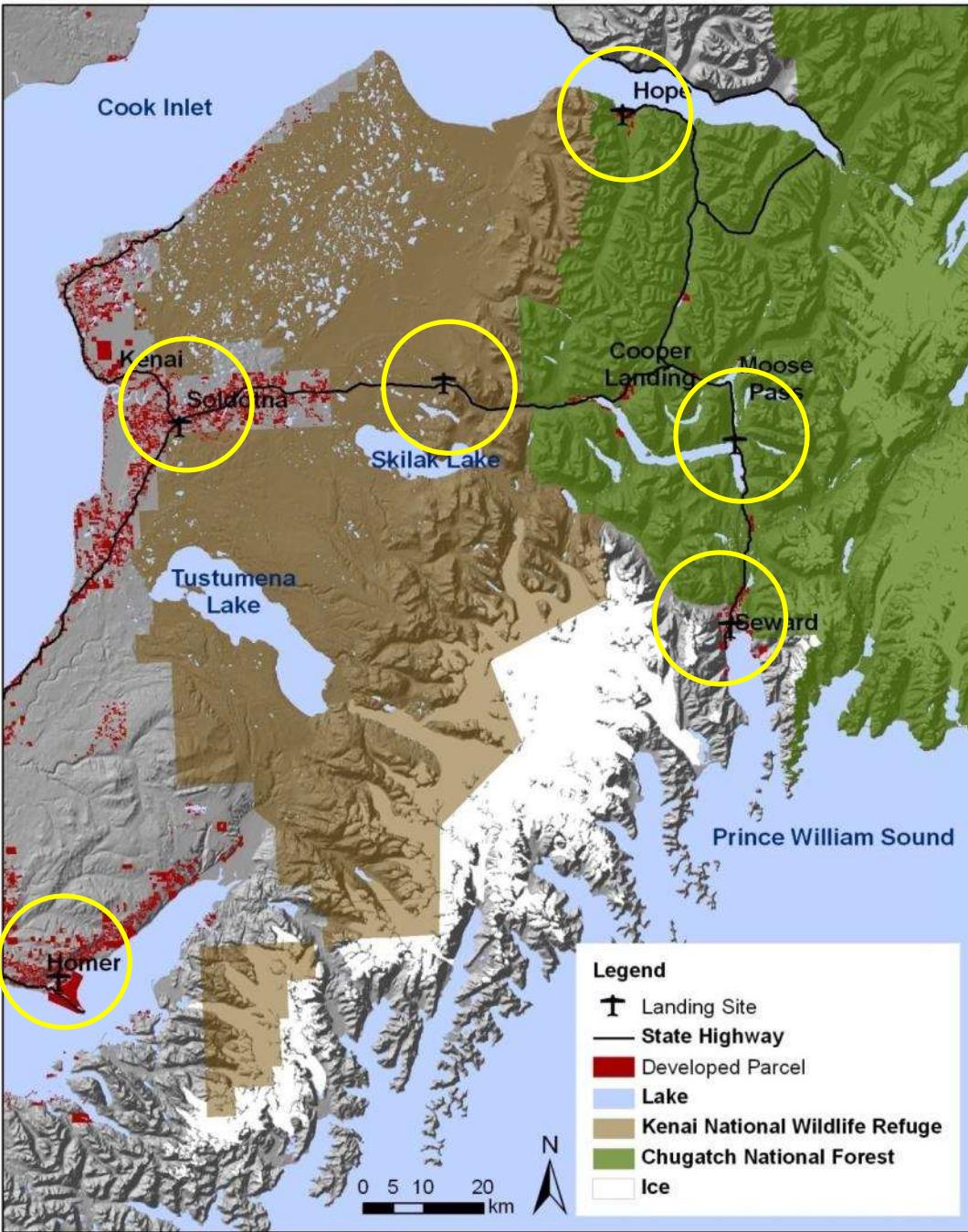
► 87% were on study area

Selection criteria for hair stations

- ✓ adequate space for helicopter access
- ✓ > 400m from trails, cabins, roads
- ✓ riparian/wetland corridors
- ✓ other travel corridors (ridges, shoulders, chutes)
- ✓ OTBE, ensure good spatial separation among sites within a cell







Four sets of 2-person field crews operated out of Moose Pass and Soldotna for 31 consecutive days













1MINUTE

07

JUN.19,10 08:00 AM



1MINUTE

07

JUN.19,10 08:01AM



1MINUTE

07

JUN.19,10 08:04 AM



1MINUTE

07

JUN.19,10 08:05 AM



1MINUTE

07

JUN.19,10 08:06 AM



1MINUTE

07

JUN.19,10 08:09 AM



1MINUTE

07

JUN.19,10 08:10 AM



1MINUTE

07

JUN.19,10 08:11AM

Hair station: _____ Barb # _____ Session # _____
Date: June _____, 2010
3 hairs or less? Y / N
Hair Location: Upper strand / Lower strand
Probable species: Brown bear / Black bear / unknown
Comments: _____

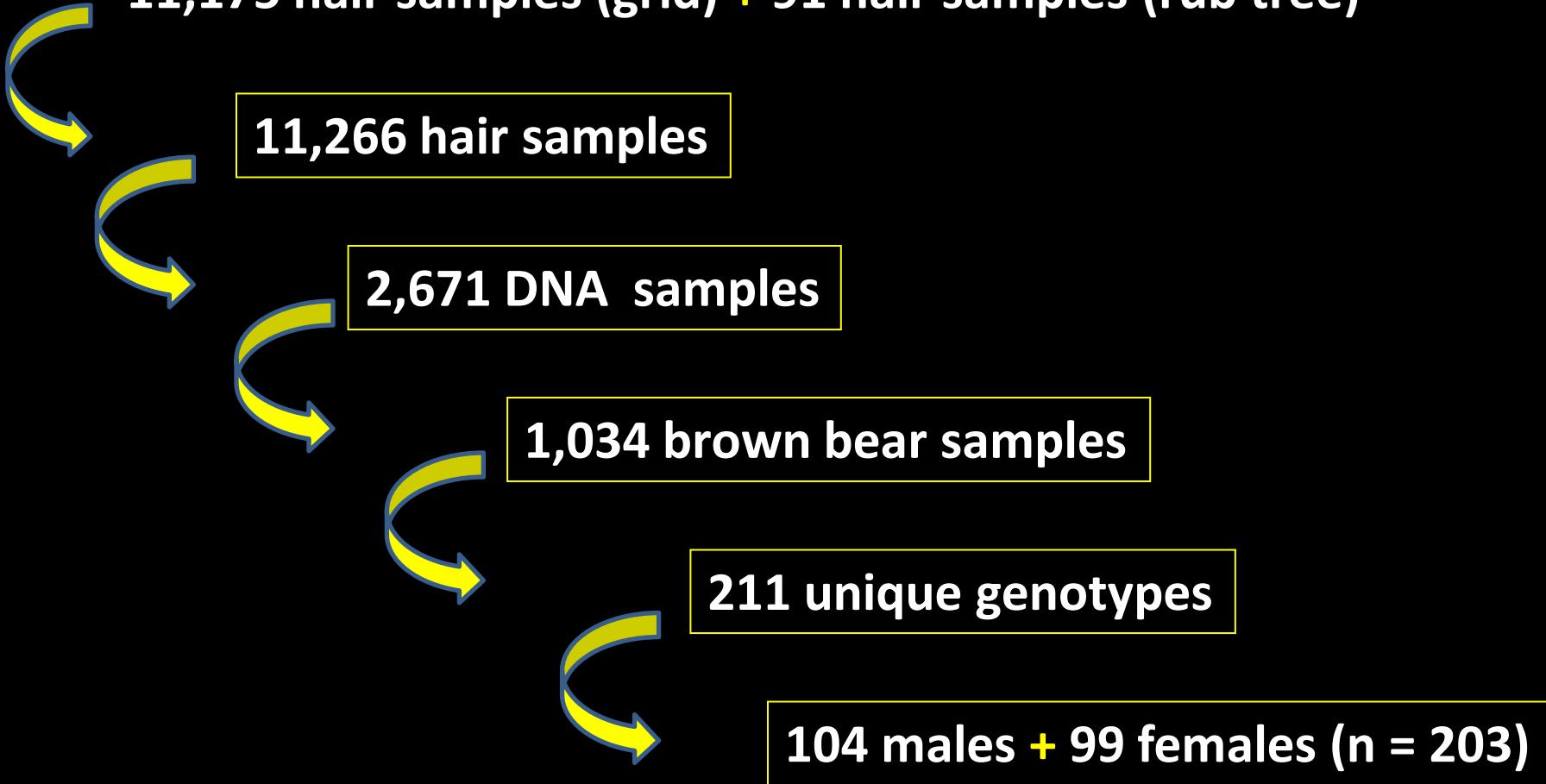
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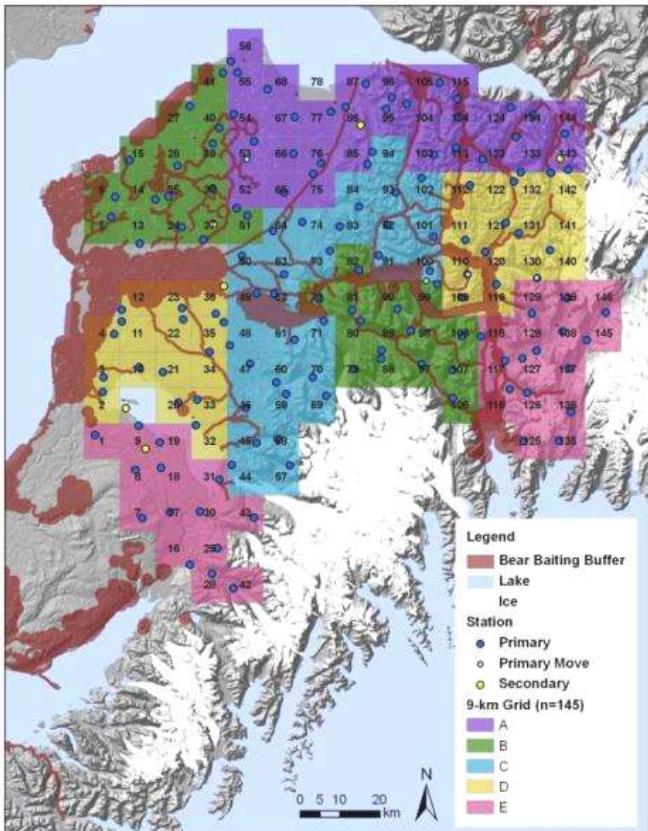


11,175 hair samples (grid) + 91 hair samples (rub tree)



How many brown bears on the Kenai Peninsula in 2010?

**428 (353-539) brown bears estimated (all ages) on 11,700 km² sample frame
of which 10,200 km² is available habitat**



≈ 42 bears per 1,000 km²

≈ 582 bears on the KP (469-719)

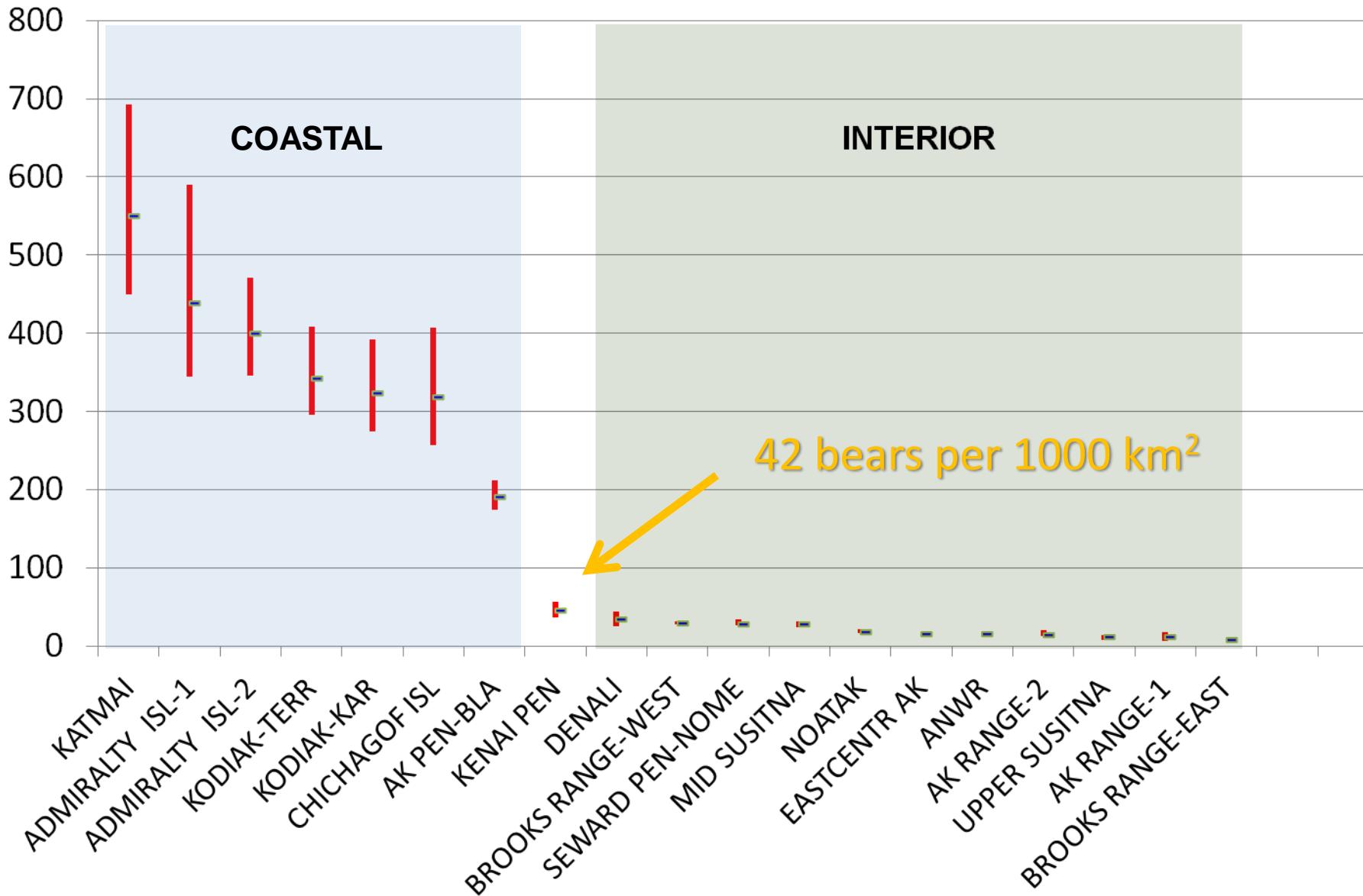
≈ 188 adult females + 188 adult males

≈ 206 dependent young

(Morton et al. 2016)

ALASKA BROWN BEAR DENSITIES (PER 1000 KM²)

(after Miller et al. 1997)

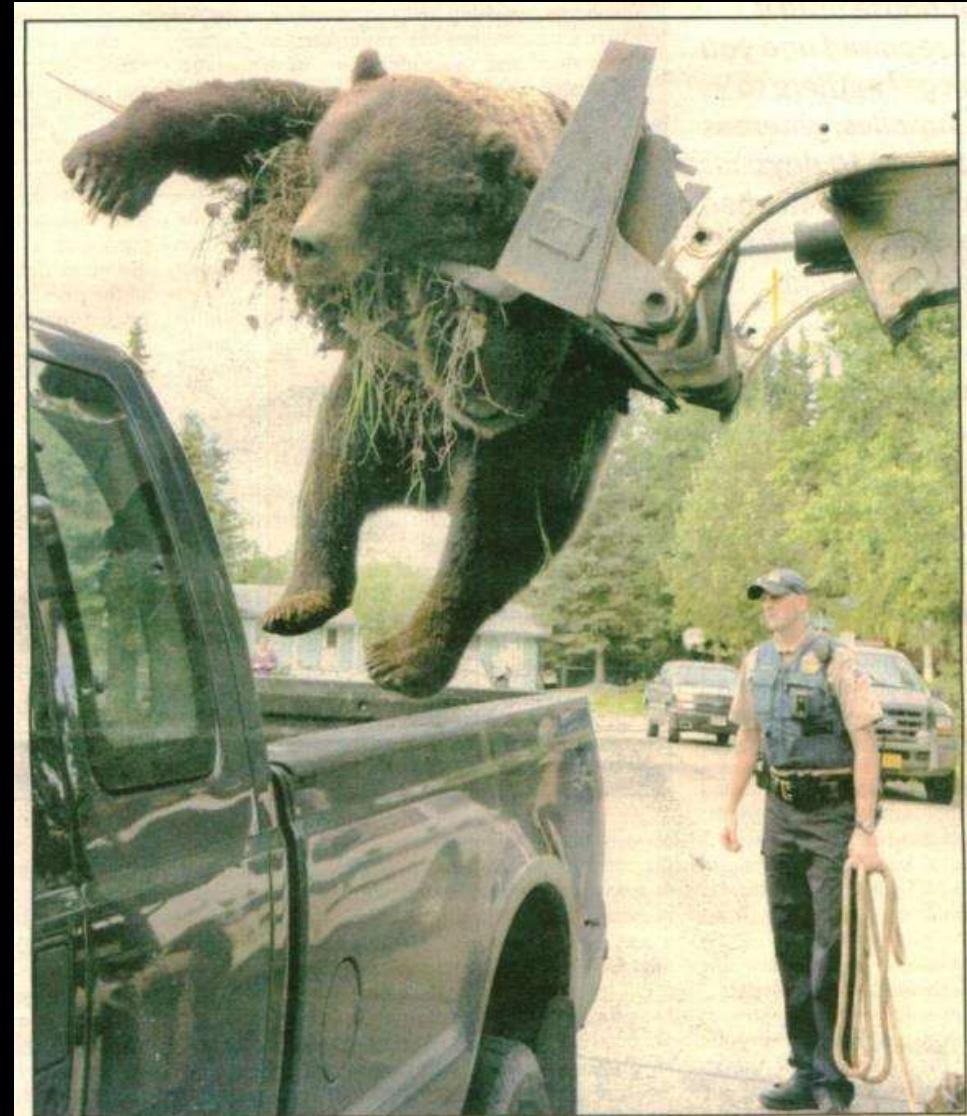


What are the management implications for Kenai brown bears?

