

# **DNA Barcoding of the Aphidoidea (Insecta: Hemiptera)**

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Agriculture and  
Agri-Food Canada

Agriculture et  
Agroalimentaire Canada

Canada

# Aphidoidea

- About 4800 described species in world
- About 1400 species in North America north of Mexico
- Feed on plant fluids (most are phloem feeders)
- Many important pests in agriculture, forestry and horticulture
  - direct feeding effects
  - virus transmission

# Aphidoidea: obstacles to taxonomy

## Morphological uniformity among species

- Simplification of structures; neoteny.
- Closely related species differ biologically but difficult to distinguish morphologically.
- Phylogenetic relationships obscured by convergent and parallel evolution of characters.

# Aphidoidea: obstacles to taxonomy

## Variation within species

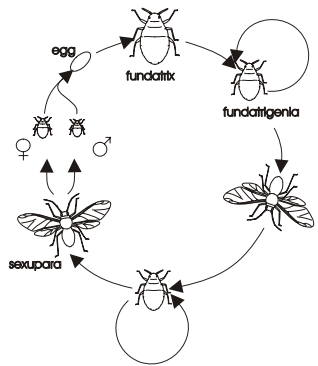
- Host plant (nutritional, physical) and temperature-induced variation
- Complex lifecycles with cyclic parthenogenesis and host alternation resulting in structural polymorphism.

# Aphidoidea: obstacles to taxonomy

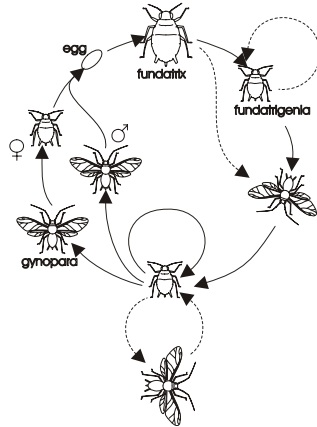
## Principal life cycle types

### Host alternating

#### Pemphigine heterocycle

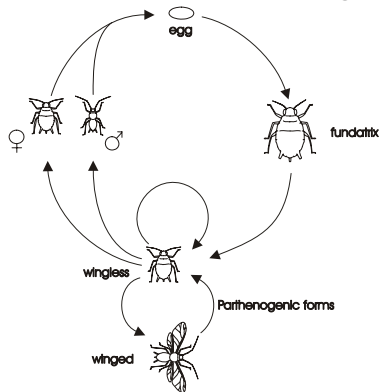


#### Aphidine heterocycle

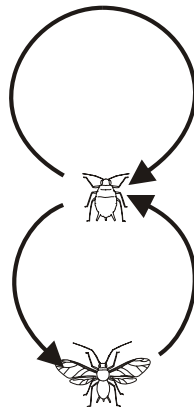


Each species may have many morphologically distinct forms

### Non-host alternating



### Permanently asexual



# Uses of barcoding in Aphidoidea

- Delineation of species boundaries
- Detection of cryptic species
- Detection of host-specific lineages
- Association of different morphological forms of same species
- Species identification/confirmation; monitoring for invasive species

# Current status of project

## barcoded taxa

	genera	species
<b>Adelgidae</b>	2	16
<b>Phylloxeridae</b>	3	3
<b>Aphididae</b> Anoeciinae	1	2
Aphidinae	66	260
Calaphidinae	17	25
Chaitophorinae	3	8
Drepanosiphinae	2	4
Eriosomatinae	18	42
Greenideinae	2	3
Hormaphidinae	9	13
Lachninae	9	19
Lizerinae	1	1
Mindarinae	1	2
Phloeomyzinae	1	1
Phyllaphidinae	2	3
Pterocommatinae	2	4
Saltusaphidinae	3	3
Tamaliinae	1	4
Thelaxinae	1	1

Samples submitted or prepared for  
submission: 1940

Sequences (with >600 bp) to date:

**1168**

representing

**414 species in 144 genera**

# Species discrimination

- **Species** mostly **well differentiated** by barcode sequence: 90% of between species sequence divergence  $> 3\%$
- Nominal species mostly well delineated: **intraspecific divergence low** ( $< 0.4\%$ ) in 90% of replicated species.
- A few very close species pairs with divergence  $< 1\%$ , but nominal species still form distinct clusters
- Six nominal species with intraspecific divergence  $> 1\%$  (3 of these  $> 3\%$ ): potential cryptic species.

# Detection of cryptic species and host-specific lineages

- **Species without a sexual generation**
  - *Aphis gossypii* (melon/cotton aphid)
  - *Pentalonia nigronervosa* (banana aphid)
  
- **Species with a sexual generation**
  - *Chaitophorus populicola*
  - *Pemphigus betae* (sugar beet root aphid)

# *Aphis gossypii*

## Melon aphid / cotton aphid



- cosmopolitan distribution.
- mostly **entirely asexual**; a few reported instances of sexual reproduction with host alternation.
- polyphagous, pest of diverse crops.
- **may represent a complex of species or biologically distinct populations.**

# *Aphis gossypii*

Specimens from a wide variety of hosts:

Agavaceae (*Cordyline*)

Araceae (*Colocasia*)

Asteraceae (*Bidens*, *Chromolaena*, *Chrysanthemum*, *Senecio*,  
*Youngia*)

Bignoniaceae (*Catalpa*, *Spathodea*)

Crassulaceae (*Kalanchoe*, *Sedum*)

Curbitaceae (*Cucumis*, *Cucurbita*)

Euphorbiaceae (*Euphorbia*)

Fabaceae (*Cajanus*)

Goodeniaceae (*Scaevola*)

Lamiaceae (*Clerodendron*)

Malvaceae (*Abelmoschus*, *Gossypium*, *Hibiscus*, *Sida*)

