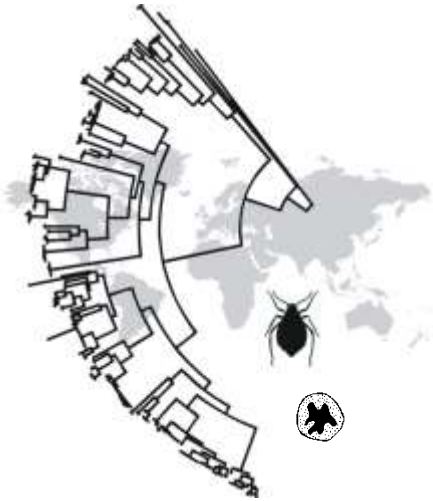


Evolution of obligate bacterial endosymbiosis in aphids



AAP grands projets
de séquençage
2015: *MicroPhan*

Symbiosis with bacteria, has favored the colonization of « nutrient poor » environments



Deep sea tube worms use bacteria for chemosynthesis



AA Nikoh, Hosokawa, Moriyama, Oshima, Hattori & Fukatsu. 2014.

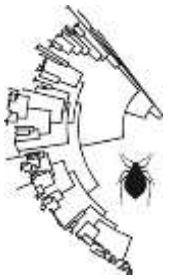
Blood-feeding arthropods host bacteria that synthesize Vitamins



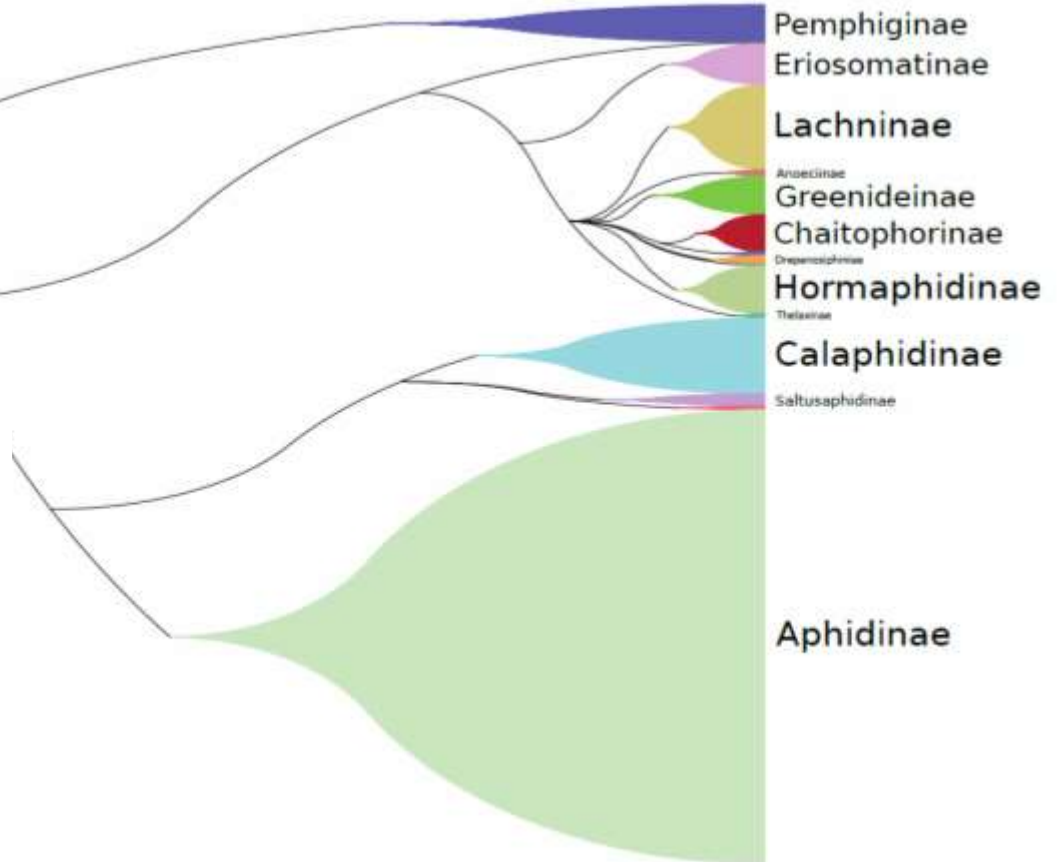
Sap-feeding insects host bacteria that synthesize AAs and vitamins

Mutualistic bacteria synthesize nutrients that are missing in their hosts diet
Bacteria that live within their host=endosymbionts

Bacterial endosymbiosis in aphids

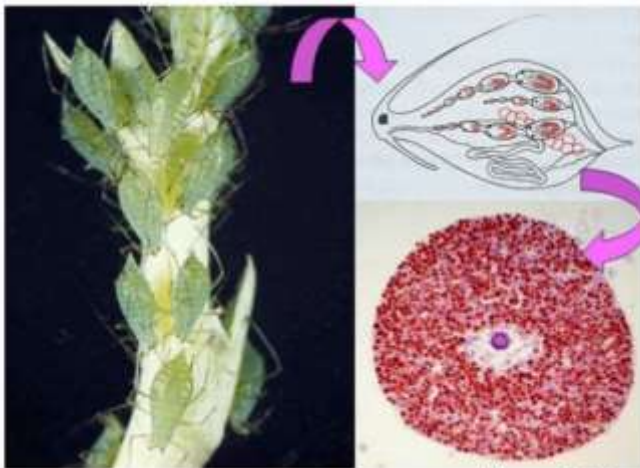


Ancestral acquisition of *Buchnera aphidicola*, coevolving with its host for ≈ 200 Myrs



Vertical transmission

Novakova et al., 2013



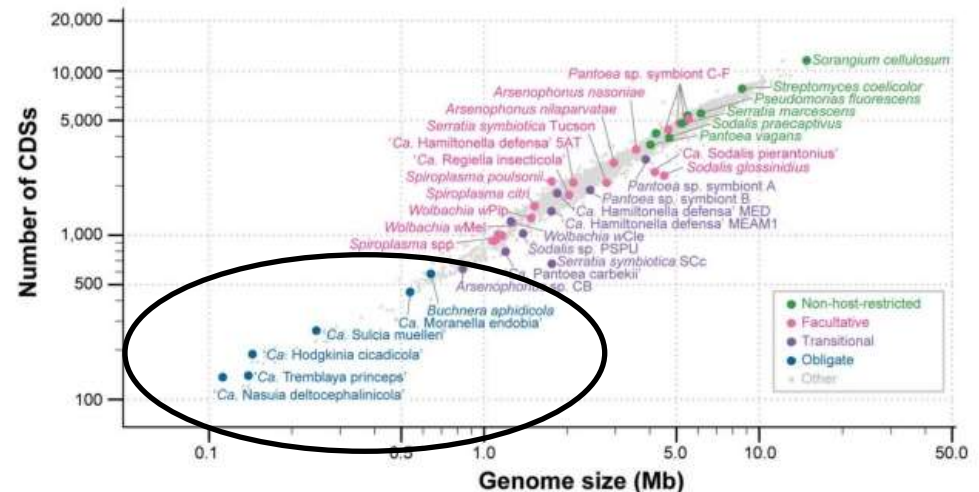
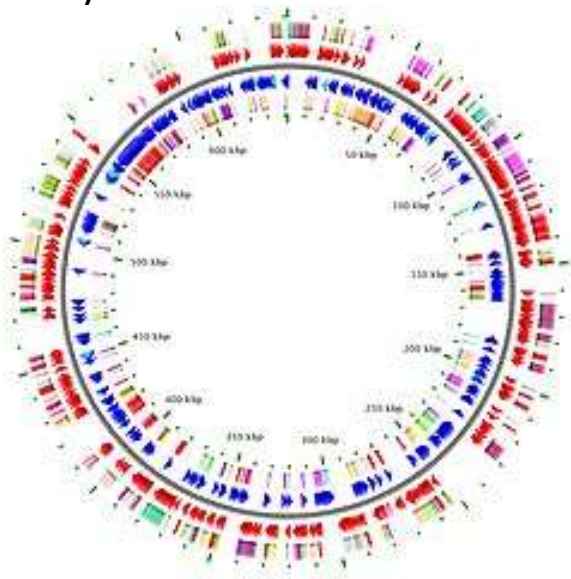
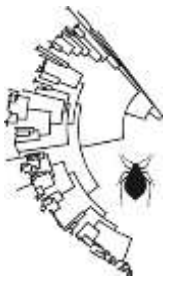
Takema Fukatsu