Represents 1st empirically-based estimate of the Kenai brown bear population

Puts human-caused mortality (legal harvest, illegal take, vehicle collisions, agency kills and DLPs) into better demographic context

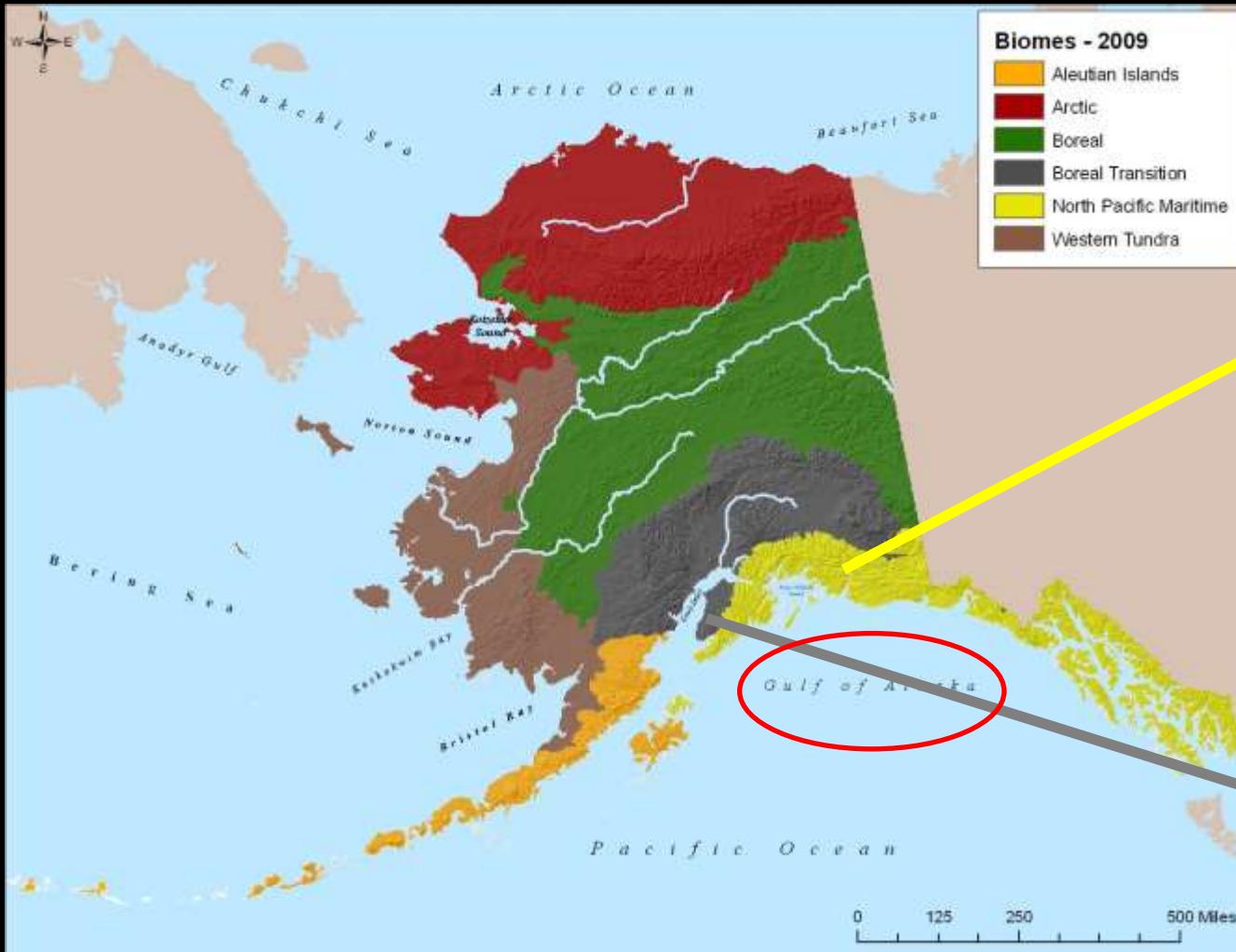
Helps determine sustainable harvest



Why care about Lutz spruce?



Kenai Peninsula is the nexus of two biomes





White spruce arrived first
8500 YBP



Sitka spruce arrived later
4000 YBP



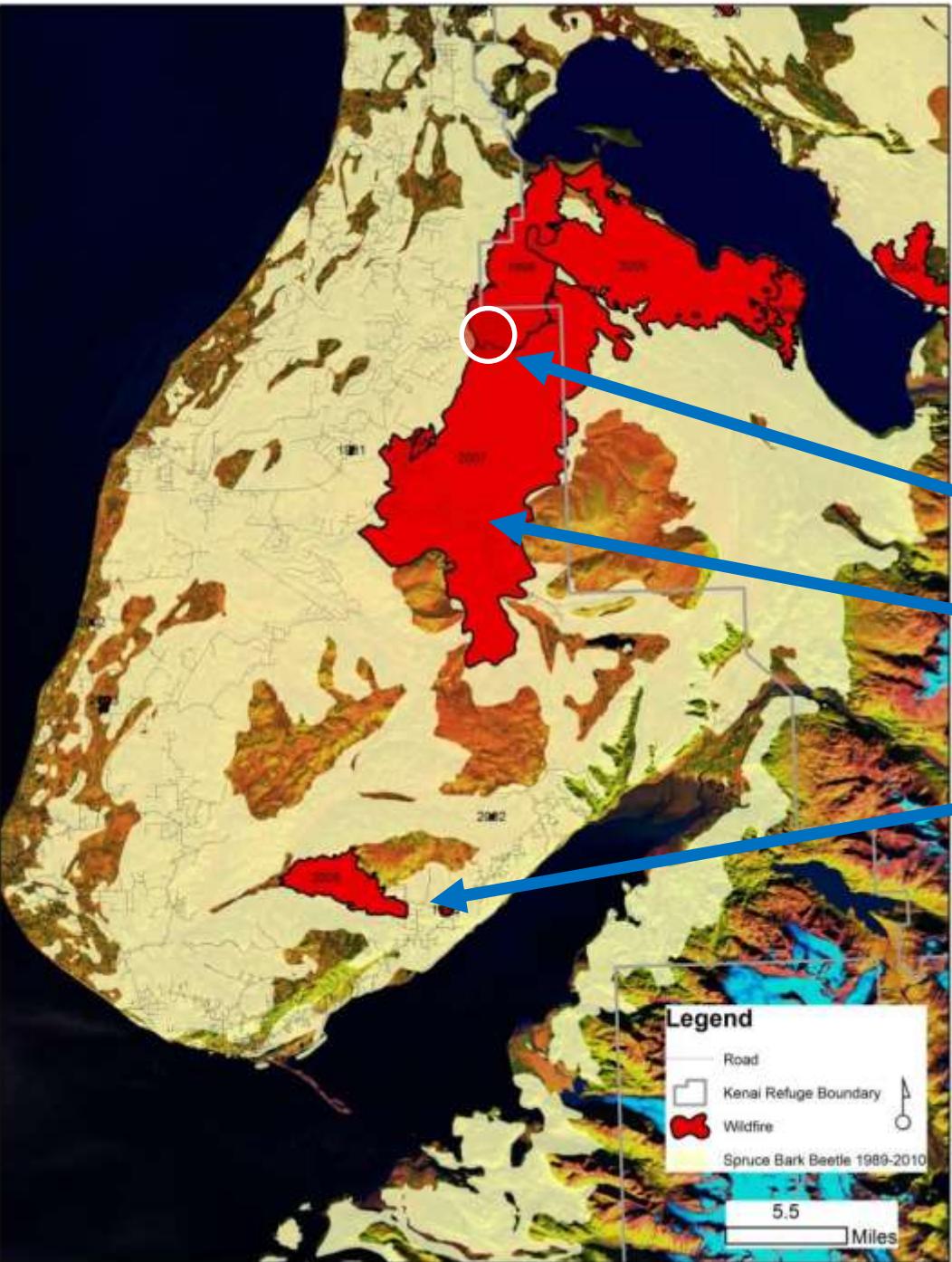
White spruce (1)



Lutz spruce (3-4)



Sitka spruce (6-7)



Southwestern Kenai Peninsula was the epicenter of 1990s spruce bark beetle outbreak

- 2019 Tustumena Lake**
- 2007 Caribou Hills**
- 2005 Tracy Avenue**
- 2005 Fox Creek**
- 2005 Glacier Creek**
- 1996 Crooked Creek**
- 1994 Windy Point**

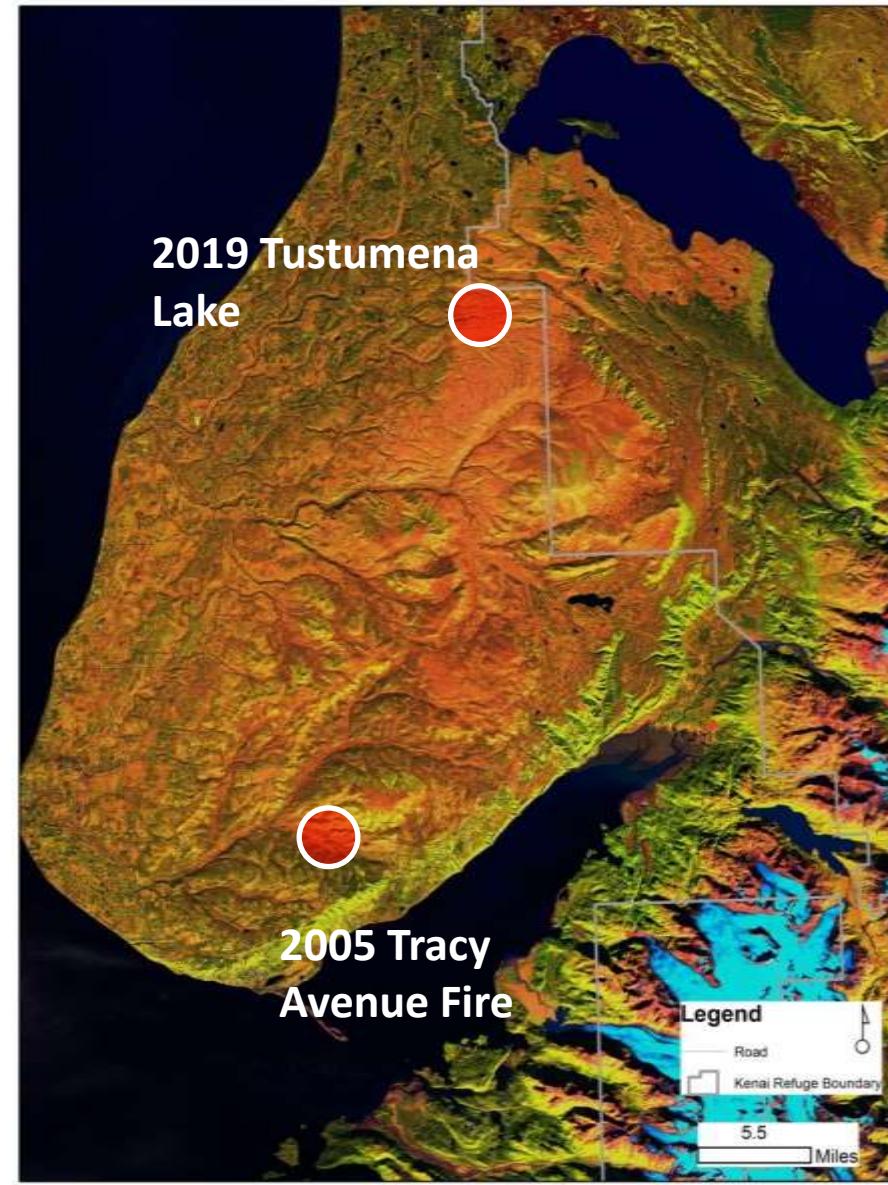
First lightning-caused grassland fire in spring on Kenai Peninsula

2019 Tustumena Lake Fire





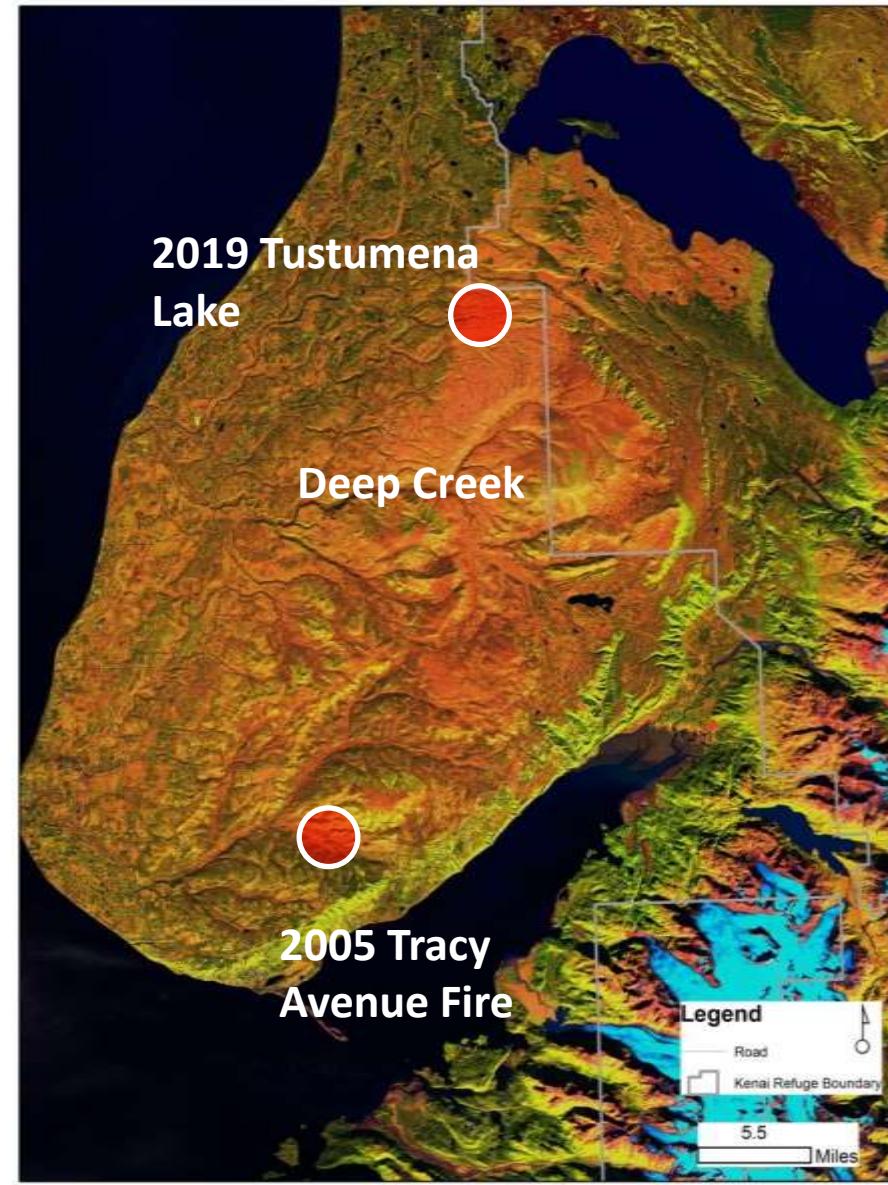
SEPT 1985



SEPT 2014



SEPT 1985



SEPT 2014

An aerial photograph showing a vast, hilly landscape. The terrain is covered in brown, sparsely vegetated areas, indicating deforestation. A winding blue river or creek bed cuts through the center of the image. In the background, a range of mountains is visible under a clear sky.