Poaceae (*Triticum*)

Portulacaceae (*Portulaca*)

Rosaceae (*Potentilla*)

Rubiaceae (*Ixora*)

Solanaceae (*Solanum*, *Capsicum*)

Urticaceae (*Pipturus*)

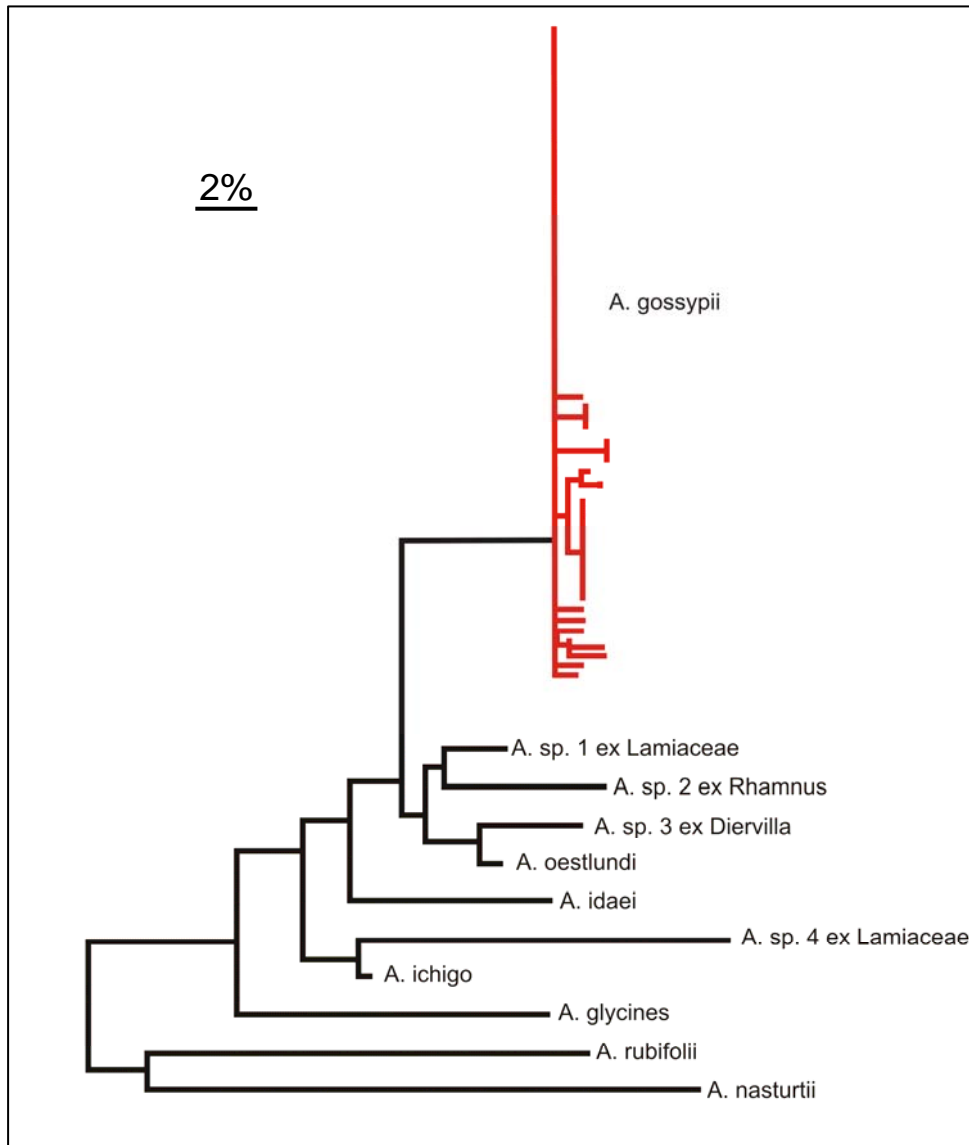
Verbenaceae (*Lantana*)

# *Aphis gossypii*

Specimens from a wide geographic range:

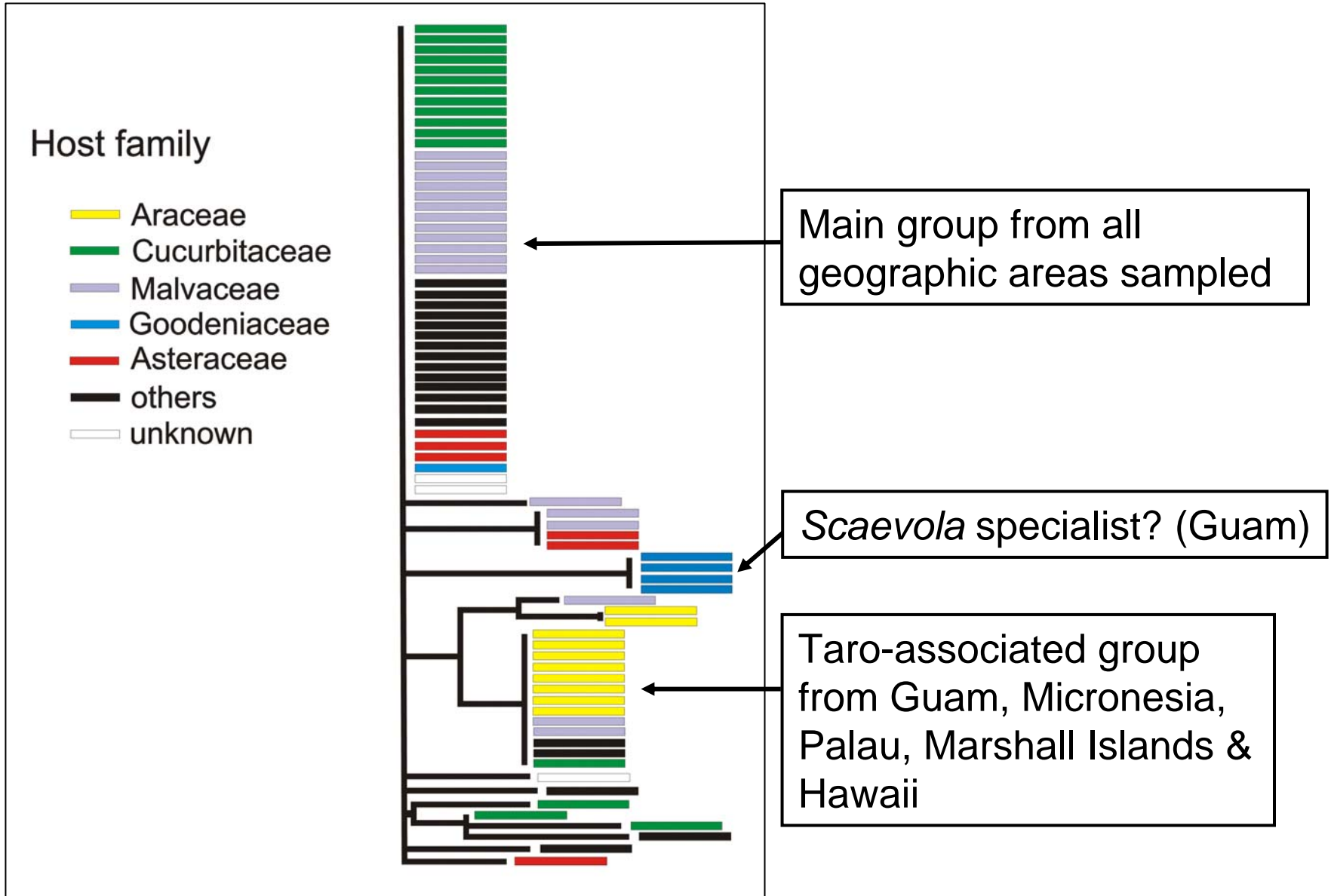


# *Aphis gossypii*



***A. gossypii* well defined:** low within-species sequence divergence relative to divergence among related species

# *Aphis gossypii*: within species variation



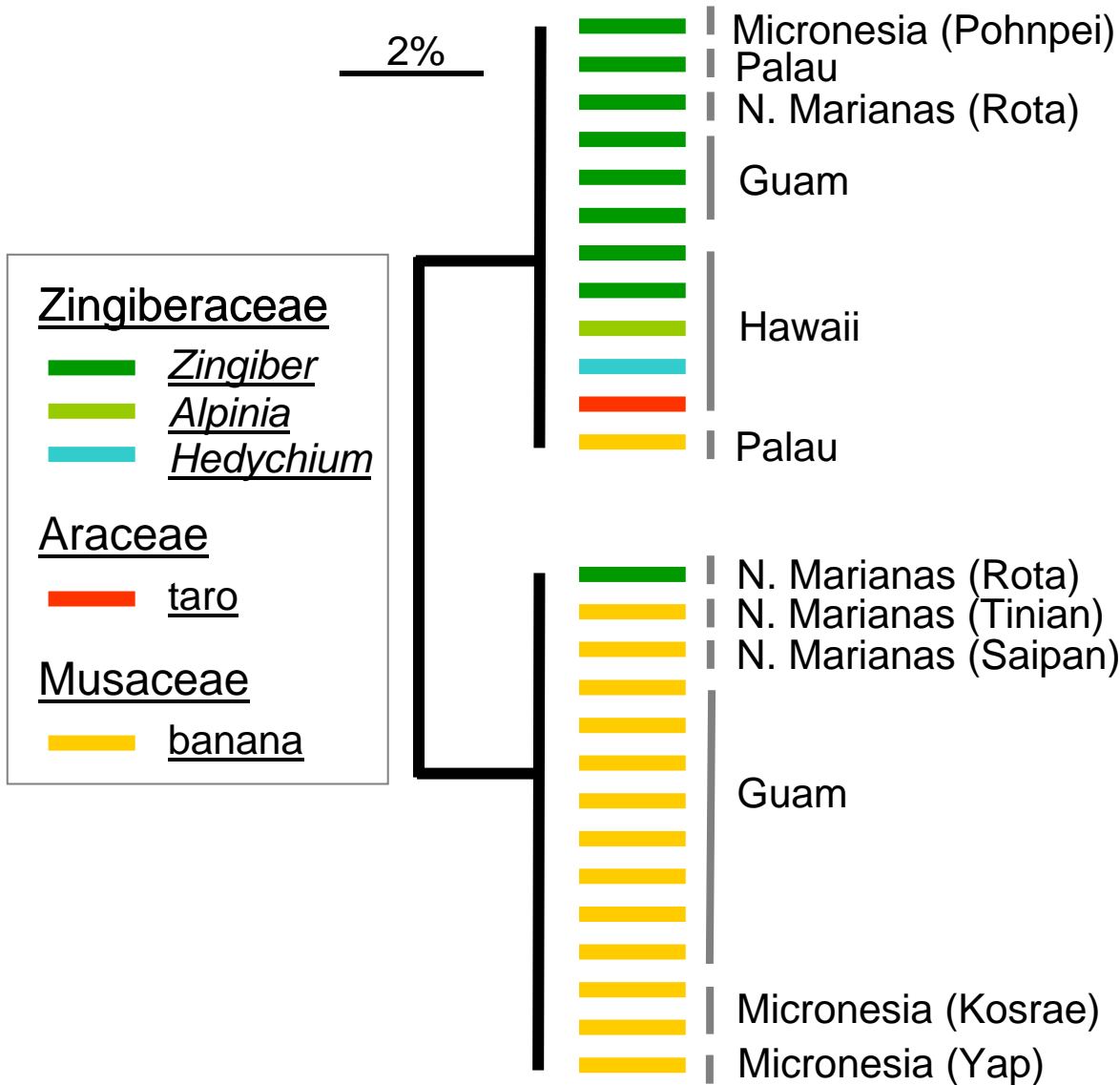
# *Pentalonia nigronervosa*



## Banana aphid

- **No sexual forms known**
- Pantropical
- Feeds on Musaceae, Araceae and Zingiberaceae
- Vector of banana bunchy-top virus