Bacterial endosymbiosis in aphids

Buchnera aphidicola (Buchner 1965)

Synthesizes AA and vitamins for its host (i.e. nutrients lacking in the phloem)

First obligate endosymbiont genome to be sequenced: it is small 0,6Mb (Shinegobu, 2000)

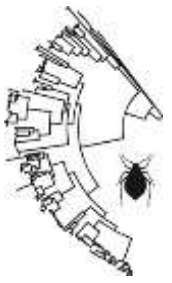


Lo et al. (2016). *FEMS Microbiol Rev* **40**(6):855-874

Endosymbionts preserve genes that serve the hosts

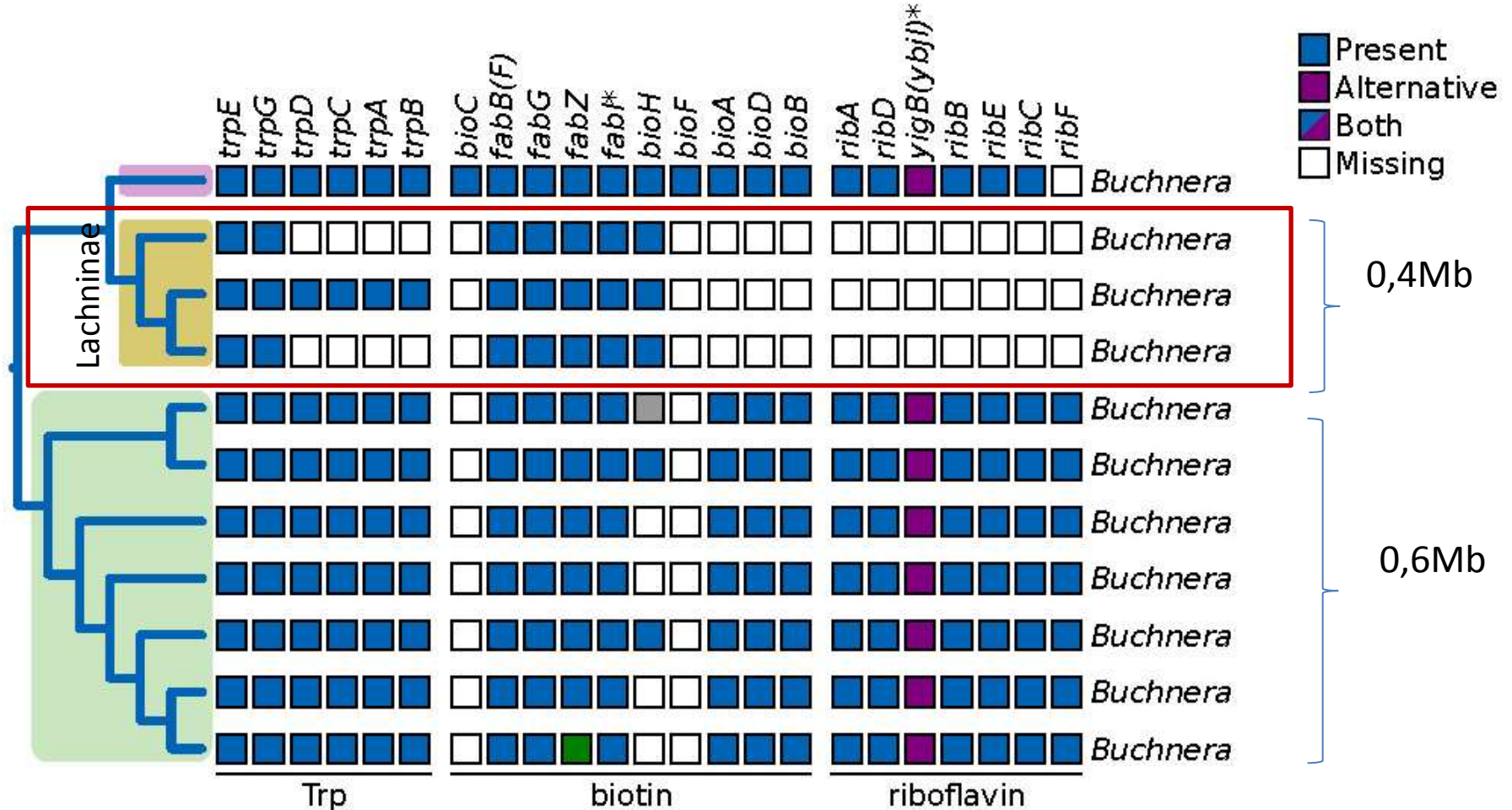
Genomic erosion caused by : deletion of genes useless to intracellular life-style , strong genetic drift due to recurrent bottlenecks (fixation of deleterious mutations, deletion of genes)

Bacterial endosymbiosis in aphids



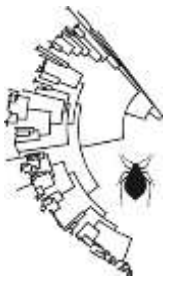
In *Lachninae* *Buchnera* only 0,4Mb and losses of key metabolic functions

Who does it if not *Buchnera*?