Deforestation North Fork of Deep Creek

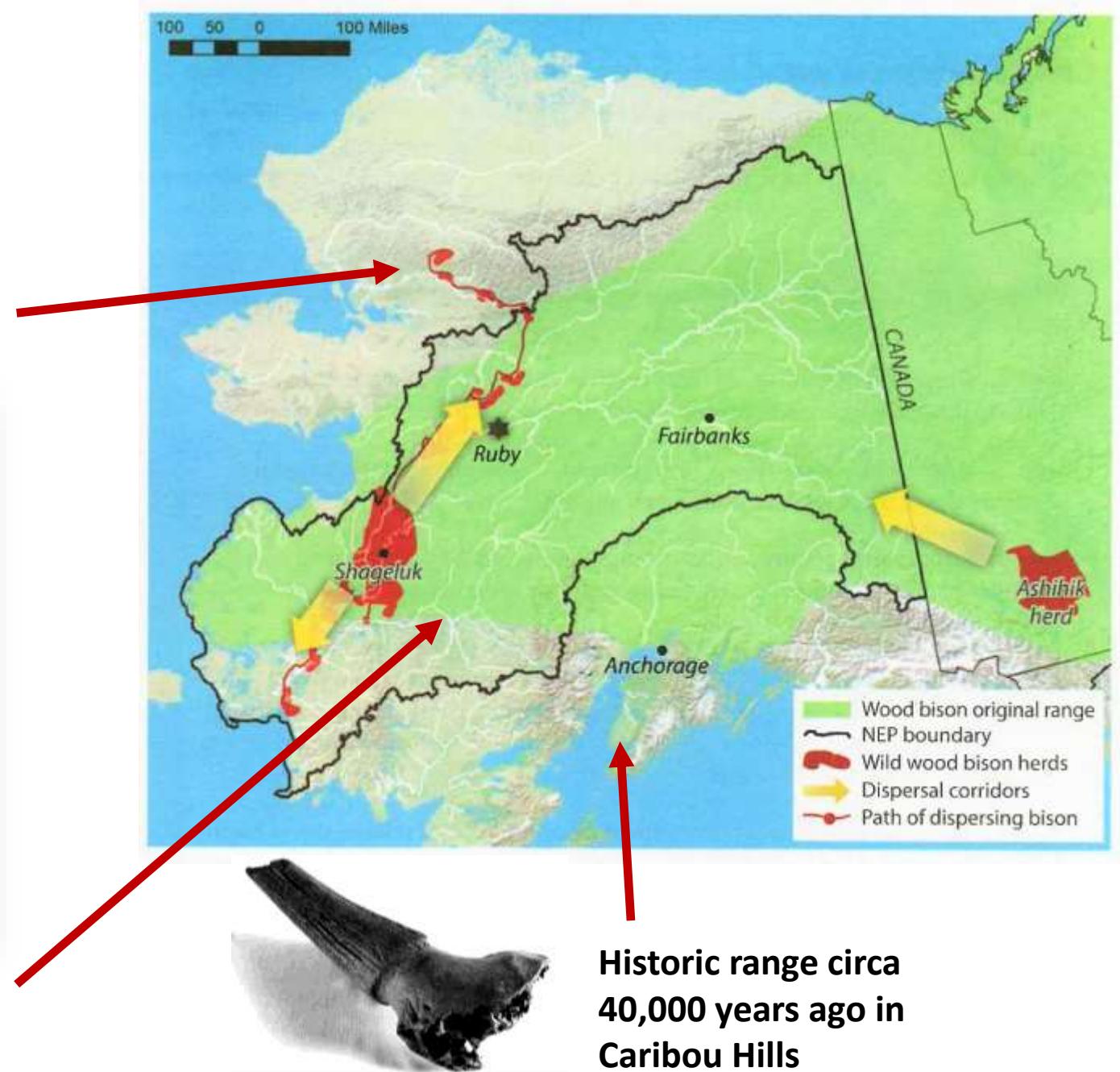


Bison? Grasslands need a functional grazer

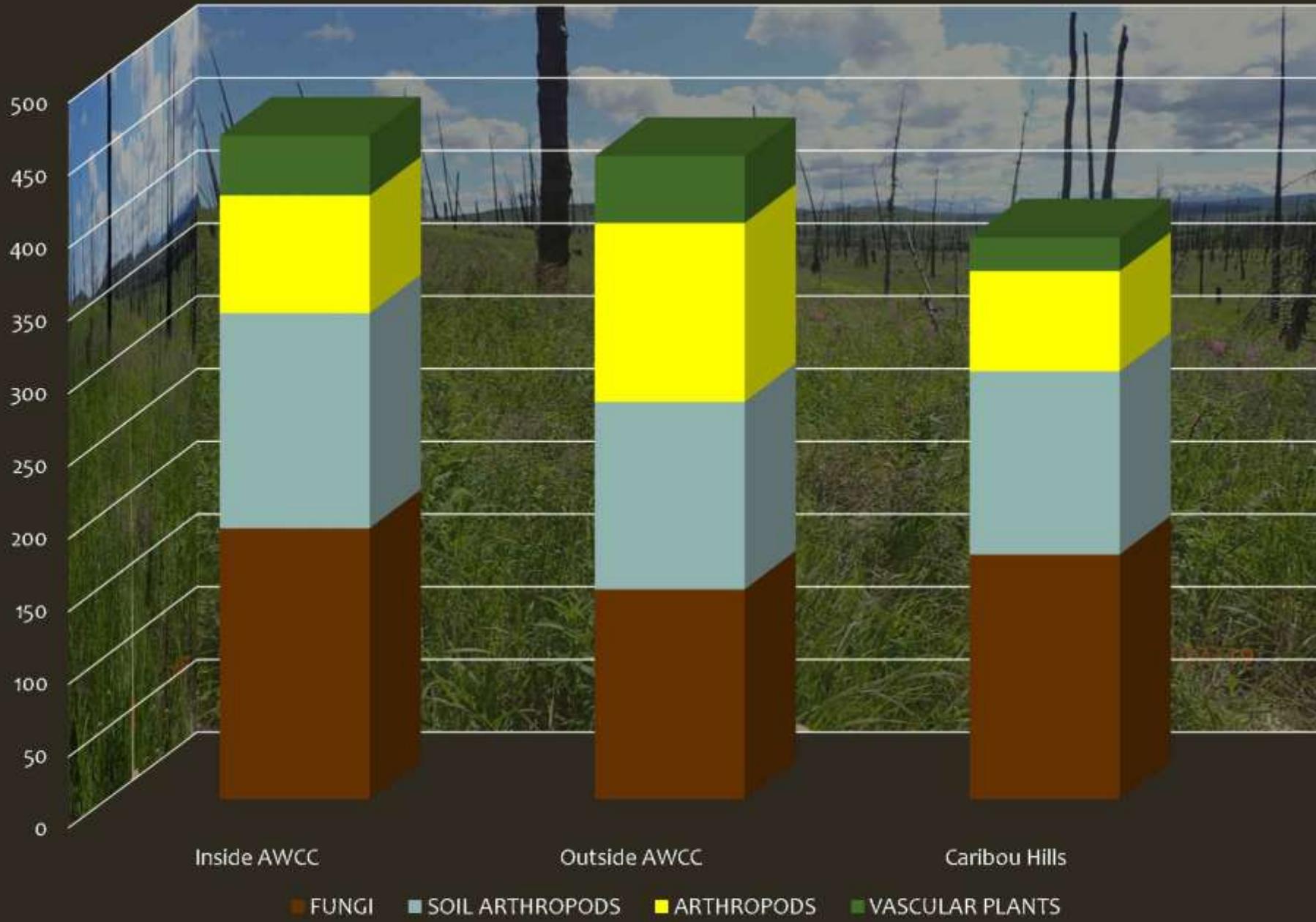


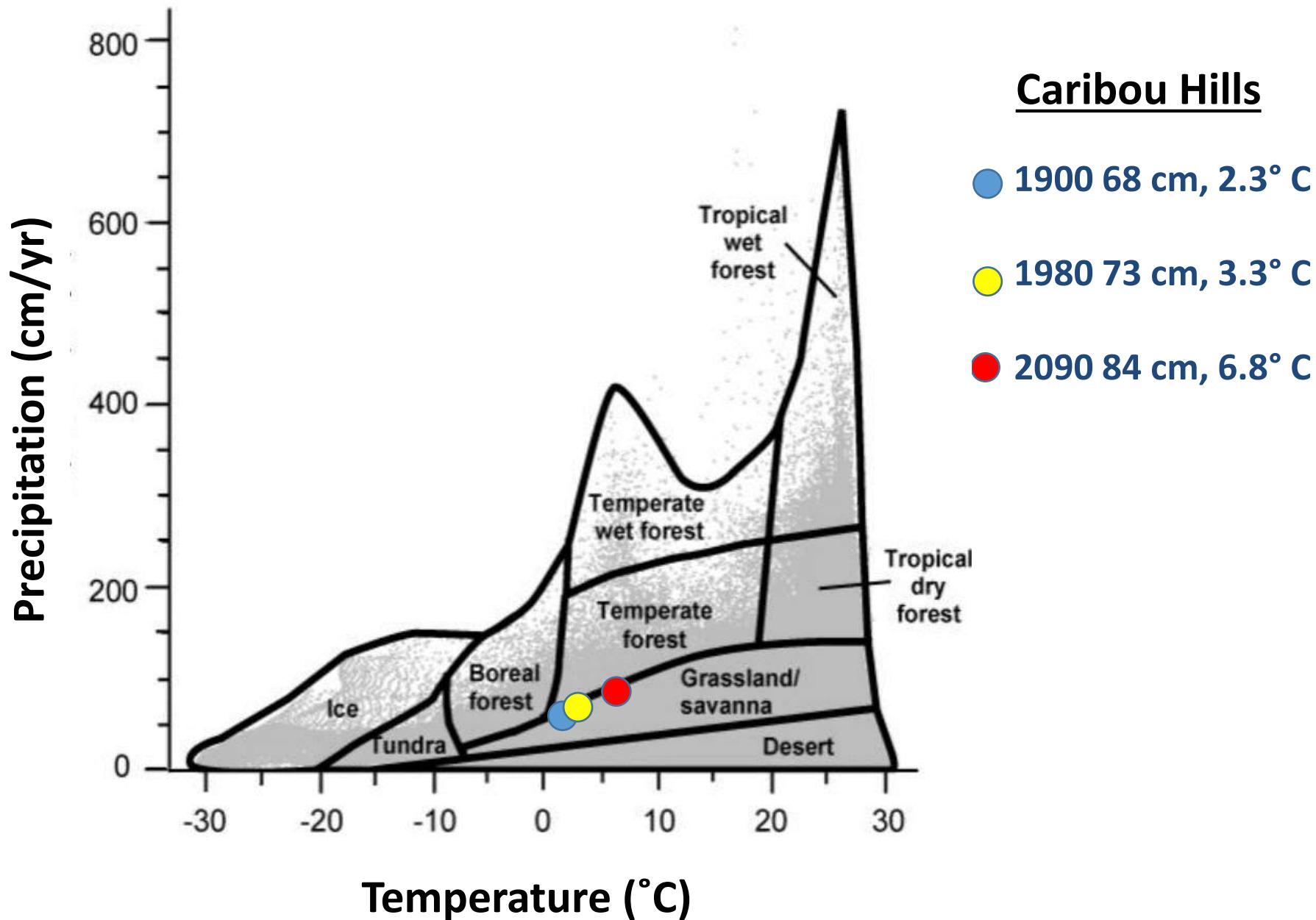
2018 outside
historic range

Historic range circa
19th century



SPECIES RICHNESS



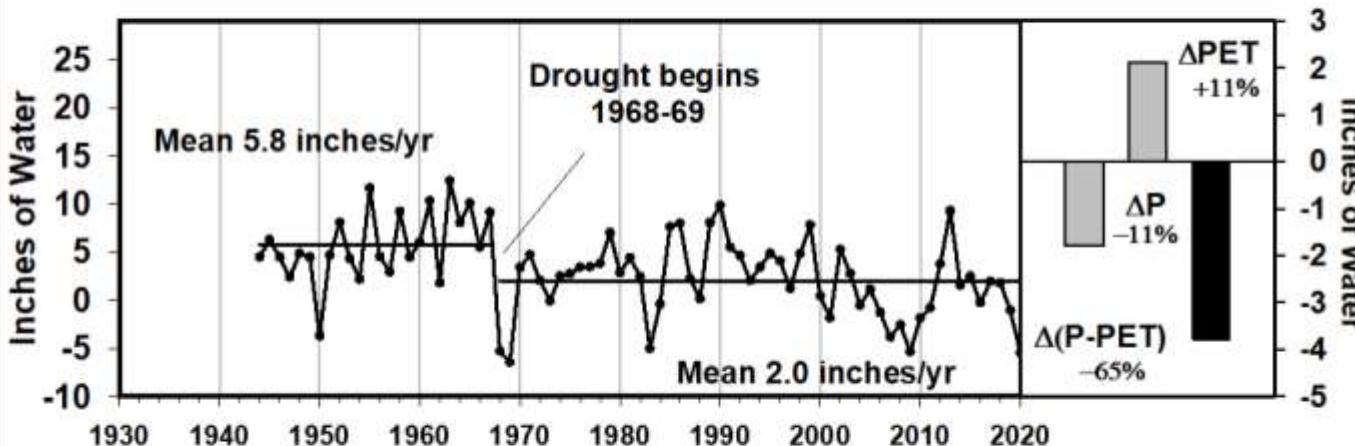


Staudinger et al. 2012. Impacts of Climate Change on Biodiversity, Ecosystems, and Ecosystem Services: Technical Input to the 2013 National Climate Assessment.

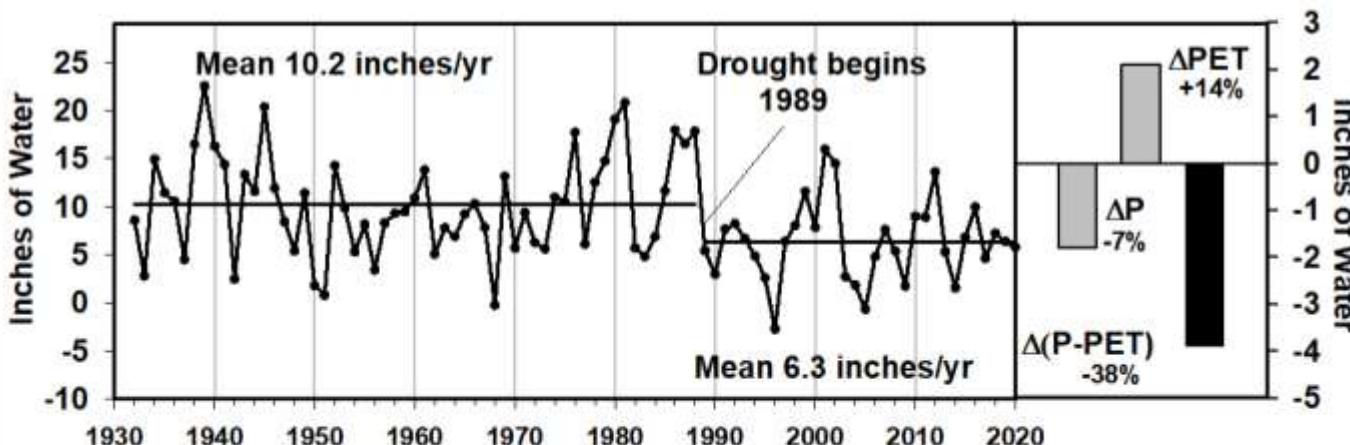
Available Water

Precipitation (P) - Potential Evapotranspiration (PET)

Kenai



Homer



Potential Evapotranspiration was calculated from mean monthly temperatures by the Thornthwaite-Mather method.