# *Pentalonia nigronervosa*



• **Distinct ginger- and banana-associated groups**

• Groups correspond to morphometrically distinct groups

• Specimens from other parts of world to be analysed

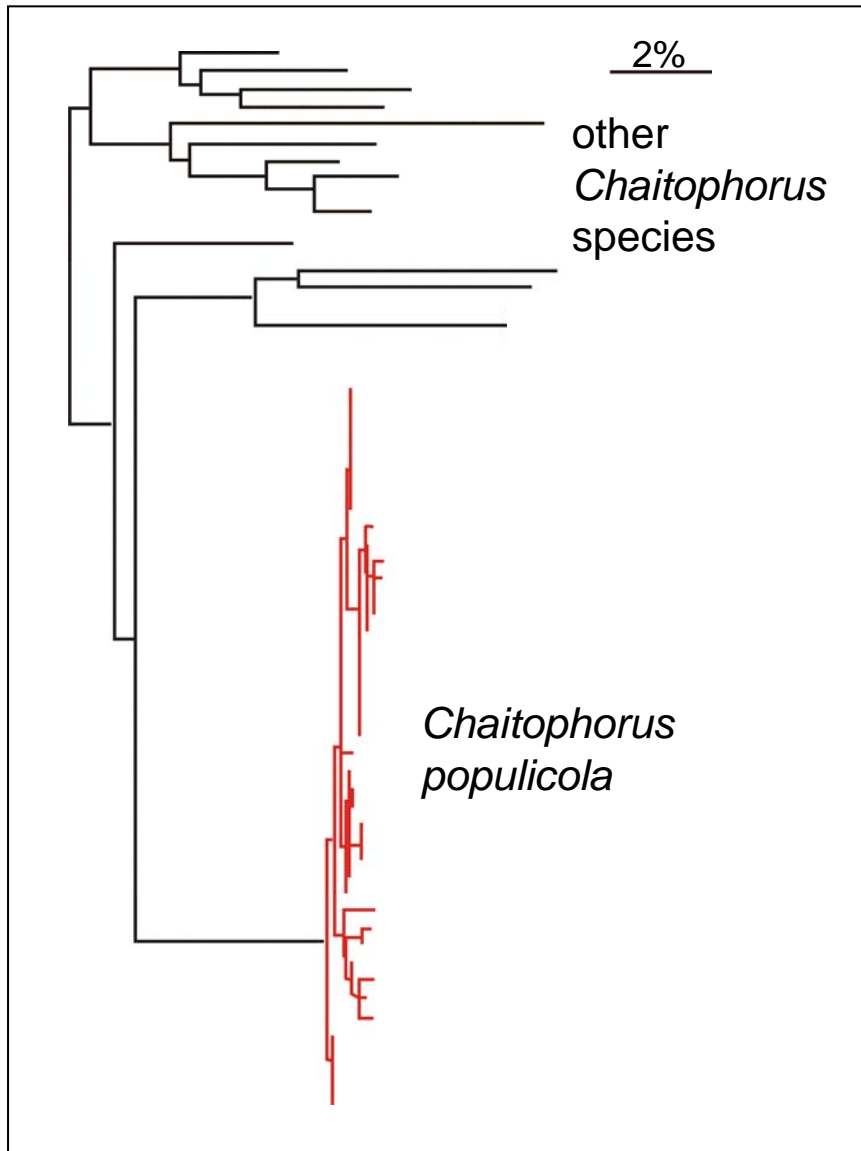
• To be confirmed with nuclear genes

# *Chaitophorus populicola*



- Transcontinental distribution in North America
- Feeds on various *Populus* species
- Variation in colour, body size and setal shape
- Obligatory sexual generation