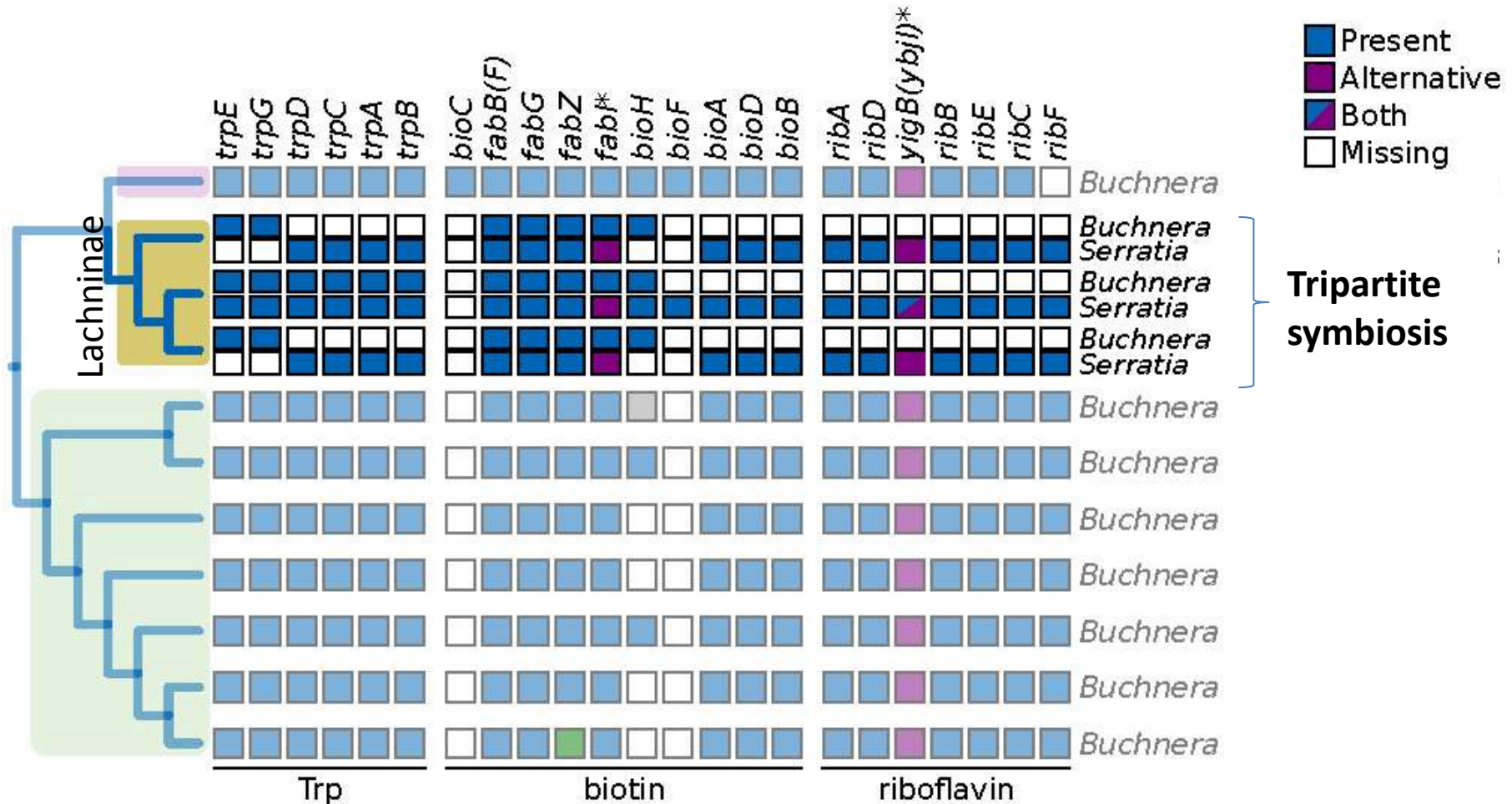


(Perez brocal et al, 2006, Lamelas et al. 2011, Manzano-Marin et al. 2014, 2016)

Bacterial endosymbiosis in aphids

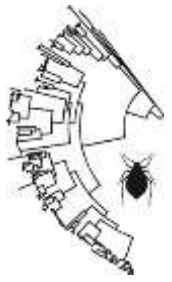


A previously facultative symbiont is complementing *Buchnera*



(Perez brocal et al, 2006, Lamelas et al. 2011, Manzano-Marin et al. 2012, 2016)

Questions



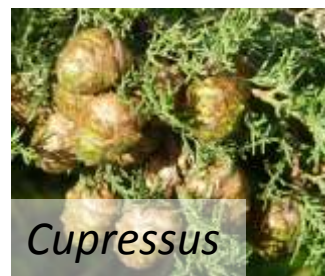
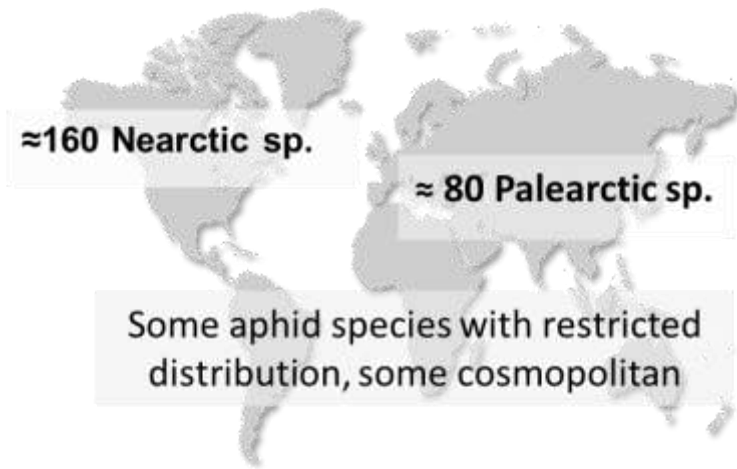
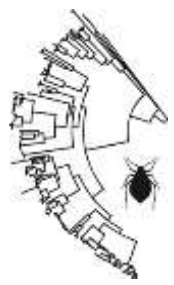
- History of the associations between Lachninae aphids and their new co-obligate symbionts: who, when ?
- Role of bacterial associations in aphids evolutionary transitions: do new symbionts carry new metabolic functions?
- Rates and patterns of molecular evolution in bacterial endosymbionts:
 - horizontal gene transfer between symbionts?
 - pace of genome “decay” in symbionts ?

What evolutionary processes drive the evolution of obligate symbionts ?

Bacterial endosymbiosis in aphids

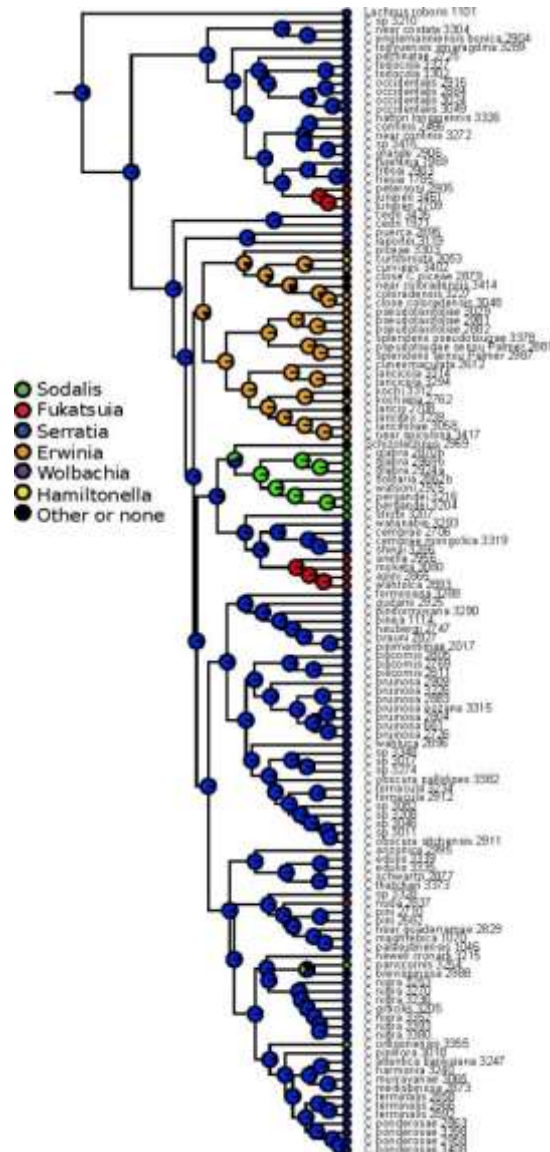
240 spp. of *Cinara* (Lachninae)

They have diversified 40-50mya



Evolution of endosymbiosis throughout the ecological diversification of *Cinara*?