Ed Berg 5-2-2021

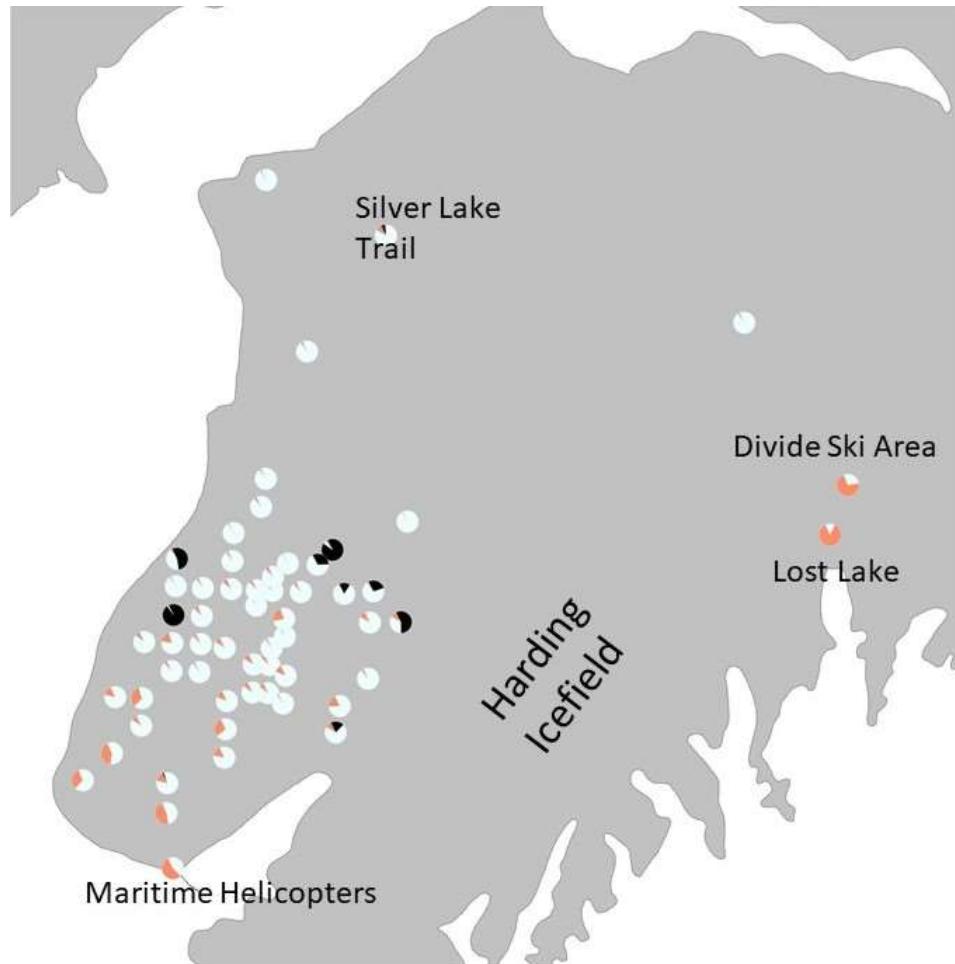
Kenai and Homer airports have declined 65% and 38%, respectively, in mean annual available water in last 5 decades

Hybrid spruce (Lutz) attributes

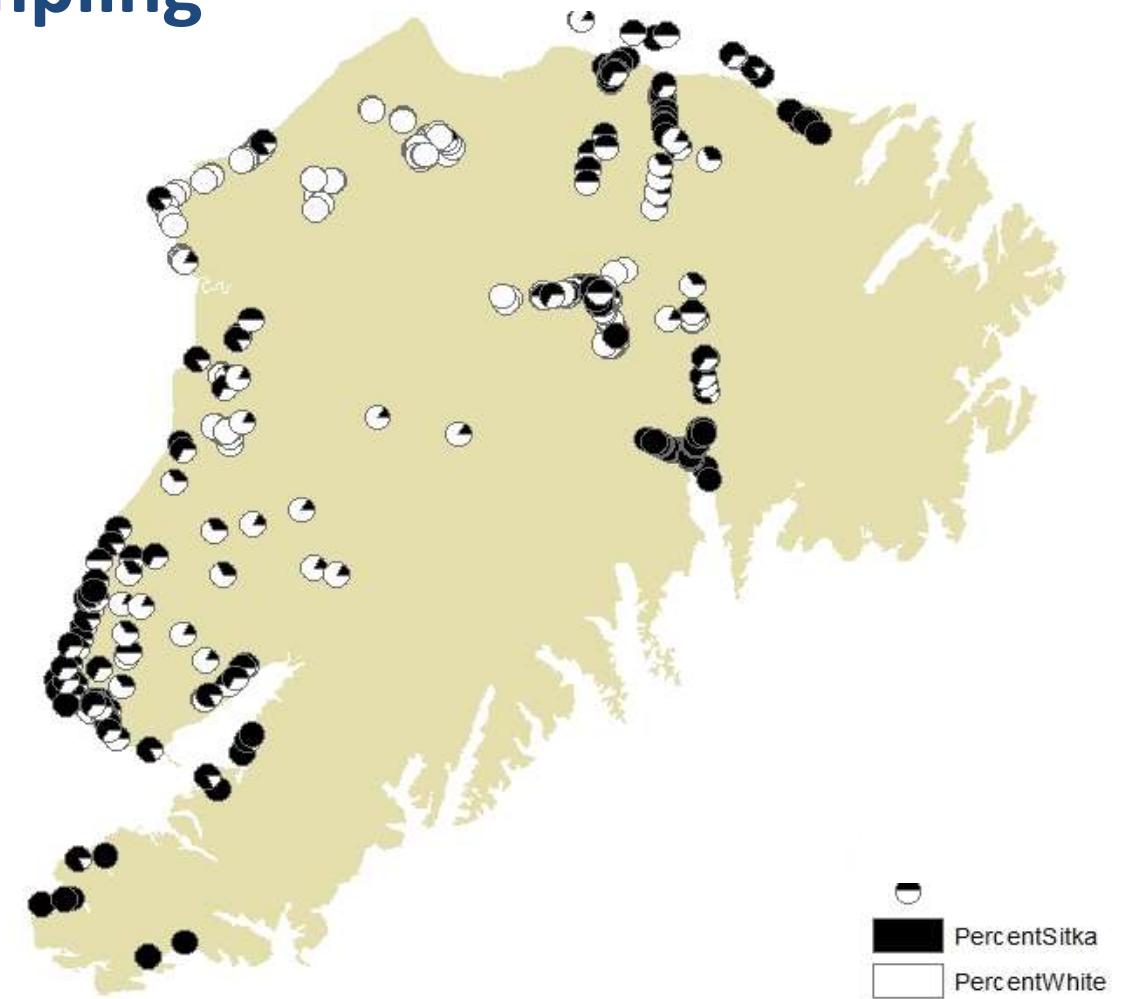
Bioclimatic variable	KP mean (SD)	White mean (SD)	Lutz mean (SD)	Sitka mean (SD)
BFFP = Julian date on which the frost-free period begins	144.1 (9.0)	149.1 (5.8)	149.0 (5.7)	141.5 (4.1)
EXT = extreme maximum temperature over 30 years (°C)	28.4 (1.6)	29.8 (0.9)	28.8 (0.7)	28.6 (0.6)
MAP = mean annual precipitation (mm)	1725.5 (1223.8)	564.6 (134.7)	782.7 (298.3)	1332.1 (641.6)
MAR = mean annual solar radiation (MJ m ⁻² d ⁻¹)	12.1 (0.8)	11.5 (0.3)	11.6 (0.6)	11.9(0.6)
MAT = mean annual temperature (°C)	1.9 (1.2)	2.2 (0.6)	1.8 (0.6)	2.5(0.6)
TD = difference between mean temperatures (°C) of coldest and warmest months (a measure of continentality)	20.0 (2.4)	21.3 (2.1)	20.6 (2.0)	19.7 (1.3)

- **Cooler MAT**
- **Intermediate MAP, TD**
- **Greater intolerance to spruce bark beetle attack**

Sitka, Lutz and White spruce sampling

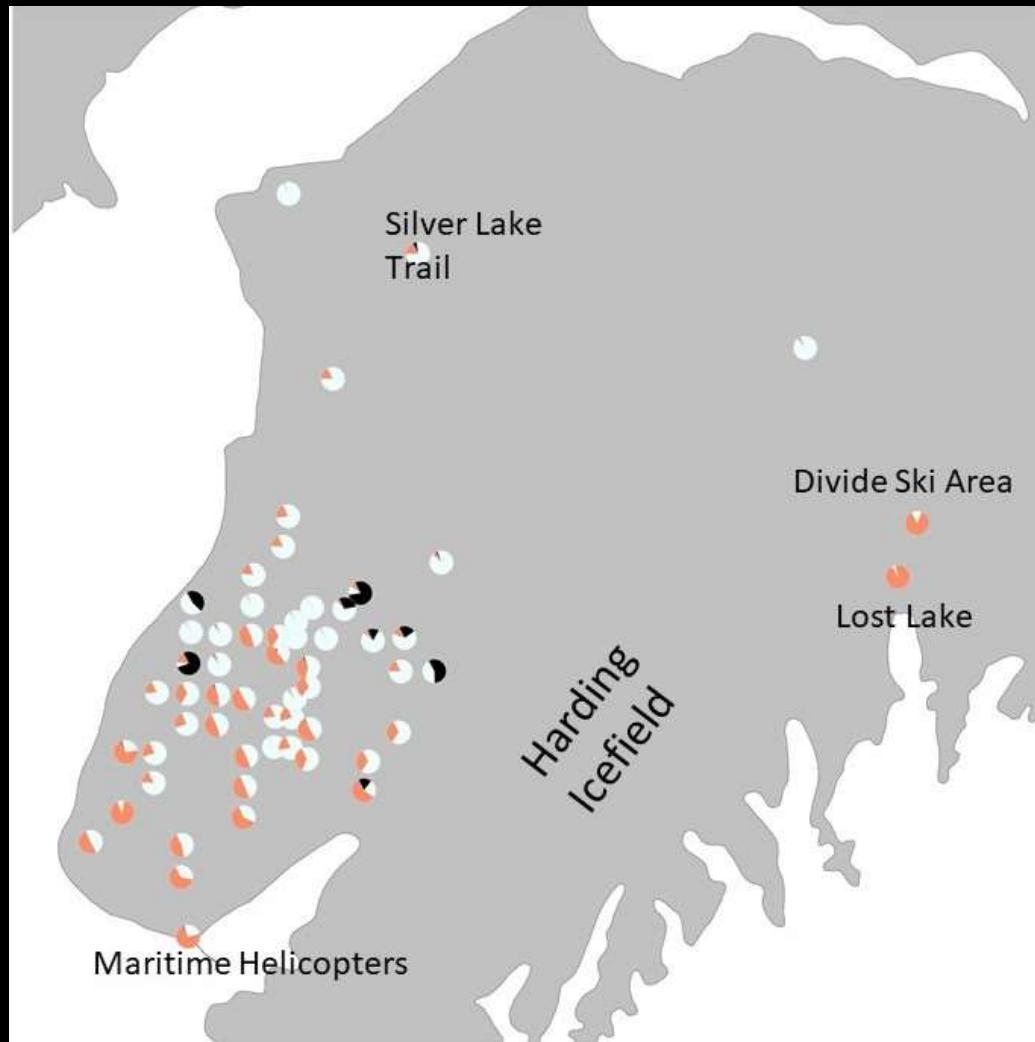


microsatellites (genotypic)

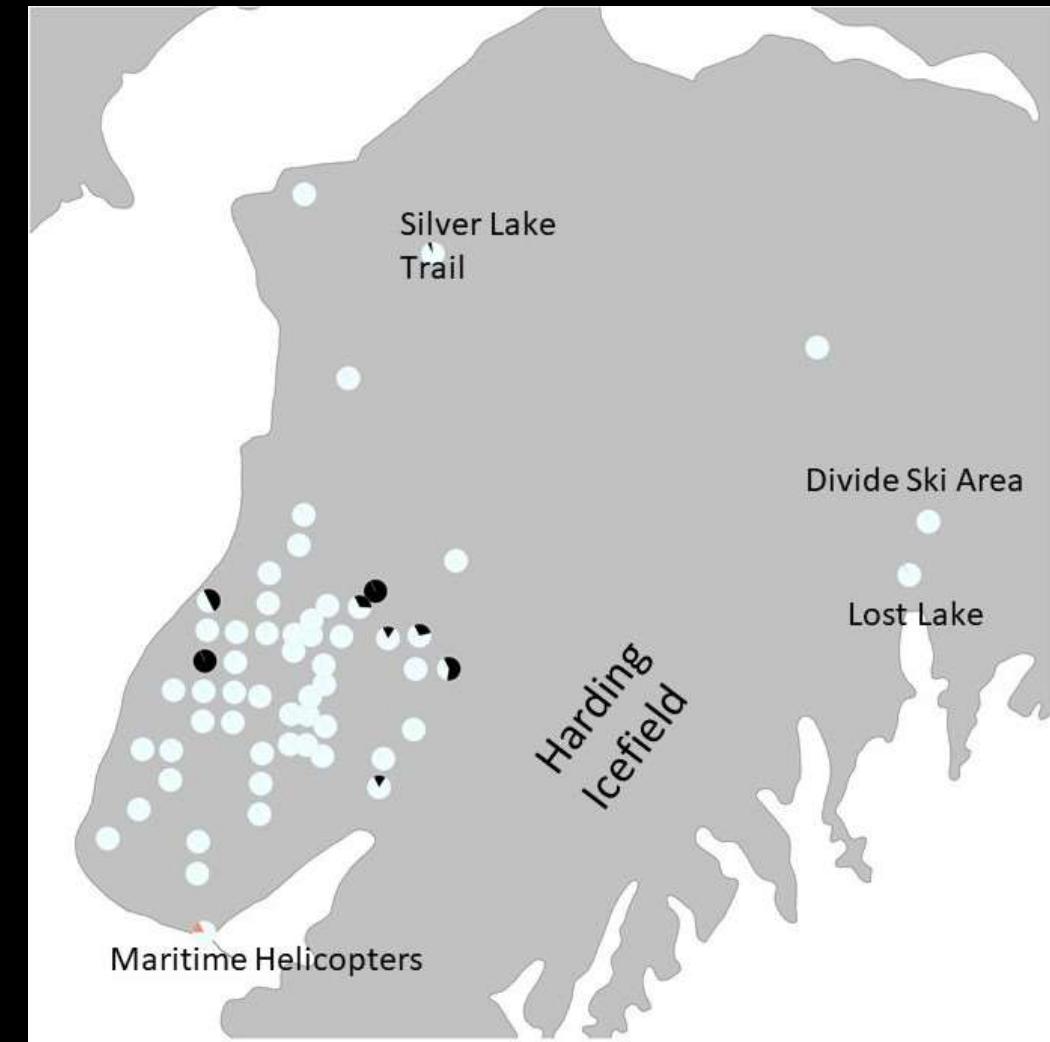


cones and foliage (phenotypic)