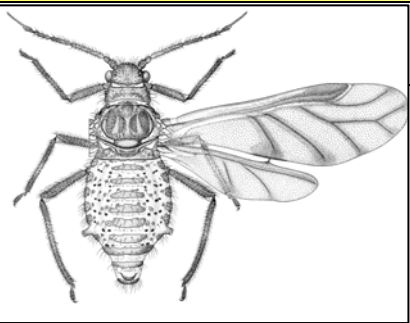
# *Chaitophorus populicola*



***C. populicola* variable  
but well defined:**

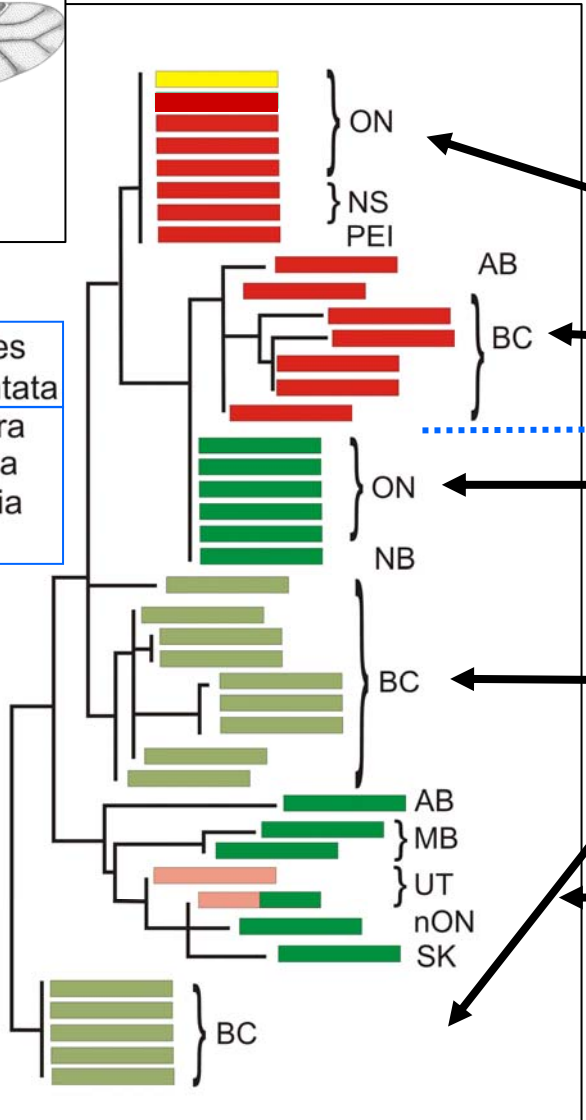
within-species sequence  
divergence low relative to  
among-species  
divergence within genus

# Chaitophorus populicola



Within species: geographic segregates + sympatric host-associated lineages

- Populus sp.
- tremuloides
  - grandidentata
  - balsamifera
  - trichocarpa
  - angustifolia
  - hybrid



eastern

on Populus (Populus)

western

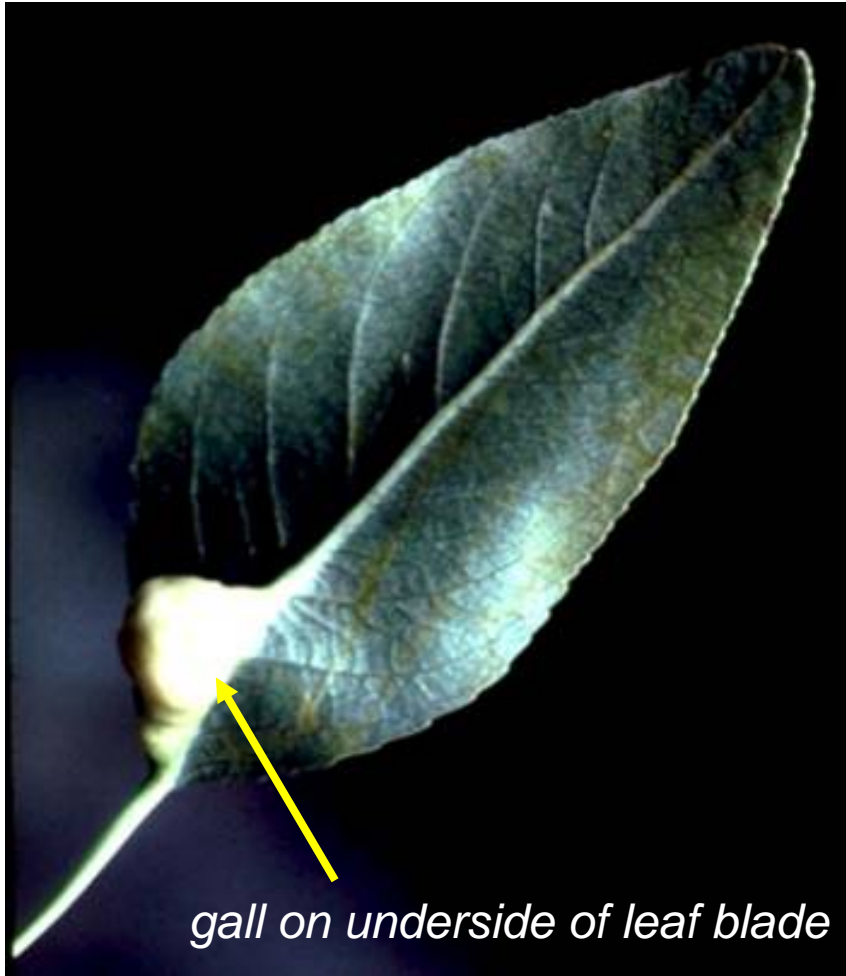
eastern

western

on Populus (Tacamahaca)