Evolution of co-obligate symbionts in *Cinara*



16S rRNA gene survey (high-throughput sequencing)
on 500 samples (≈100 species)

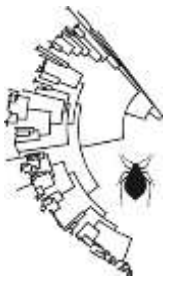
Association with *Serratia* in most species

When *Serratia* is « lost », it is replaced by
another endosymbiont (*Erwinia*, *Sodalis*,
Wolbachia)

Metabolic complementarity of Buchnera and
these new endosymbionts?

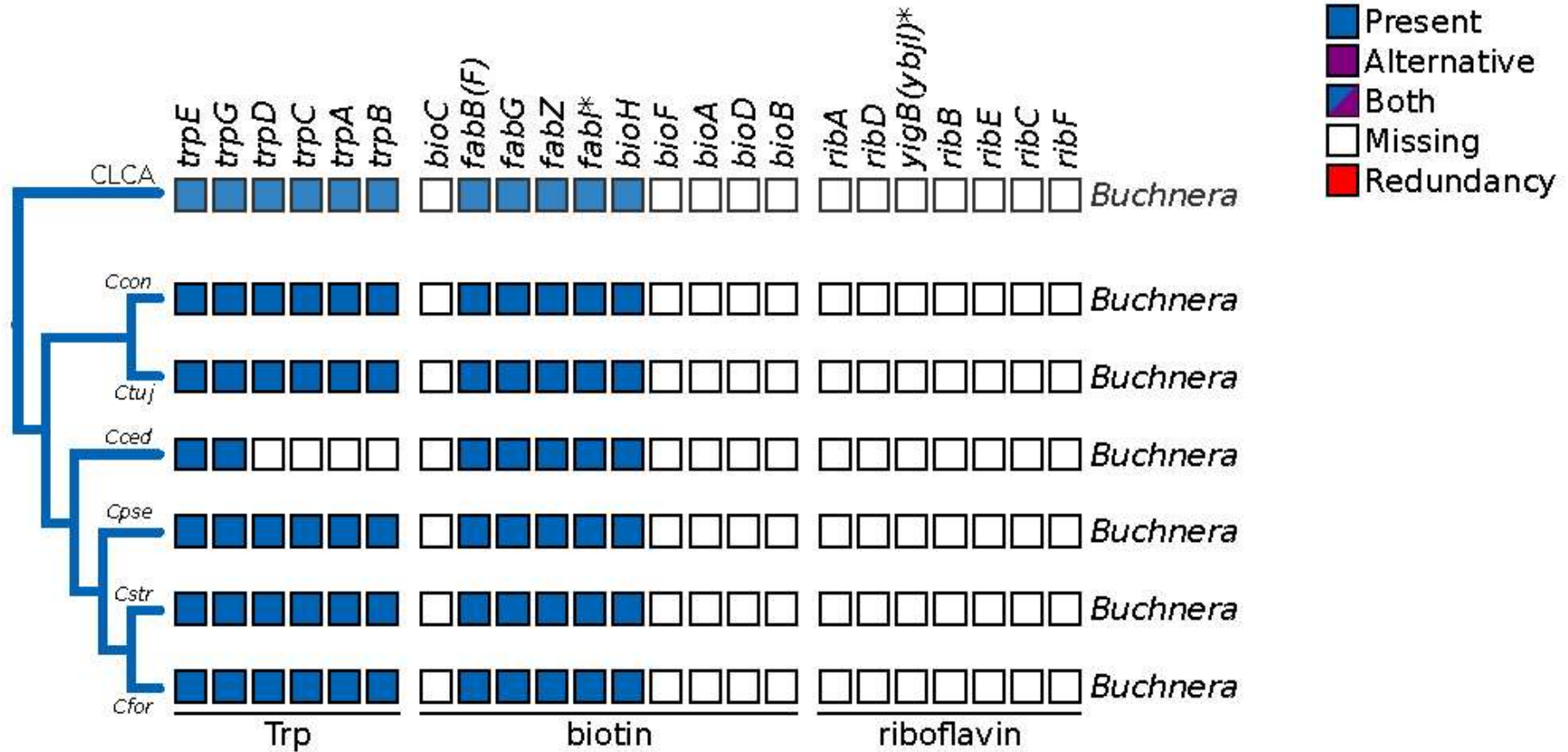
- whole genome sequencing of the endosymbionts
of about 50 *Cinara* species

Evolution of co-obligate symbionts in *Cinara*



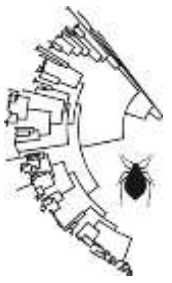
Buchnera genome Evolution

Losses of the same key functions in *Buchnera* of all *Cinara* species

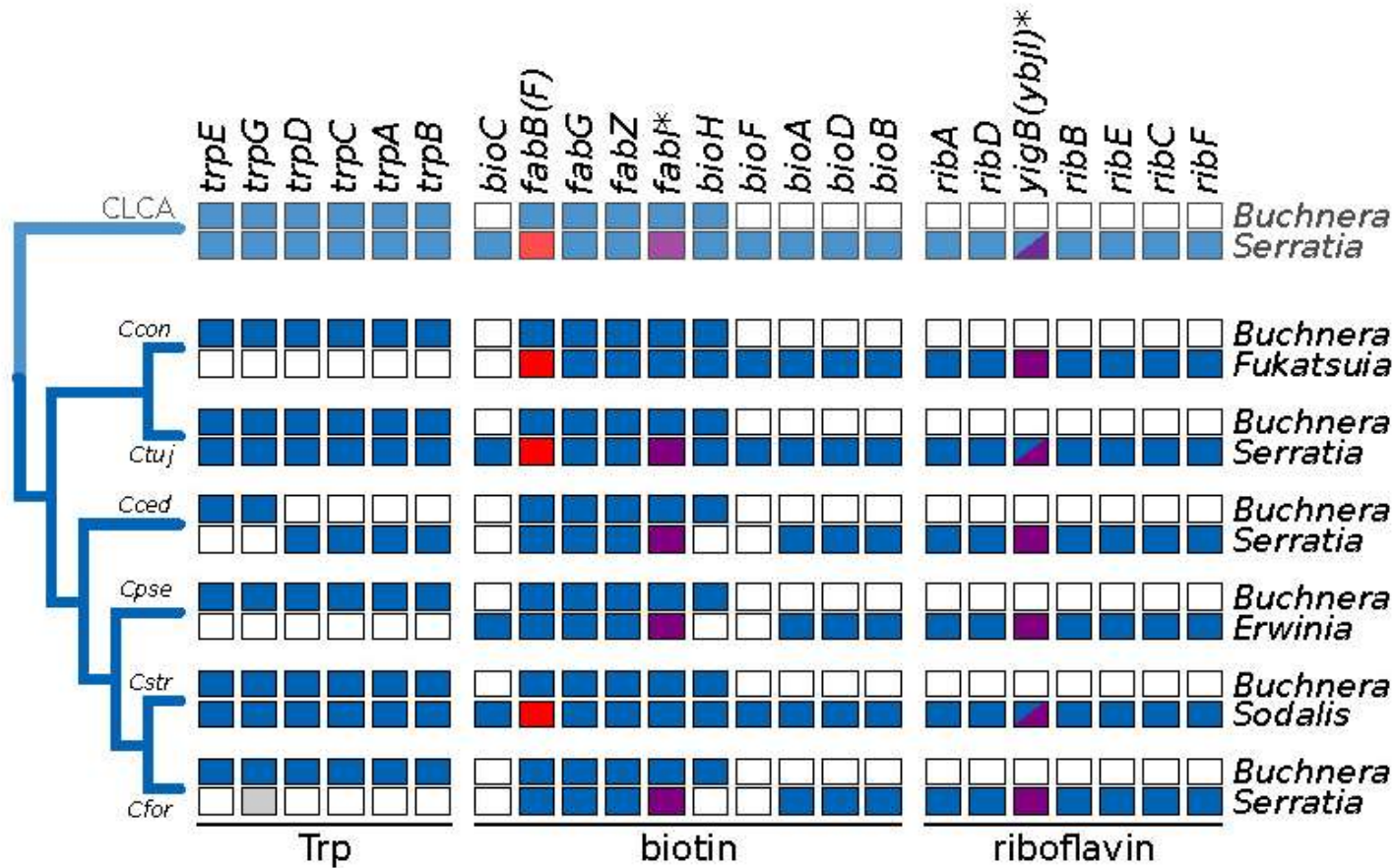


Ancient loss of biotin and riboflavin genes followed by genome « stasis »