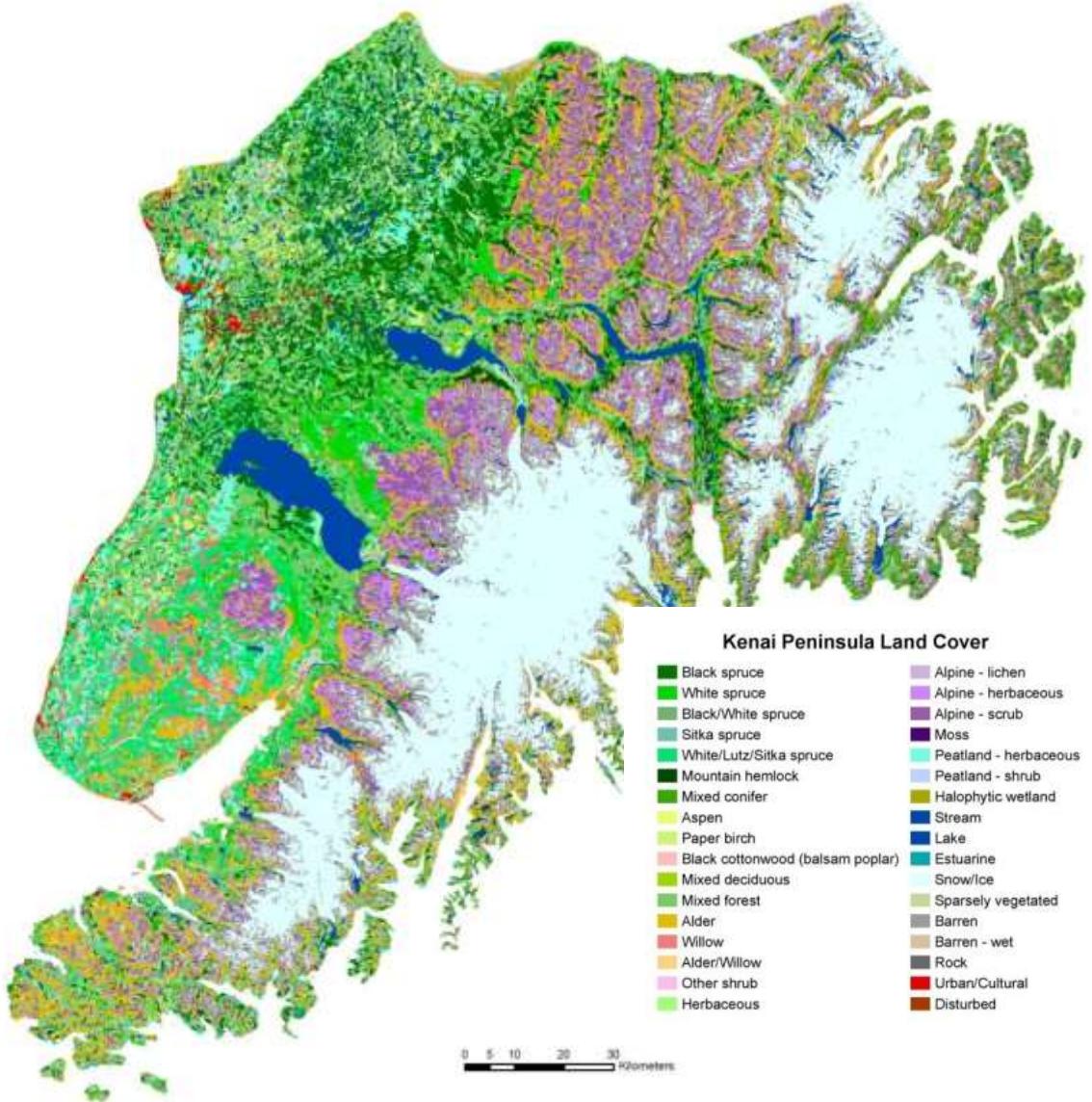
black = black spruce, white = white spruce, and orange = Sitka spruce



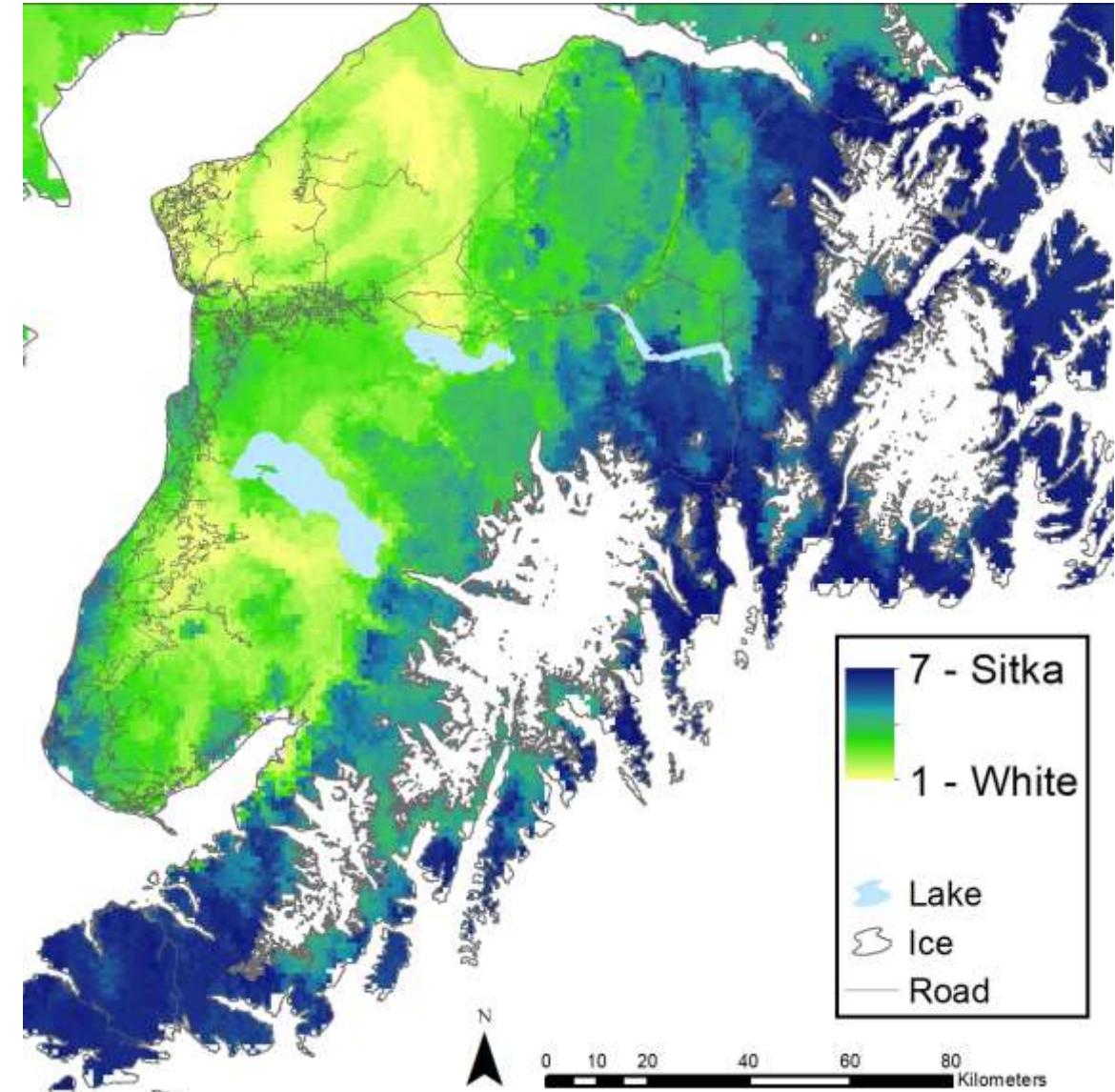
trnT-L (chloroplast♂)



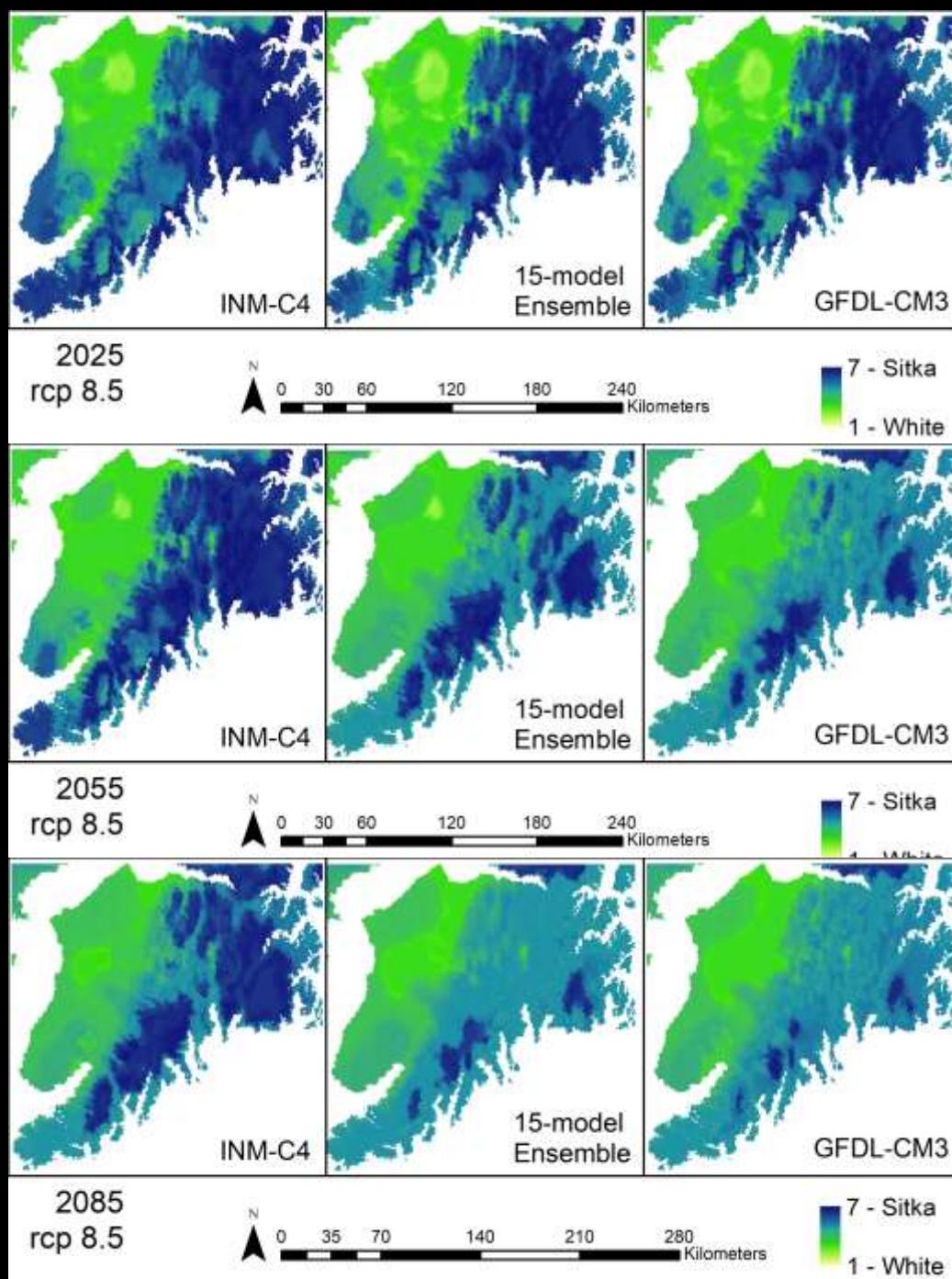
Nad5 (mitochondrial♀)



Landcover



Lutz spruce distribution



Future

White spruce likely to be extirpated and conditions for Sitka spruce decline

Kenai National Wildlife Refuge's Long Term Ecological Monitoring Program

- **Inventory** the co-occurrence and distribution of selected terrestrial floral & faunal species and assemblages

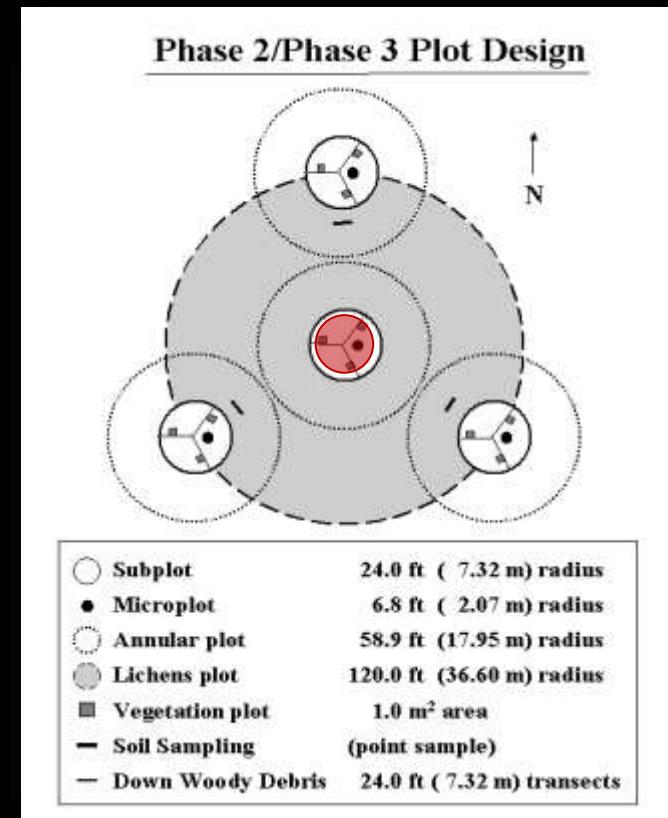
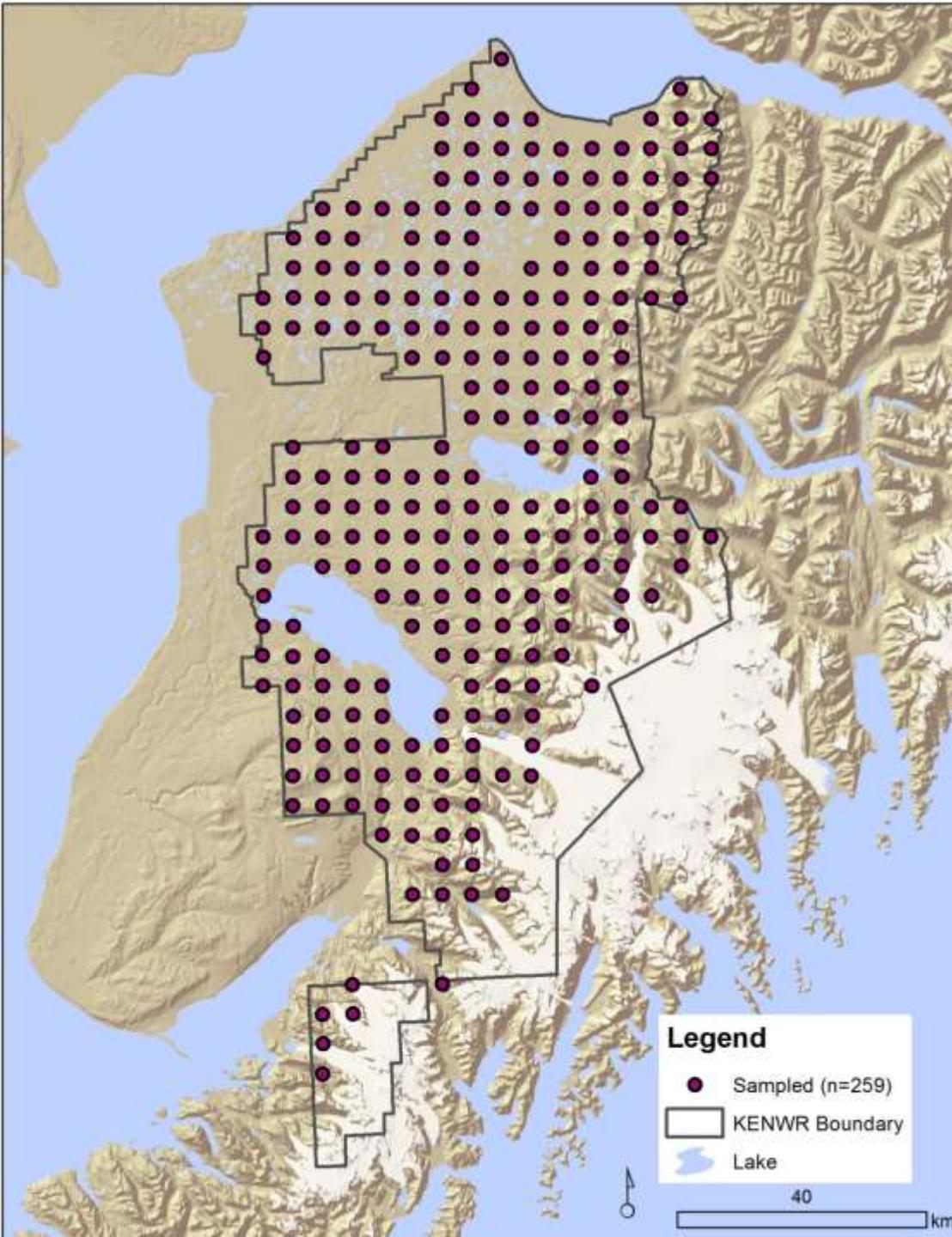
- **Monitor** trends in co-occurrence and distribution of selected terrestrial floral & faunal species and assemblages