central

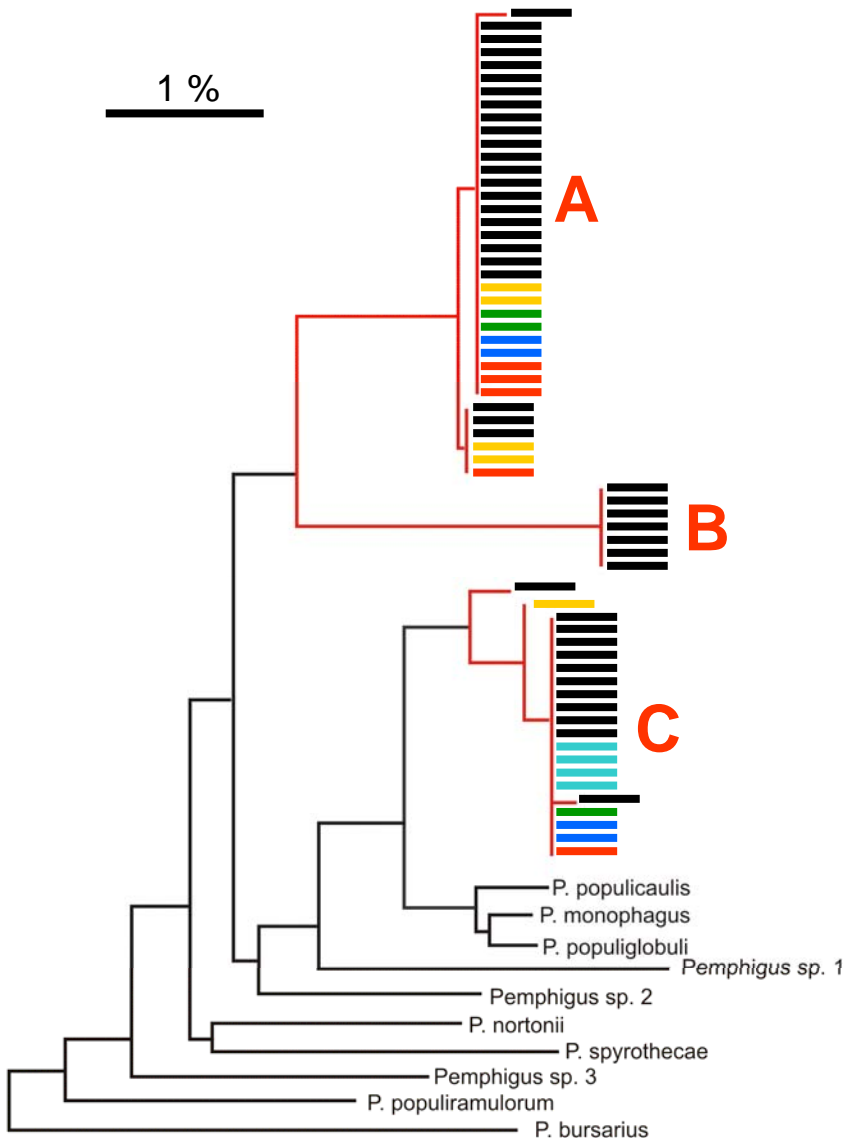
# *Pemphigus betae*



## *Pemphigus betae*

- Pest of sugar beet and other *Chenopodiaceae*.
- has a sexual generation with host alternation between *Populus* (gall) and herbaceous plants.
- normally lays over-wintering eggs on *Populus*.
- part of population may continue to reproduce asexually on secondary host and over-winter in soil.

# *Pemphigus betae*



- **Three distinct clusters** of “*Pemphigus betae*”; divergence as great as interspecific divergence among other species of genus
- Clusters correspond exactly to clusters independently determined by nuclear genes
- All clusters **sympatric** (samples belonging to 2 different clusters sometimes occur on same tree [indicated by bar colour; black bars are single samples all from different trees])
- “A” and “C” clusters found from Alberta to Arizona

# Life stage association

## Adelgidae:



Biennial alternation between spruce (sexual generations, in galls) and some other conifer (parthenogenetic generations, no galls).

Some populations (species?) apparently without alternation – restricted to either spruce or the alternate conifer.

# Life stage association

[with Nathan Havill, Yale University]

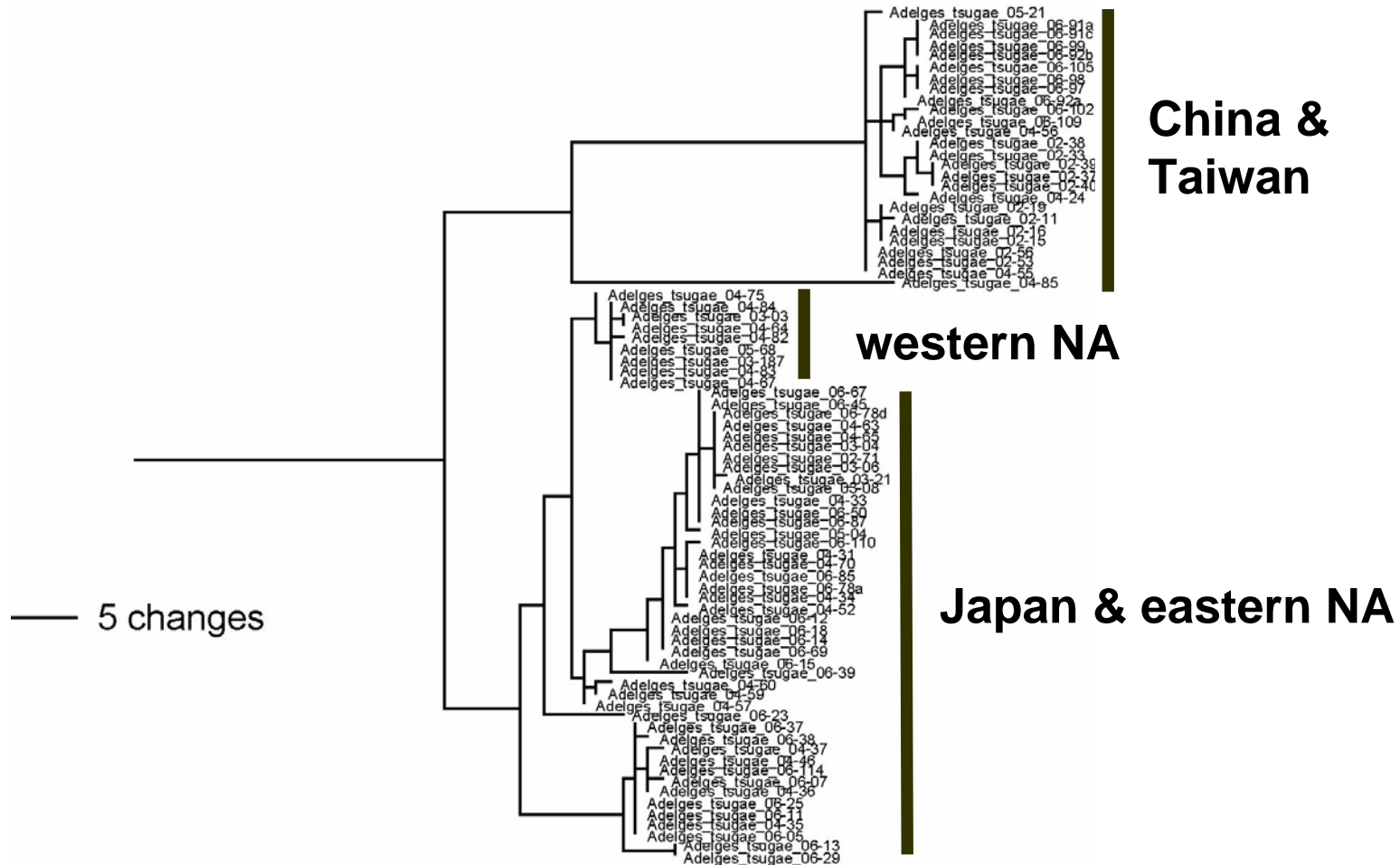
***Adelges tsugae*** - hemlock adelgid (hemlock woolly aphid)