Evolution of co-obligate symbionts in *Cinara*

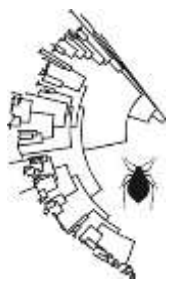


All co-symbiotic lineages can complement *Buchnera*



Tri-partite symbiosis in which one of the partners is repeatedly replaced

Evolution of co-obligate symbionts in *Cinara*



Ecological factors associated with the replacement of co-symbionts?



Is the identify of the symbiont lineage dependant on host plant genus? *Logistic phylogenetic regression*

Not significant, changes in symbiotic partner are not associated with shifts to new host-plant genera (nor feeding site etc..)

Is the presence/absence of a symbiont lineage dependant on climatic factors?
(we retrieved climatic data for all 500 samples, from WorldClim)

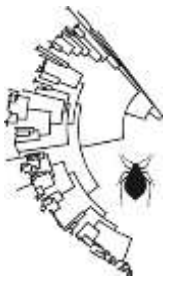


Climatic envelopes of specimens hosting different symbionts similar

No association between symbiont swaps and aphid's niche shifts

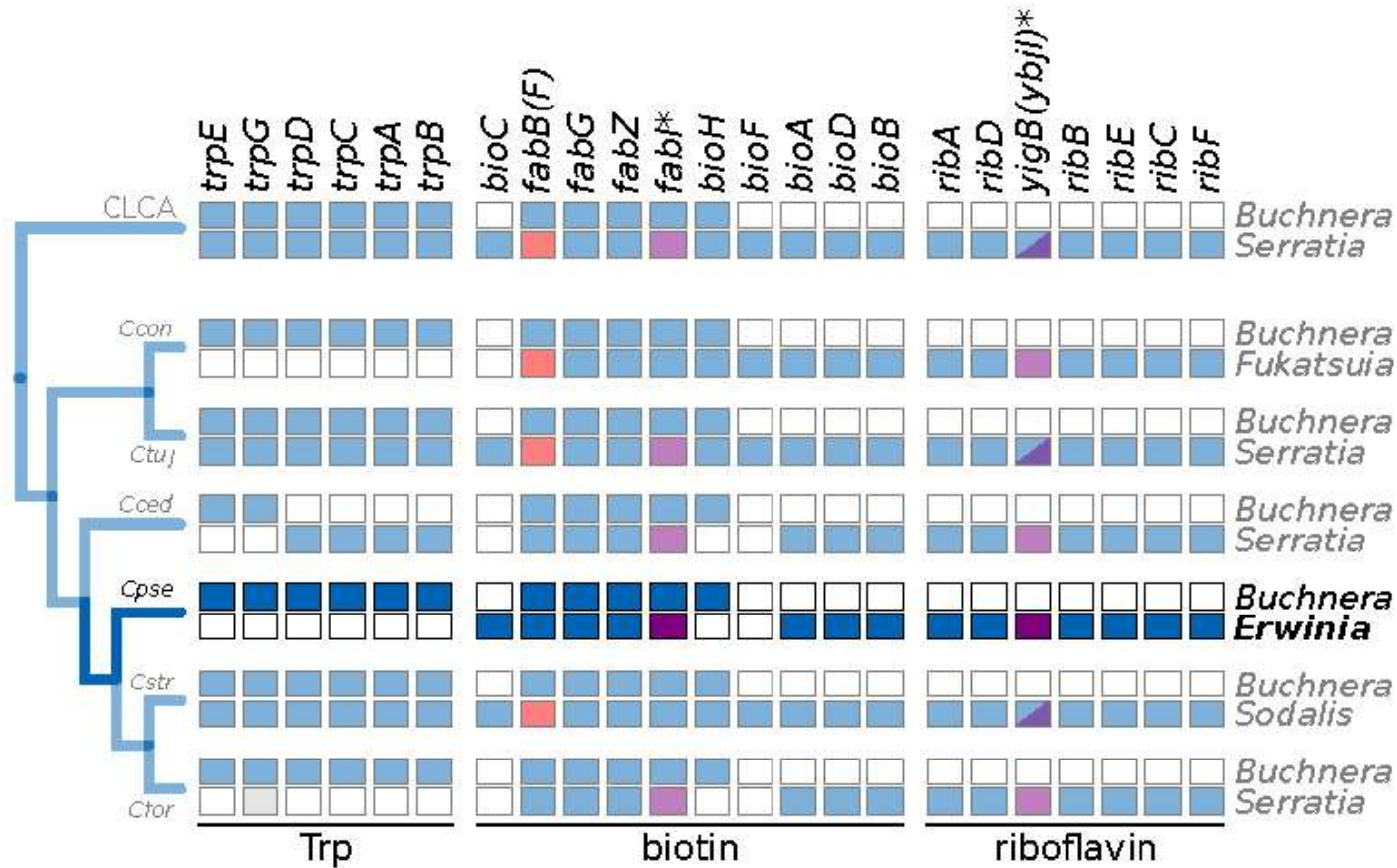


Evolution of co-obligate symbionts in *Cinara*



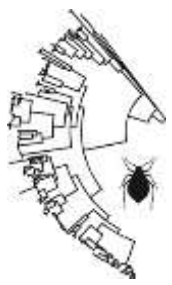
Endosymbiont genome evolution

Erwinia as a co-obligate symbiont



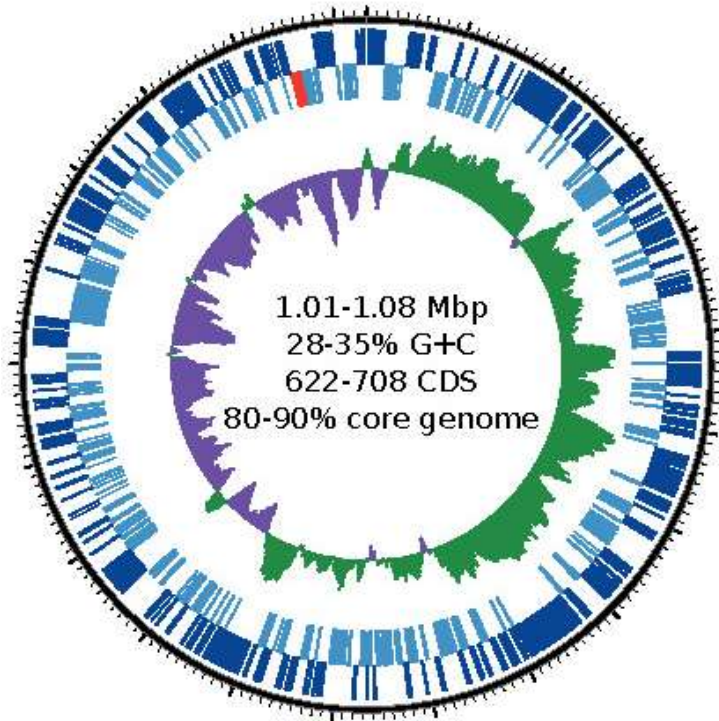
(Manzano-Marin et al. unp.)