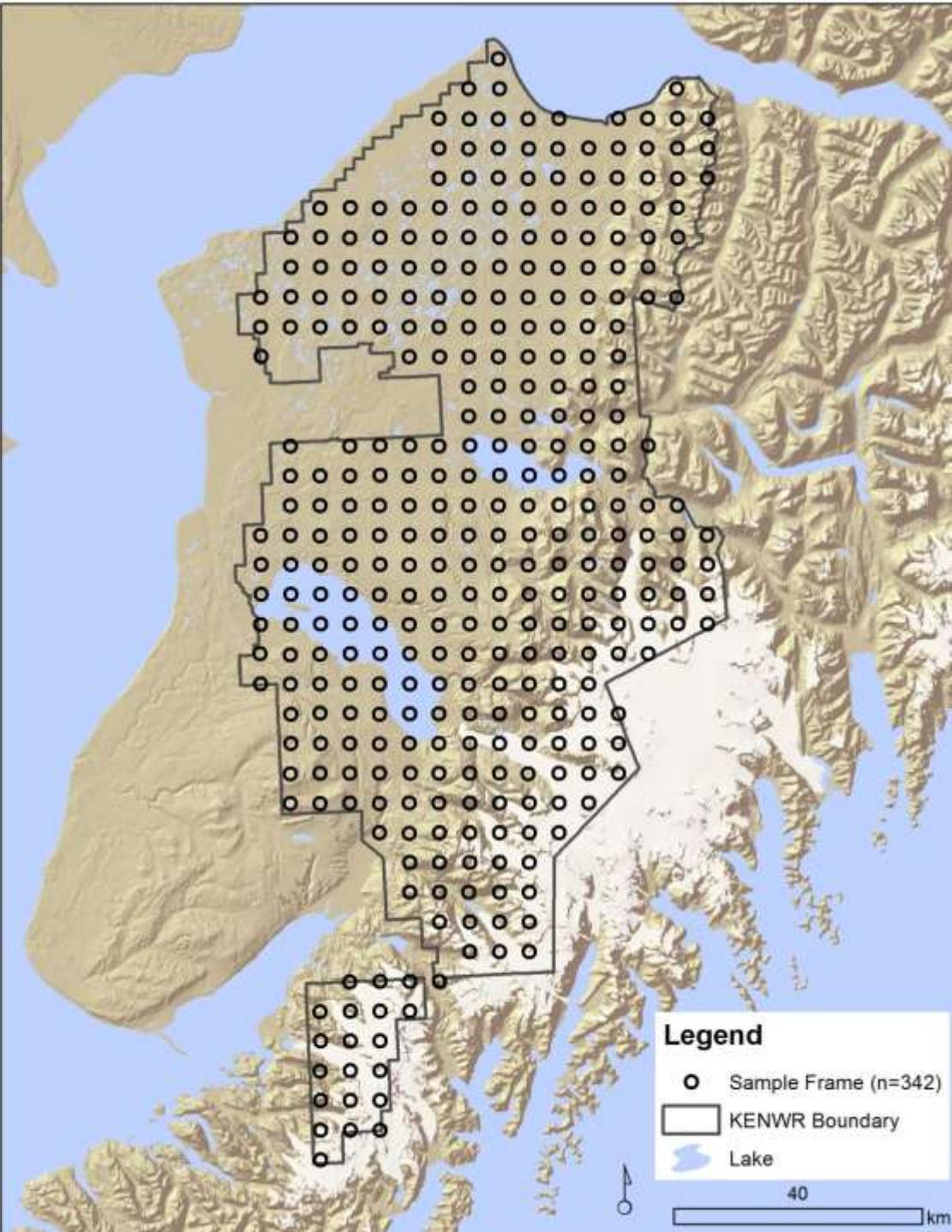
- **Model** the effects of physical, biological, and anthropogenic factors on occurrence and distributions



LTEMP

342 permanent plots systematically arrayed at 5-km intervals, of which 259 plots were sampled cooperatively with USDA-FIA





Although regionally scaled, data from LTEMP are representative of Kenai Refuge...

HABITAT	PLOTS (%)	ACRES (%)
Forest	161 (47)	945,896 (48)
conifer	105 (31)	550,996 (28)
deciduous	12 (4)	72,805 (4)
mixed	44 (13)	322,095 (16)
Shrub/grass	26 (7)	141,819 (7)
Barren/sparsely vegetated	59 (17)	329,293 (17)
Wetlands	20 (6)	122,292 (6)
Snow/ice	51 (15)	289,974 (15)
Water	25 (7)	159,242 (8)
Σ	342 (100)	1,988,516 (101)

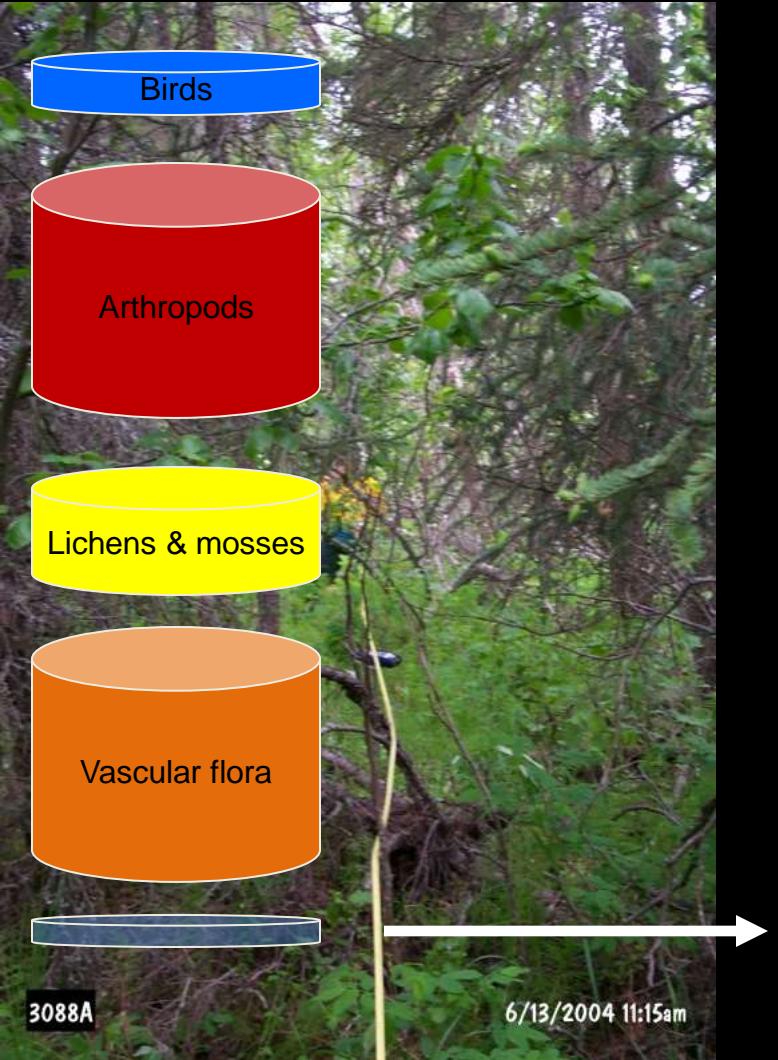
Data collected in 2004, 2006 & 2008

- Vascular (including exotics) & nonvascular flora on nonforested points (**modified line intercept**)
- Breeding bird densities (**VCP**)
- Insect relative abundance (**sweep nets**)
- Heavy metal uptake (**Hylocomium**)
- Rose galls as disturbance index
- Ambient noise recordings



32510

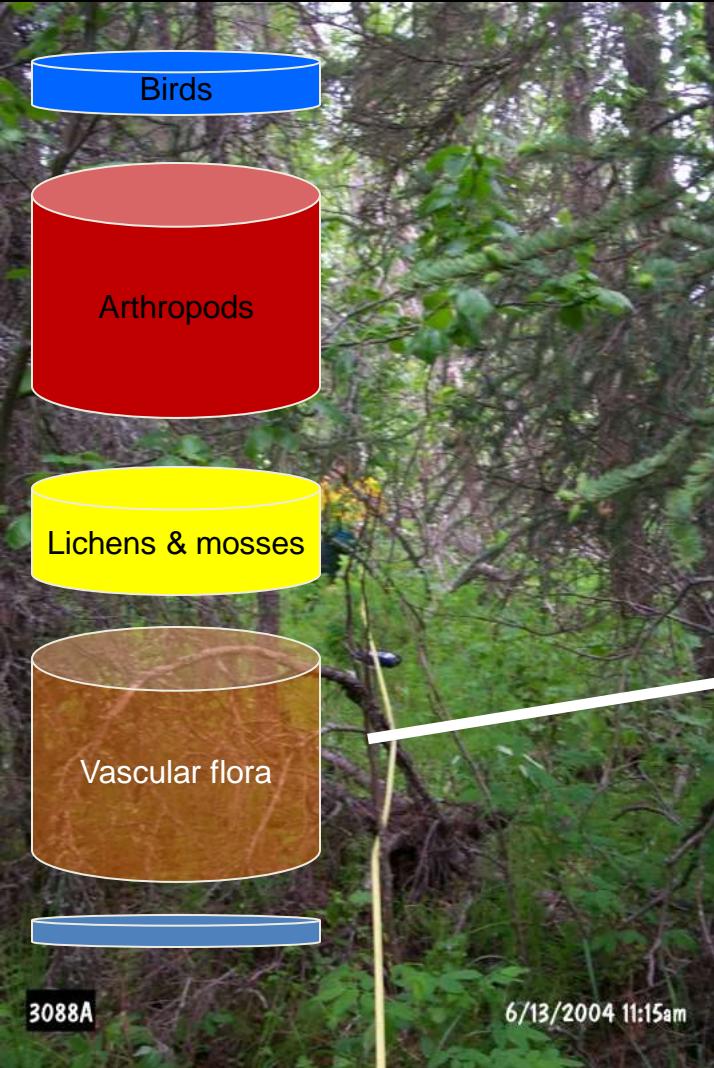
LTEMP site 3088



GIS derived data

black spruce (IA2F), 40 years old
elevation = 20 m
patch size = 57 ha
nearest stream = 105 m
nearest road = 2725 m
nearest border = 1119 m
Leq = 46.4 dBA
Rose galls = 0
mean annual temp = 3.2°C
mean annual precip = 433 mm

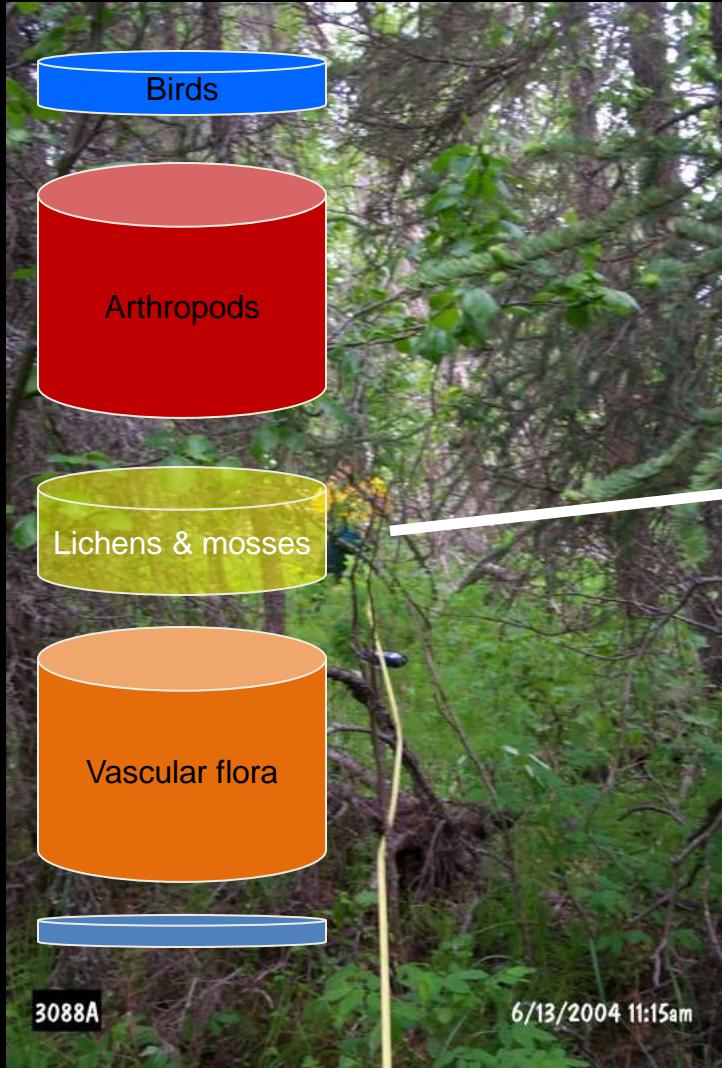
LTEMP site 3088



26 species of
vascular plants

Alanus incana
Athyrium filiz-femina
Betula nana
Calamagrostis canadensis
Chamerion angustifolium
Cornus canadensis
Dryopteris expansa
Empetrum nigrum
Equisetum sylvaticum
Equisetum arvense
Gymnocarpium dryopteris
Linnaea borealis
Mertensia paniculata
Picea mariana
Pyrola minor
Ribes glandulosum
Ribes hudsonianum
Ribes triste
Rosa acicularis
Rubus arcticus
Salix pulchra
Sanguisorba canadensis
Spiraea stevenii
Trientalis europaea
Vaccinium uliginosum
Vaccinium vitis-idaea

LTEMP site 3088



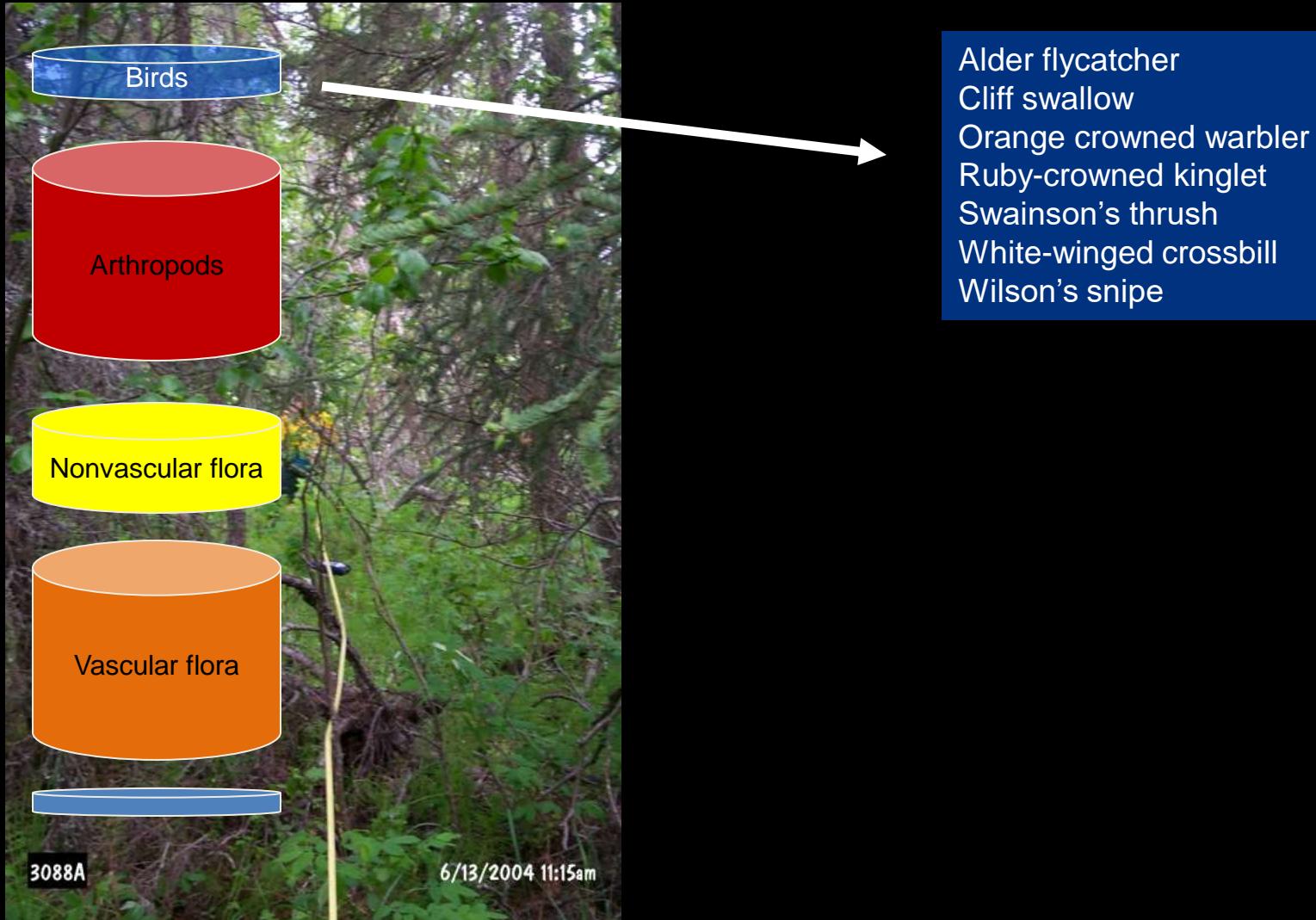
9 lichen & moss species

Ptilidium ciliare
Drepanocladus uncinatus
Stereocaulon alpinum
Aulacomnium palustre
Cladonia umbricola
Parmelia sulcata
Pleurozium schreberi
Rhizomnium nudum
Brachythecium sp.