- Origin in East Asia.
- Probably indigenous in western North America.
- Introduced in eastern North America, where it is a significant pest
- Previously known only from hemlock spp. (*Tsuga*).

# Life stage association

- Three geographically segregated clades associated with *Tsuga* under name *Adelges tsugae* (probably at least 2 species)



# Life stage association

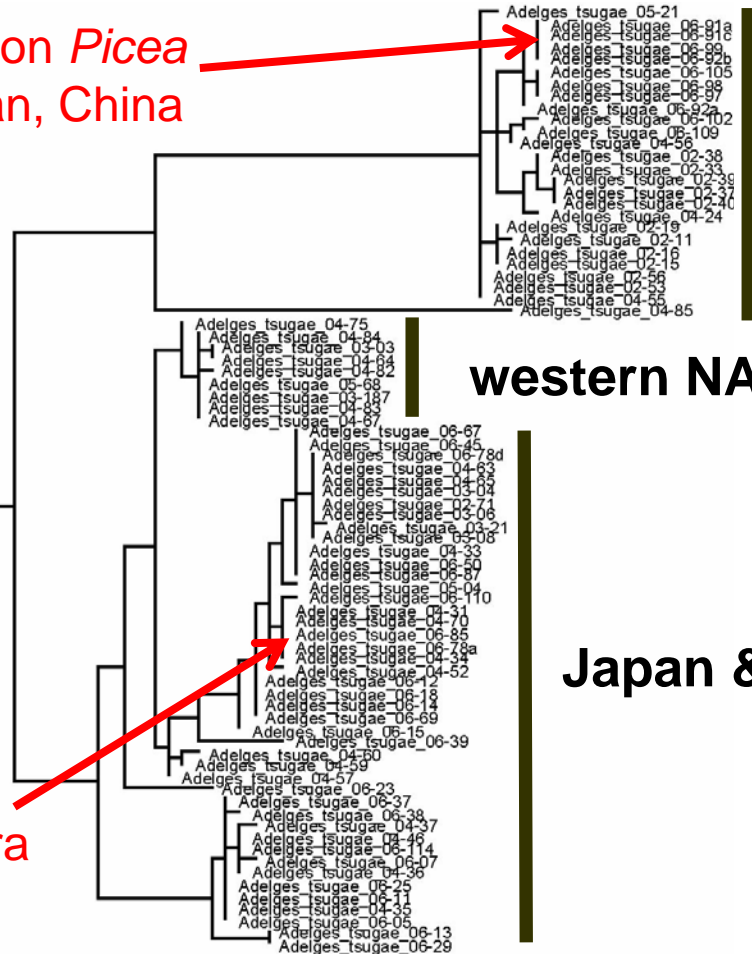
- Adelgids dissected from galls on spruce in China and Japan cluster within two of the “*A. tsugae*” groups.
- Probable previously unknown primary host forms of *A. tsugae* complex species.