Evolution of co-obligate symbionts in *Cinara*

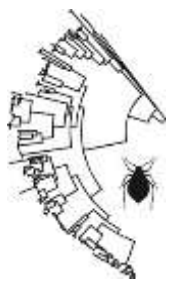


Endosymbiont genome evolution

Erwinia has a small genome, AT rich

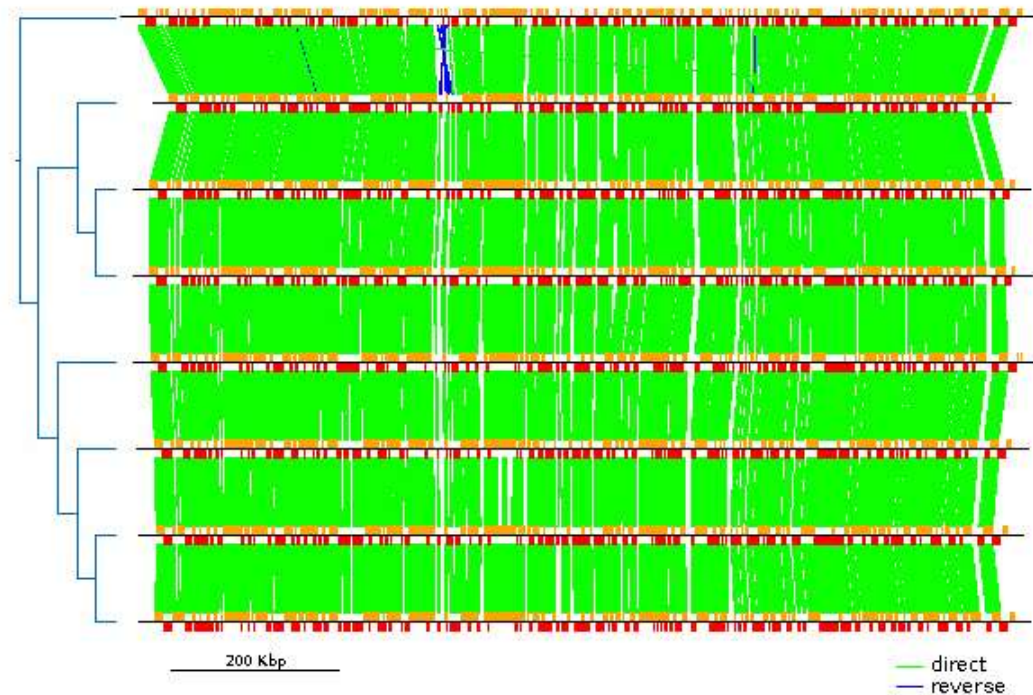
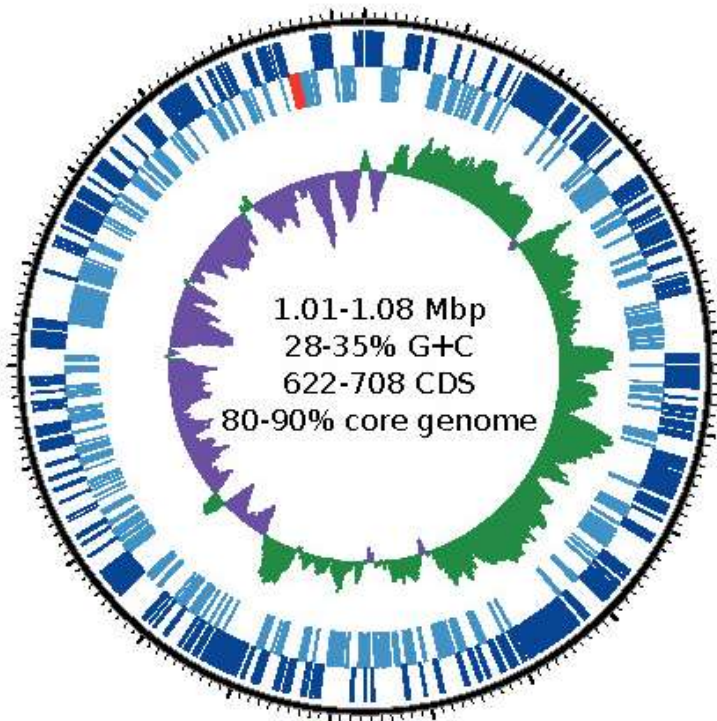


Evolution of co-obligate symbionts in *Cinara*



Endosymbiont genome evolution

Erwinia genomes are highly syntenic

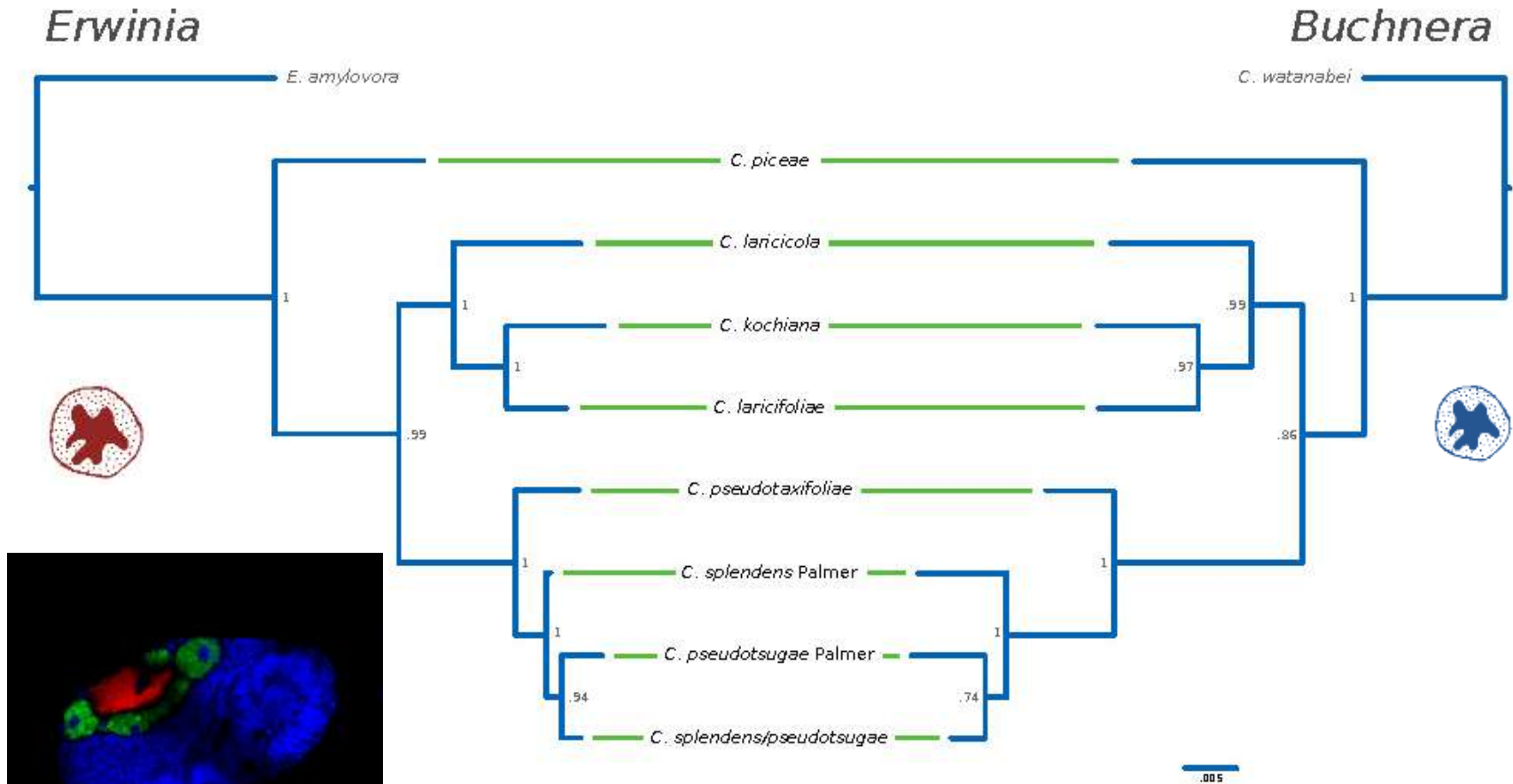
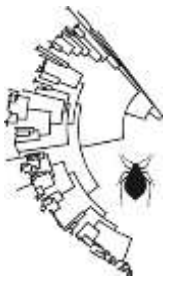