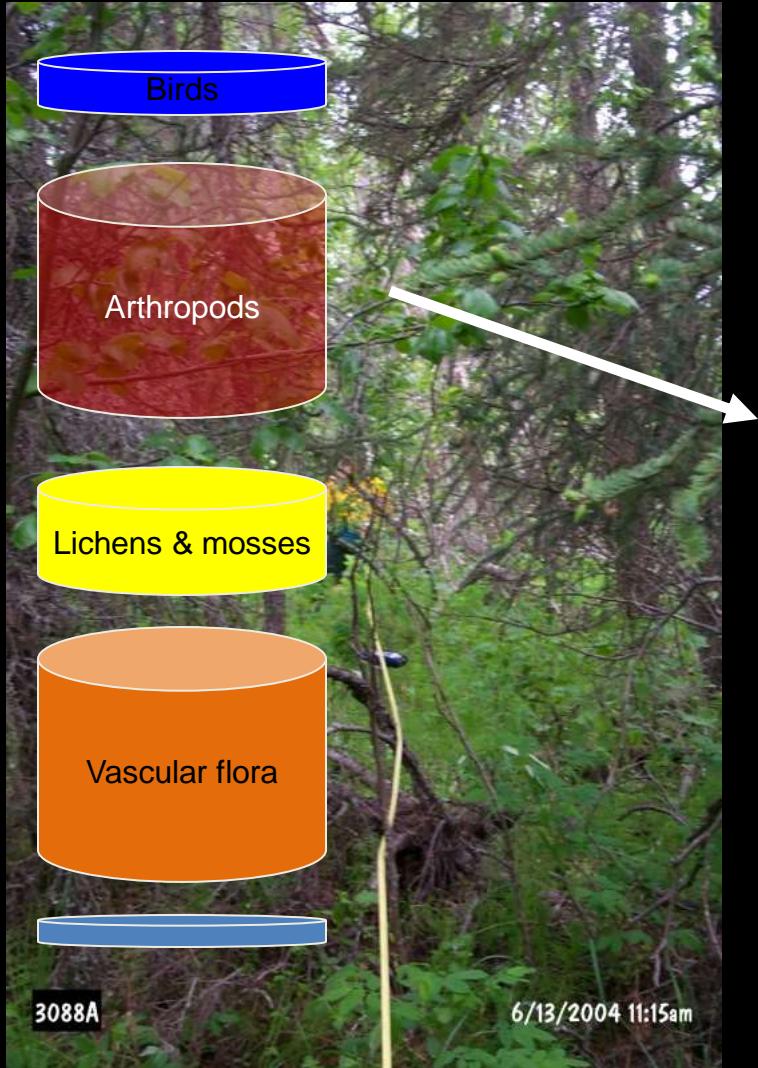
100 m²

LTEMP site 3088

7 bird species



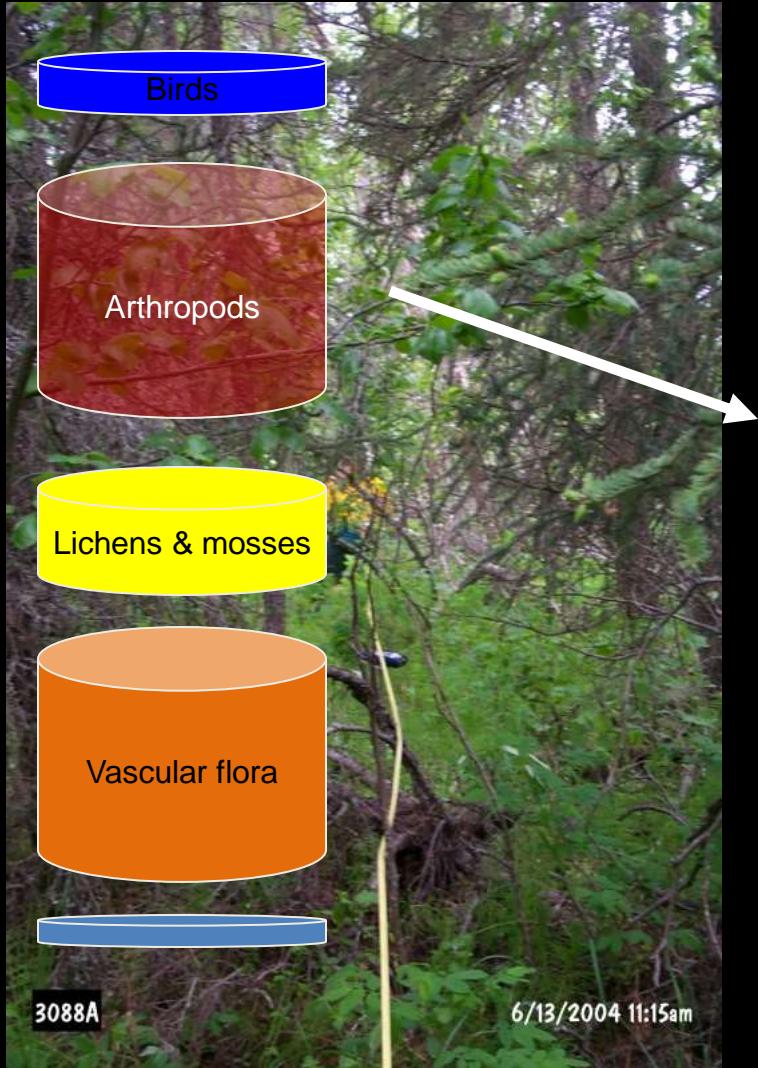
LTEMP site 3088



Aphididae
Aphidius
Araneae
Balclutha manitou
Braconidae
Craspedolepta
Culicidae
Diptera
Dismodicus modicus
Ephedrus lacertosus
Euthyneura nr. albipennis
Fannia postica
Fannia serena
Fannia spathiophora
Fannia subpellucens
Hemerobiidae
Ichneumonidae
Javesella pellucida
Lepidoptera
Melanostoma mellinum
Mycetophilidae
Phaenoglyphis kenaiii
Podabrinii
Sapromyza TAW1 BOLD:AAG6931
Simuliidae
Sminthurus

26 arthropod taxa

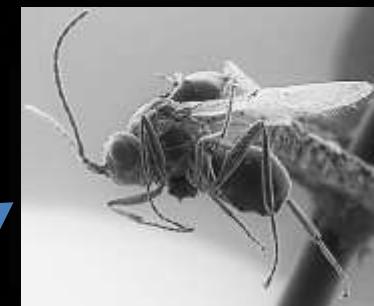
LTEMP site 3088



Birds
Arthropods
Lichens & mosses
Vascular flora

26 arthropod taxa

Aphididae
Aphidius
Araneae
Balclutha manitou
Braconidae
Craspedolepta
Culicidae
Diptera
Dismodicus modicus
Ephedrus lacertosus
Euthyneura nr. albipennis
Fannia postica
Fannia serena
Fannia spathiophora
Fannia subpellucens
Hemerobiidae
Ichneumonidae
Javesella pellucida
Lepidoptera
Melanostoma mellinum
Mycetophilidae
Phaenoglyphis kenaiii
Podabrinii
Sapromyza TAW1 BOLD:AAG6931
Simuliidae
Sminthurus

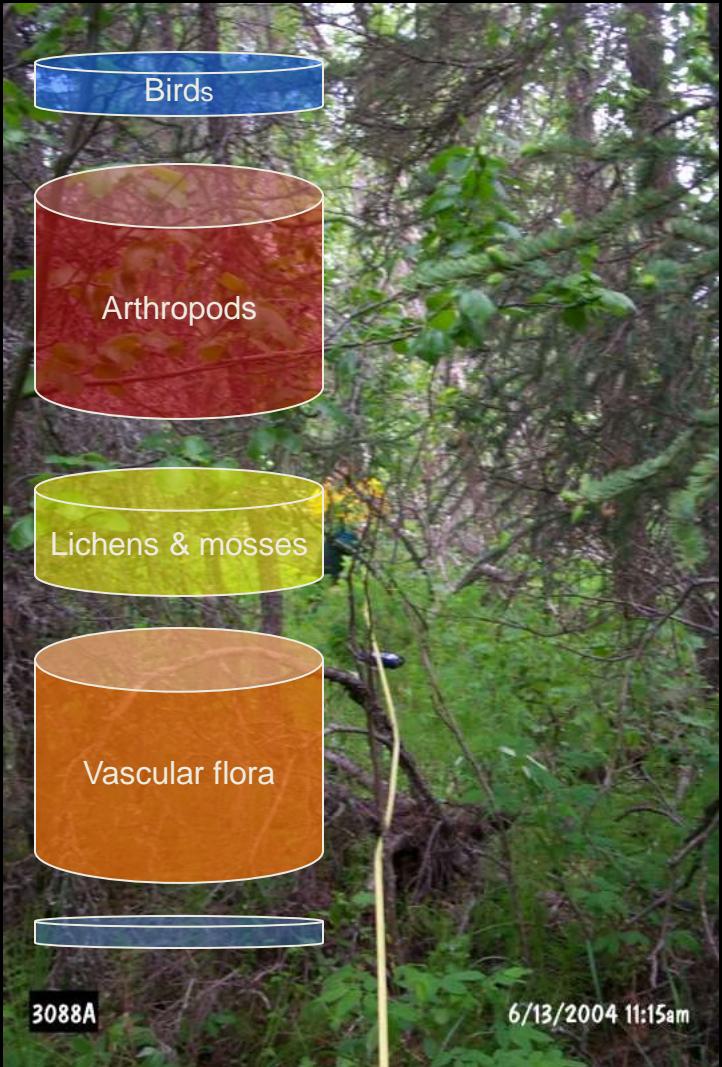


Kenai Figitid
wasp



saprophytic
fly

LTEMP site 3088



100 m²

Aphididae

Aphidius

Araneae

Balclutha manitou

Braconidae

Craspedolepta

Culicidae

Diptera

Dismodicus modicus

Ephedrus lacertosus

Euthyneura nr. albipennis

Fannia postica

Fannia serena

Fannia spathiophora

Fannia subpellucens

Hemerobiidae

Ichneumonidae

Javesella pellucida

Lepidoptera

Melanostoma mellinum

Mycetophilidae

Phaenoglyphis kenaii

Podabrini

Sapromyza TAW1 BOLD:AAG6931

Simuliidae

Sminthurus

Alder flycatcher

Cliff swallow

Orange crowned warbler

Ruby-crowned kinglet

Swainson's thrush

White-winged crossbill

Wilson's snipe

Alanus incana

Athyrium filix-femina

Betula nana

Calamagrostis canadensis

Chamerion angustifolium

Cornus canadensis

Dryopteris expansa

Empetrum nigrum

Equisetum sylvaticum

Equisetum arvense

Gymnocarpium dryopteris

Linnaea borealis

Mertensia paniculata

Picea mariana

Pyrola minor

Ribes glandulosum

Ribes hudsonianum

Ribes triste

Rosa acicularis

Rubus arcticus

Salix pulchra

Sanguisorba canadensis

Spiraea stevenii

Trientalis europaea

black spruce (IA2F), 40 years old

Vaccinium uliginosum

elevation = 20 m

Vaccinium vitis-idaea

patch size = 57 ha

nearest stream = 105 m

nearest road = 2725 m

nearest border = 1119 m

Leq = 46.4 dBA

Rose galls = 0

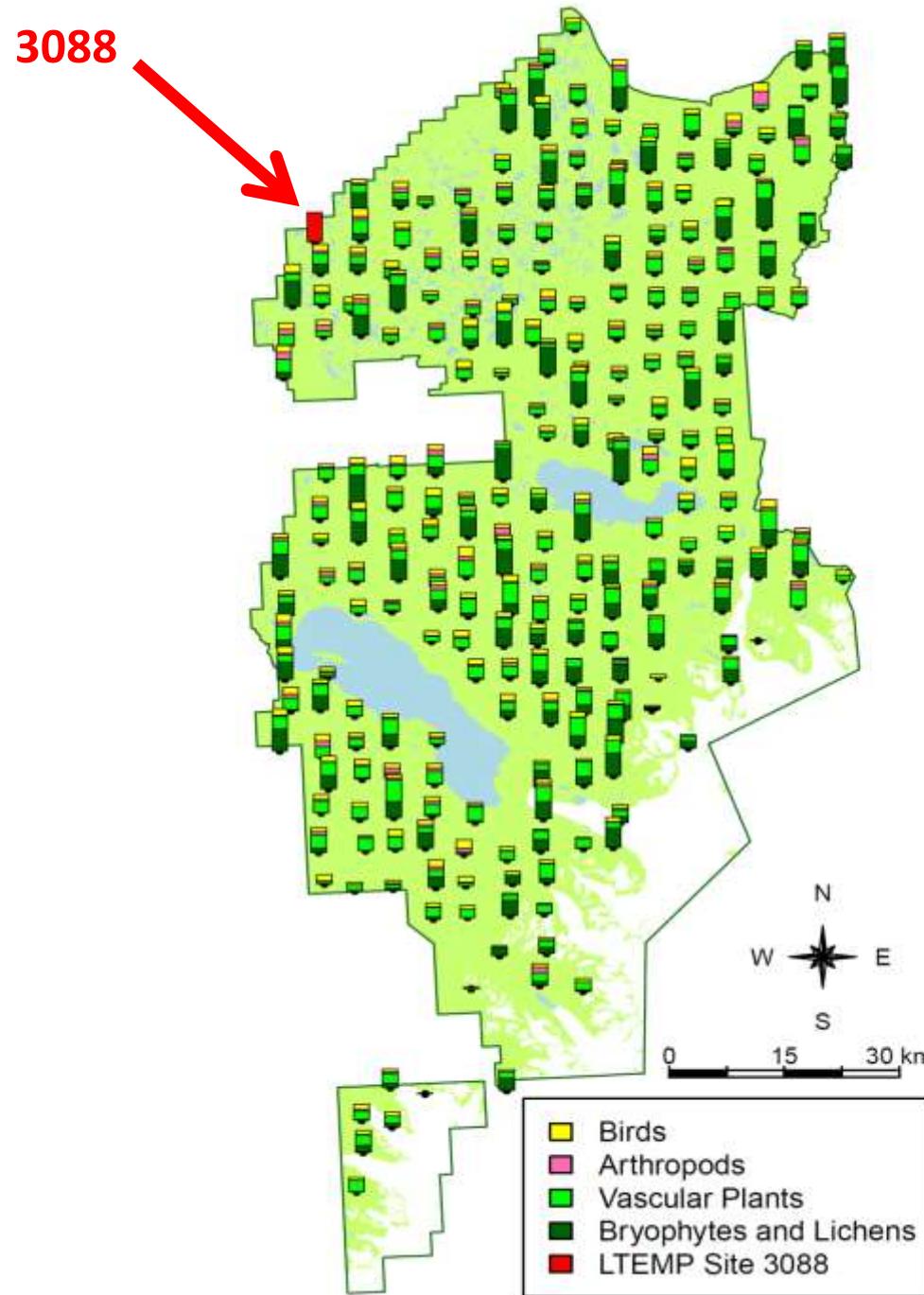
mean annual temp = 3.2°C

mean annual precip= 433 mm

**1,106 species
on 259 plots!**

- **80 birds**
- **256 invertebrates**
- **324 vascular plants**
- **297 lichens**
- **149 bryophytes**

1 – 105 species per 100m² plot with mean = 41



Taxonomic Impediment



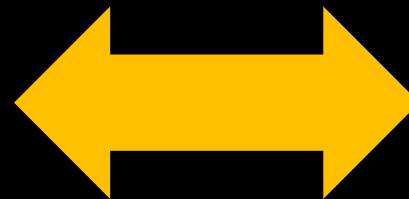
>15,000 specimens collected on
259 plots but only 256 species
identified over 8 years

Identification by morphology or
Sanger sequencing for cryptic
species is impractical for
monitoring