Dissected from galls on *Picea likiangensis* in Yunnan, China



— 5 changes

Dissected from galls on *Picea polita* in Nara Prefecture, Japan



China & Taiwan

western NA

Japan & eastern NA

# Detection of Invasive Species



*Diuraphis noxia*

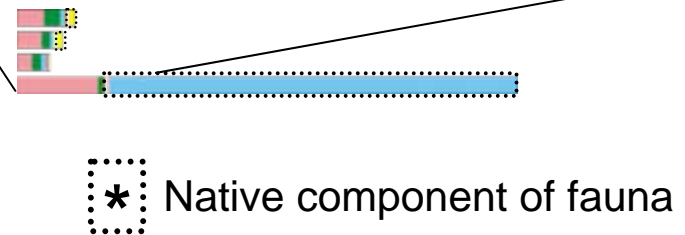
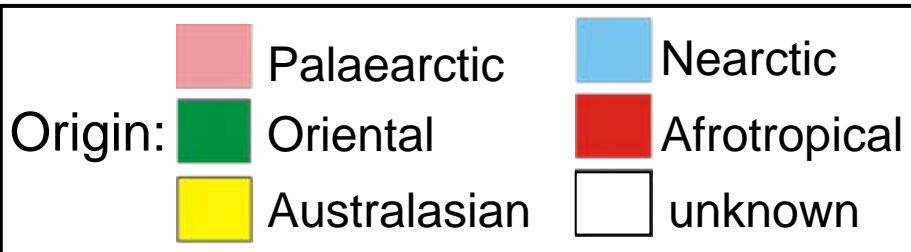
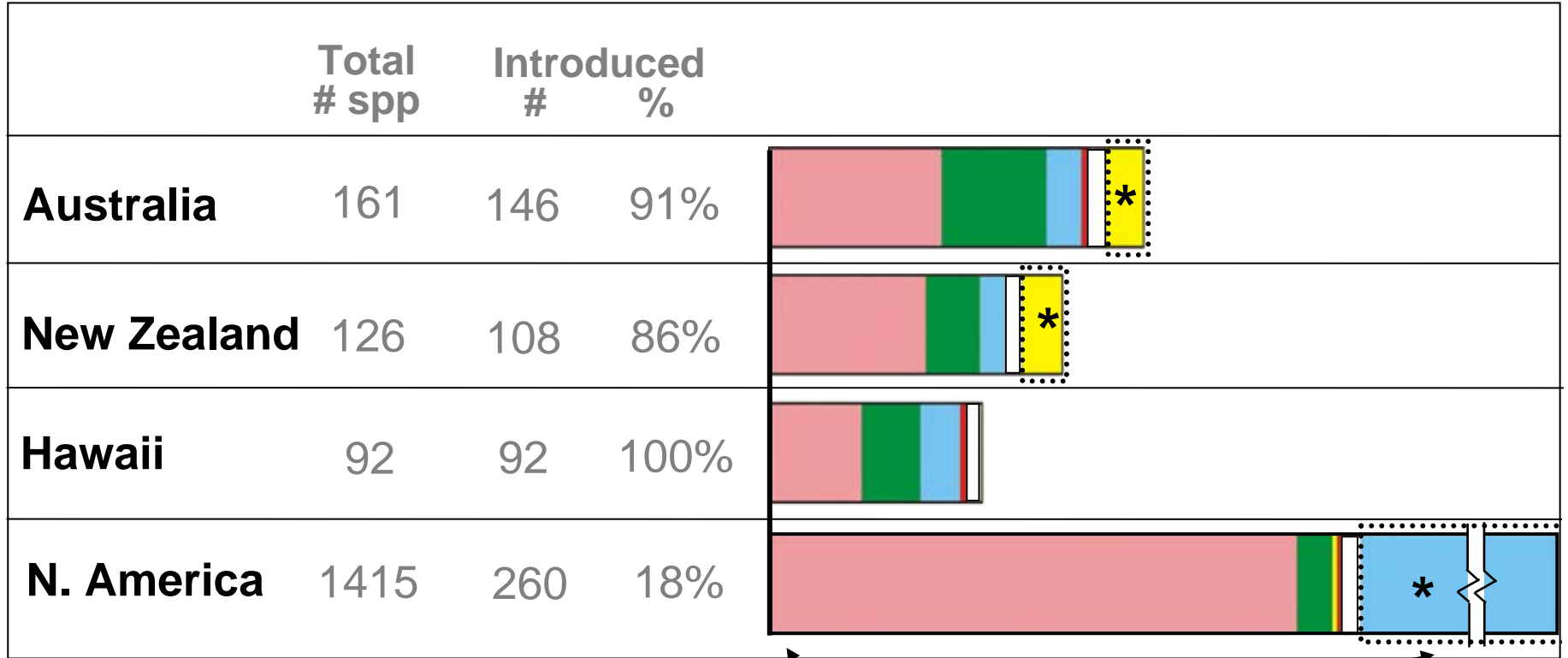


*Aphis glycines*

# Detection of Invasive Species

	<b>Total # spp</b>	<b>Introduced #</b>	<b>%</b>
<b>Australia</b>	<b>161</b>	<b>146</b>	<b>91%</b>
<b>New Zealand</b>	<b>126</b>	<b>108</b>	<b>86%</b>
<b>Hawaii</b>	<b>92</b>	<b>92</b>	<b>100%</b>
<b>N. America</b>	<b>1415</b>	<b>260</b>	<b>18%</b>

# Detection of Invasive Species



# Detection of Invasive Species

- A large proportion of adventive aphids shared

% introduced species shared with

all other  
regions

at least 2 other  
regions

Australia

31

65

New Zealand

42

78

Hawaii

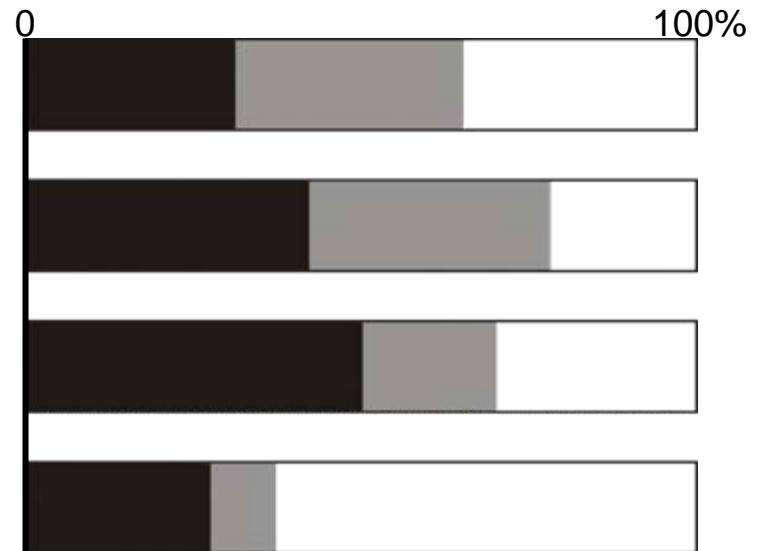
50

69

N. America

17

37



Shared by all 4 regions



Shared by 3 of 4 regions

# Detection of Invasive Species

Initiating study to:

- Barcode all known introduced species in various regions of world (pilot: Australia, New Zealand, Hawaii, North America).
- Barcode other aphids feeding on crops and crop relatives.
- Establish degree of variation within each species worldwide.

# Acknowledgements

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