Manzano-Marín *et al.* Unpublished

Evolution of co-obligate symbionts in *Cinara*

Endosymbiont genome evolution

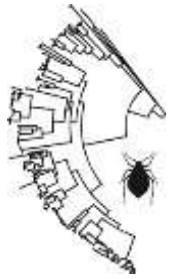
Erwinia has cospeciated with its host since its acquisition



Erwinia found in specialized cells, probably vertically transmitted

Manzano-Marín *et al.* Unpublished

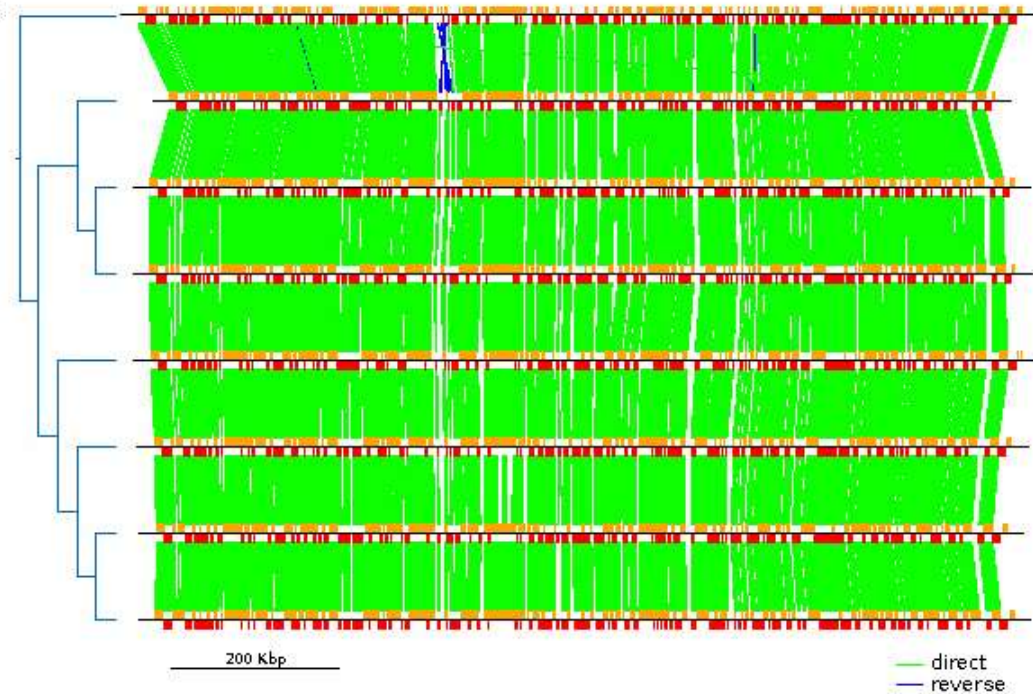
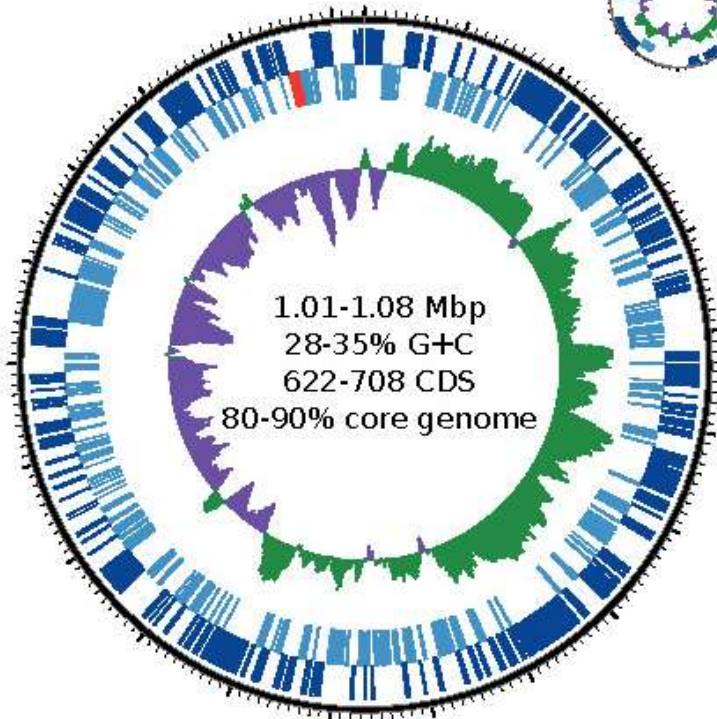
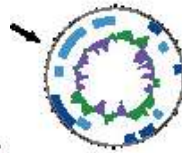
Evolution of co-obligate symbionts in *Cinara*



Erwinia Plasmid involved in biotin biosynthesis

Thiamine (B1)

Biotin (B7)