Bulk sampling (< \$100) using metagenomics or metabarcoding

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AATAATATATTATTTCGCTATGATCAGGAATAATTGGTTC  
ATCTATAAGATTATTAATTCGAATAGAATTAAGTCATCCTGGAAT  
ATGAATTAATAATGATCAGATTATAATTCTTAGTAACAGACA  
CGCATTAAATAATTAGTTATAGTTACCAATTATAATTGGA  
GGATTGGAAATTATTAATTCCATTAATATTAGGATGCCAGA  
TATAGCTTCTCGAATAATAATATTAGATTGACTTTACC  
CCCATCATTATTATACTCTATTAAAGAAATATTTACACCTAAT  
GTAGGAACAGGATGAACGTATATCCTCCTTATCCTCTTATT  
ATTCATTCTCATCTCCATCAATTGATATTGCAATCTTCTTACAT  
ATGTCAGGAATTCTTCTATTATTGGATCATTAAATTATTGTT  
ACTATTAAATAATAAAAAATCTTCATTAAATTATGACCAAATT  
AATTATTCTCATGATCAGTATGTATTACTGTAATTATTAAATT  
TATCTTACCGGTTTAGCCGGAGCTATTACTATATTACTATTG  
ATCGAAATTAAACTTCATTCTTGATCCTATGGGAGGAGG  
GGATCCAATTATACCAACATTATT
```



Western bumble bee
(*Bombus occidentalis*)

Building a DNA barcode library of Alaska's non-marine arthropods

Derek S Sikes, Matthew Bowser, John M. Morton, Casey Bickford, Sarah Meierotto, Kyndall Hildebrandt

Published on the web 21 October 2016.

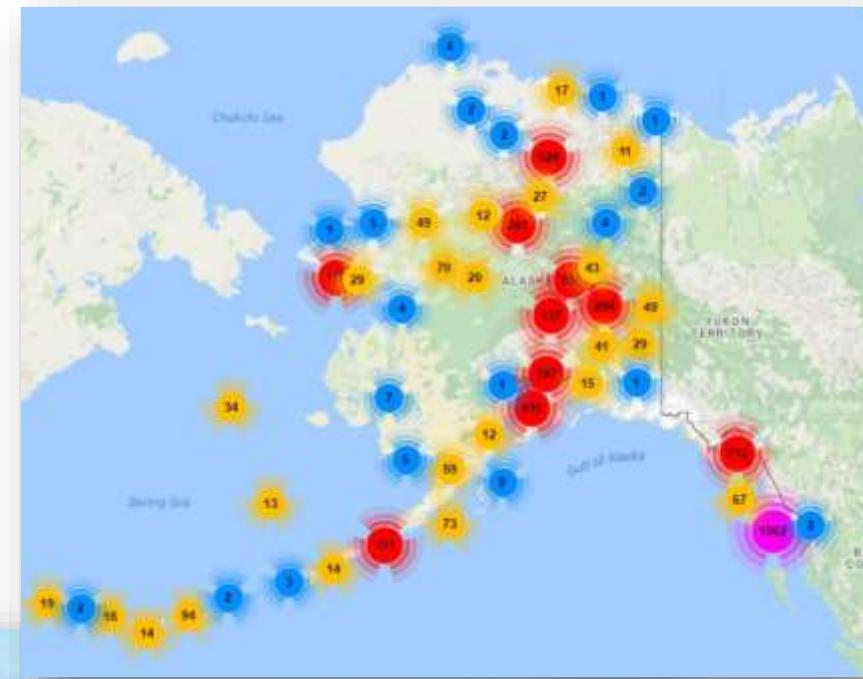
Received December 01, 2015.

Genome, 10.1139/gen-2015-0203

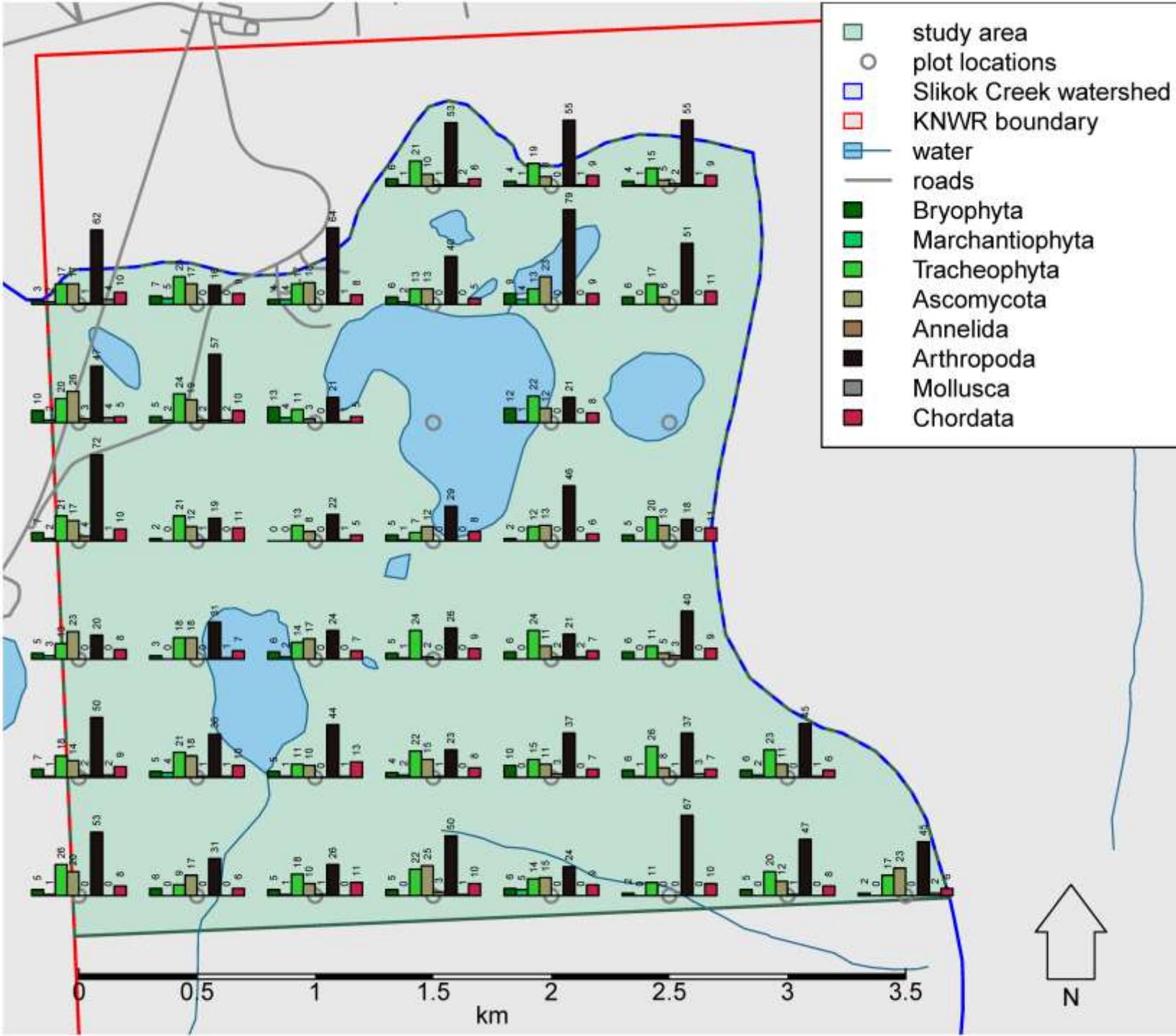
 [PDF \(1095 K\)](#)
 [PDF-Plus \(600 K\)](#)

ABSTRACT

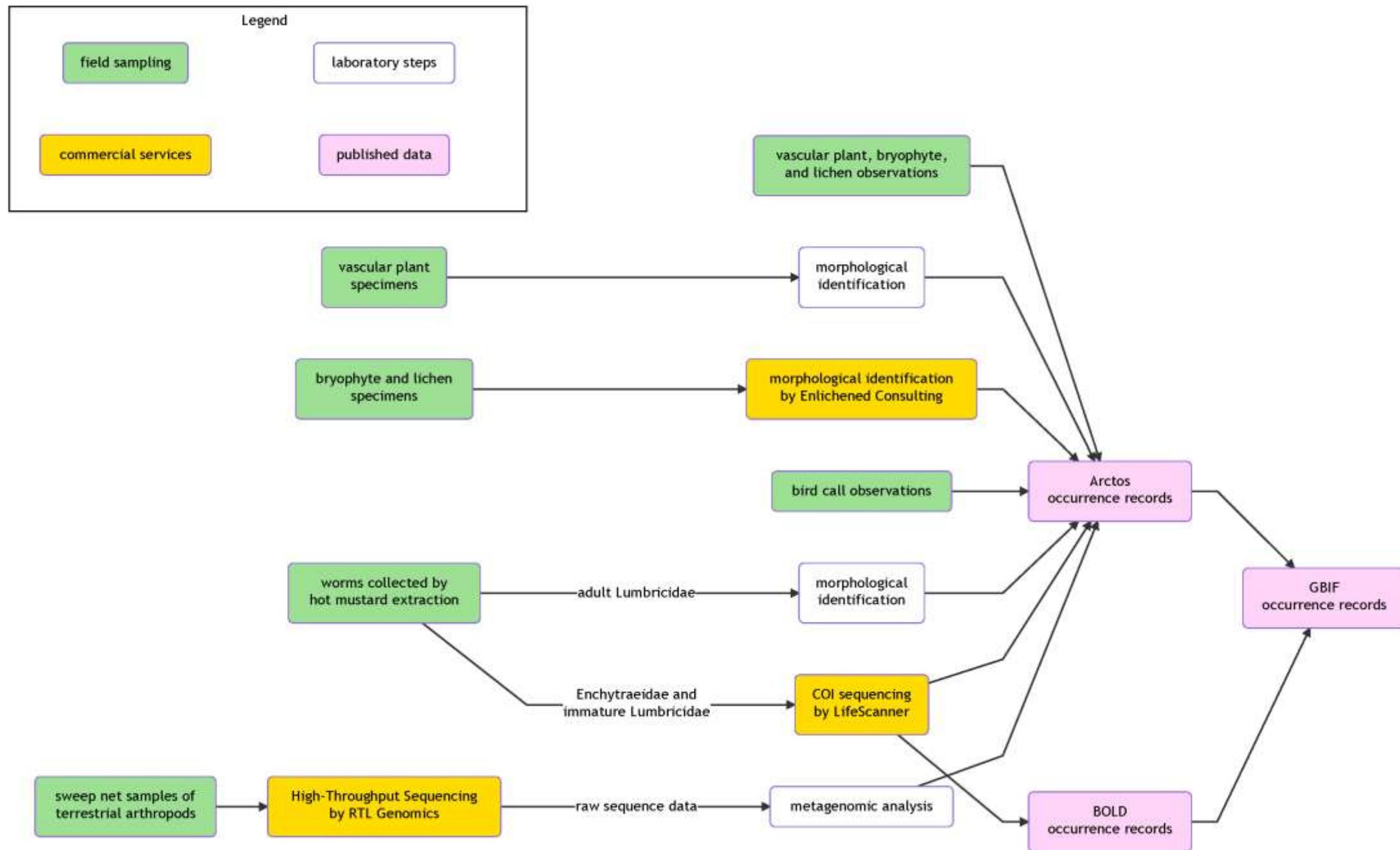
Climate change may result in ecological futures with novel species assemblages, trophic mismatch, and mass extinction. Alaska has a limited taxonomic workforce to address these changes. We are building a DNA barcode library to facilitate a metabarcoding approach to monitoring non-marine arthropods. Working with the Canadian Centre for DNA Barcoding, we obtained DNA barcodes from recently collected and authoritatively identified specimens in the University of Alaska Museum Insect Collection and the Kenai National Wildlife Refuge collection. We submitted tissues from 4,776 specimens, of which 81% yielded DNA barcodes representing 1,662 species and 1,788 BINs, of primarily terrestrial, large-bodied arthropods. This represents 84% of the species available for DNA barcoding in the UAM Insect Collection. There are now 4,020 Alaskan arthropod species represented by DNA barcodes, after including all records in BOLD of species that occur in Alaska – i.e. 48.5% of the 8,277 Alaskan, non-marine-arthropod, named species have associated DNA barcodes. An assessment of the identification power of the library in its current state yielded fewer species-level identifications than expected, but the results were not discouraging. We believe we are the first to deliberately begin development of a DNA barcode library of the entire arthropod fauna for a North American state or province. Although far from complete, this library will become increasingly valuable as more species are added and costs to obtain DNA sequences fall.

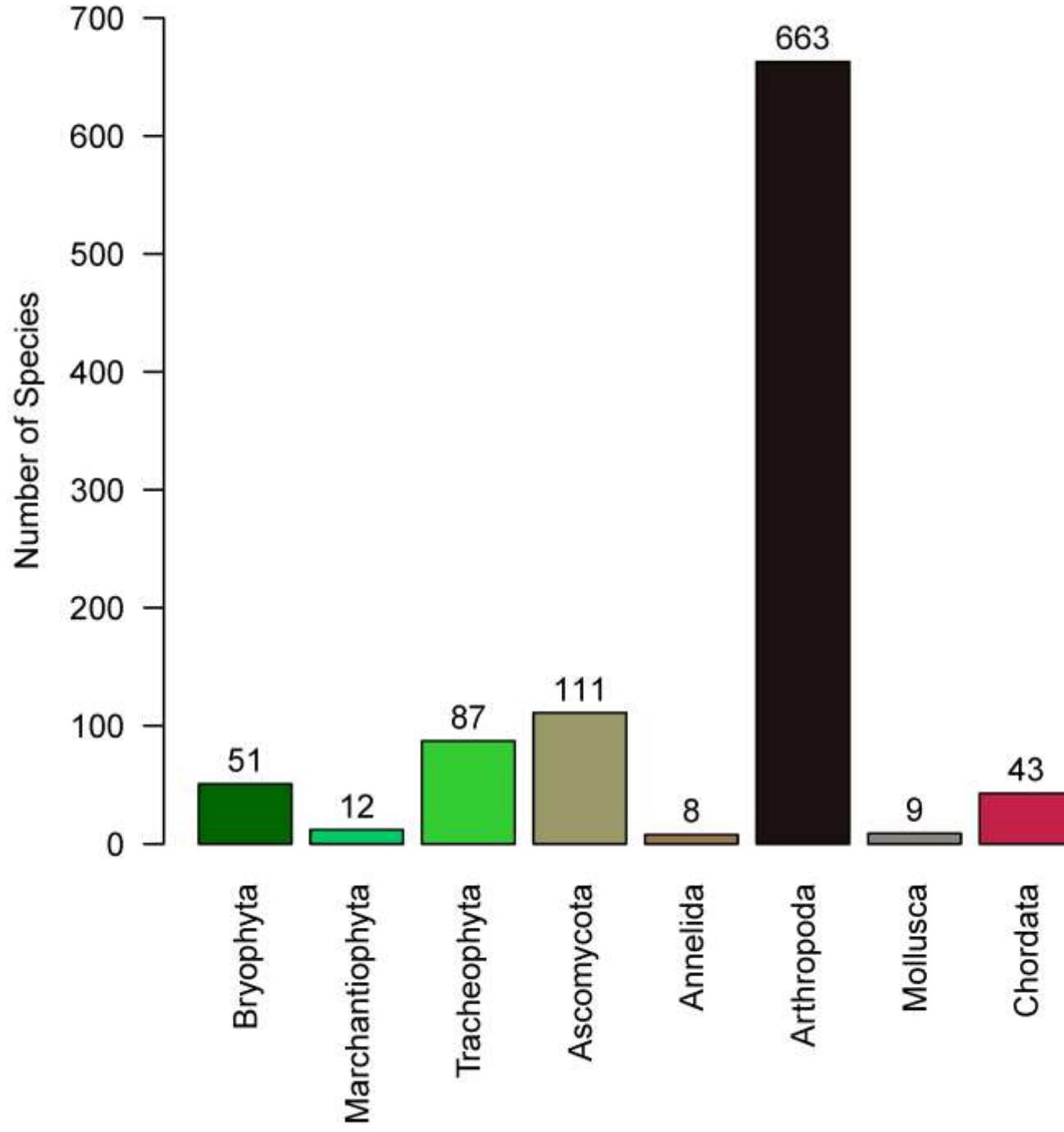


50% of 8,421 terrestrial arthropod species known from Alaska now barcoded!



40 sites 500 meters apart on 2300 acres





4,764 occurrences of 984 species and molecular operational taxonomic units:

- 87 vascular plants
- 51 mosses
- 12 liverworts
- 111 lichens
- 43 vertebrates
- 663 arthropods
- 9 molluscs
- 8 annelid worm

102 arthropod species new records for Alaska!

Inventorying species richness

The current list is 2251 species, including
2140 native, 116 non-native, 1 vagrant, and
3 of uncertain origin



Questions?