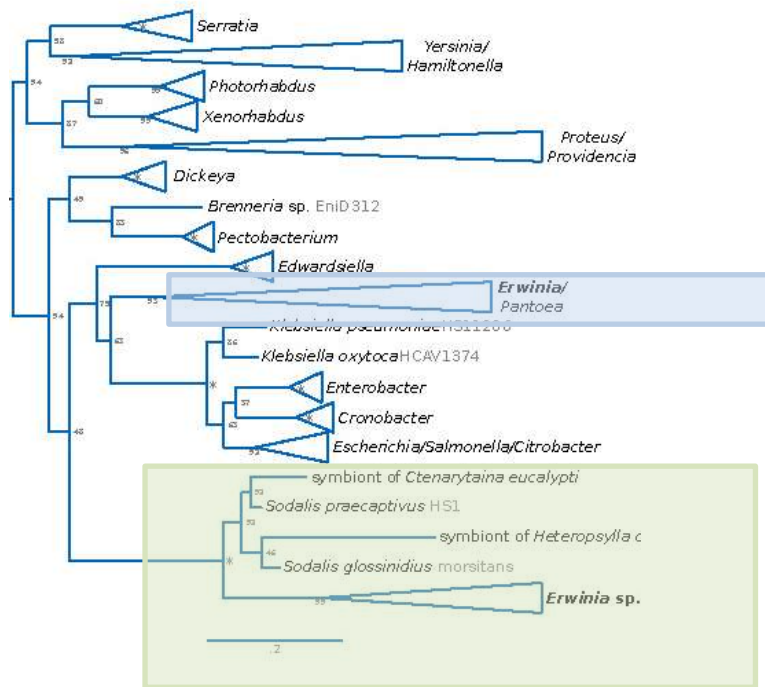
Manzano-Marín *et al.* Unpublished

Evolution of co-obligate symbionts in *Cinara*

Endosymbiont genome evolution

Erwinia has acquired a biotine gene via horizontal transfer

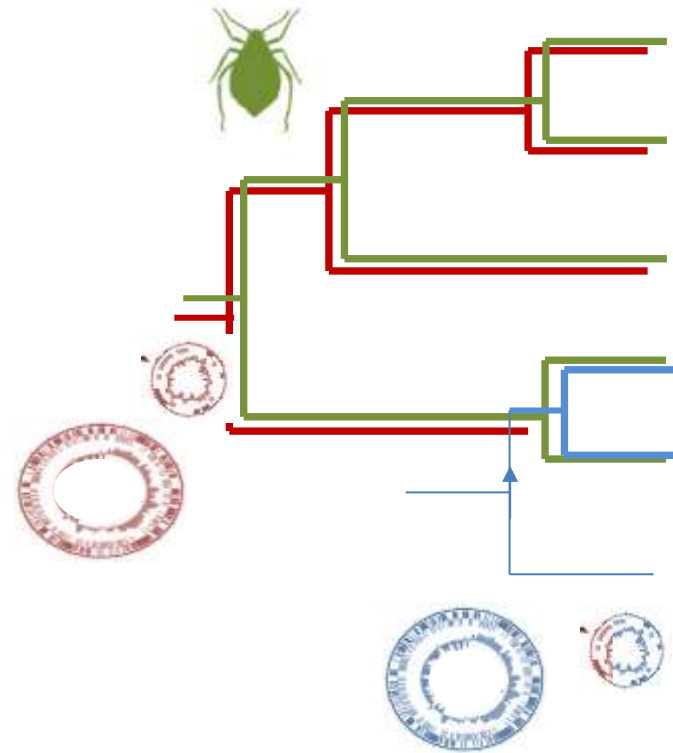
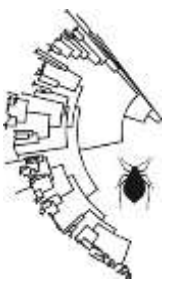
BioA



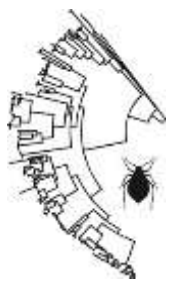
Horizontal gene transfers could play a role in symbiont replacements

Ancestral acquisition of *Serratia*, loss of *Serratia* and acquisition of *Erwinia* followed by cospeciation

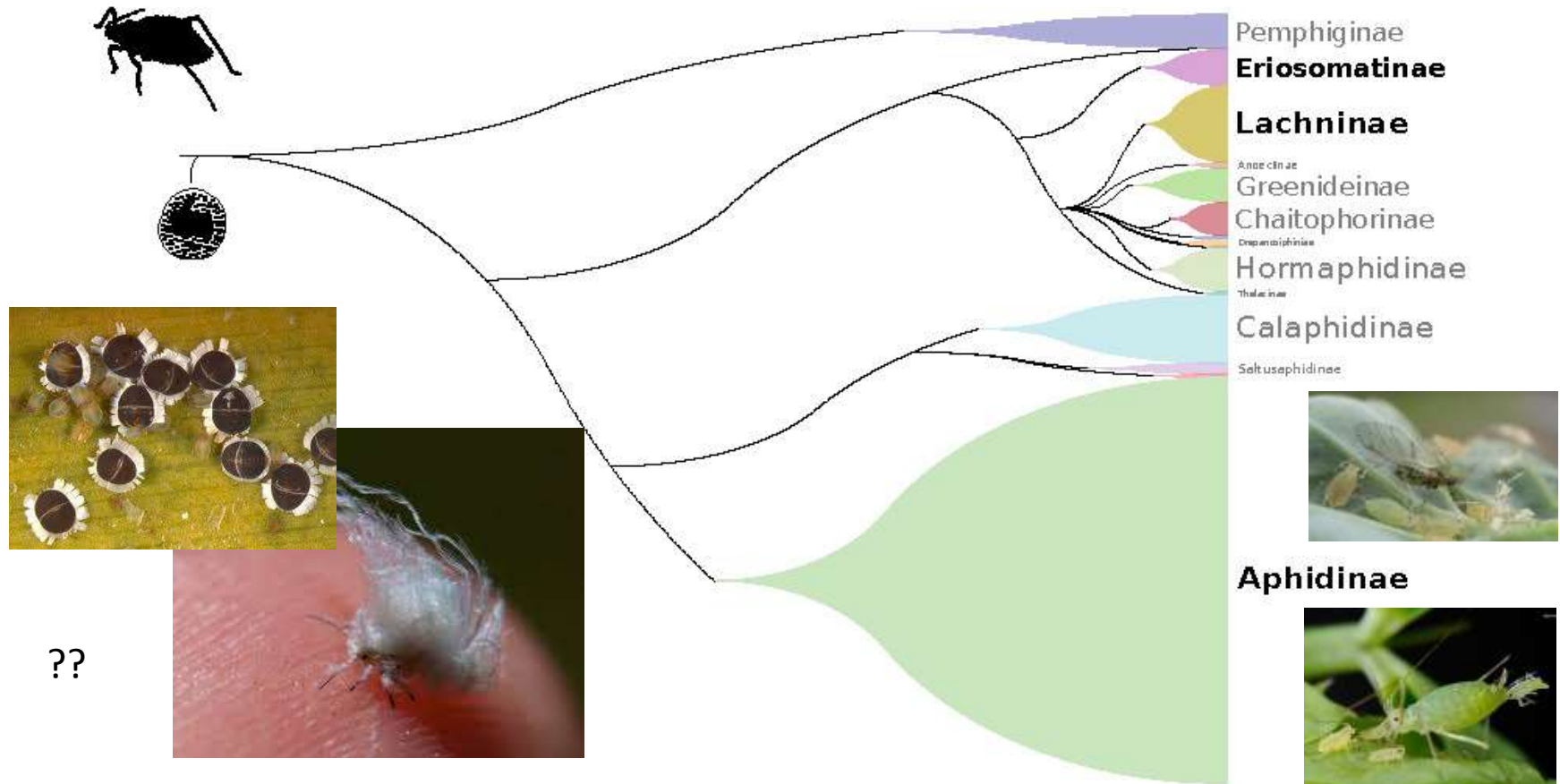
(Manzano-Marin et al. unp.)



Outside of *B. aphidicola* from the pea aphid and now from *Cinara* spp. we know little about *Buchnera*



Our knowledge comes from three subfamilies

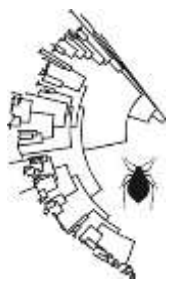


Based on Nováková *et al.* (2013). *Mol Phylogenet Evol* **68**(1):42-54

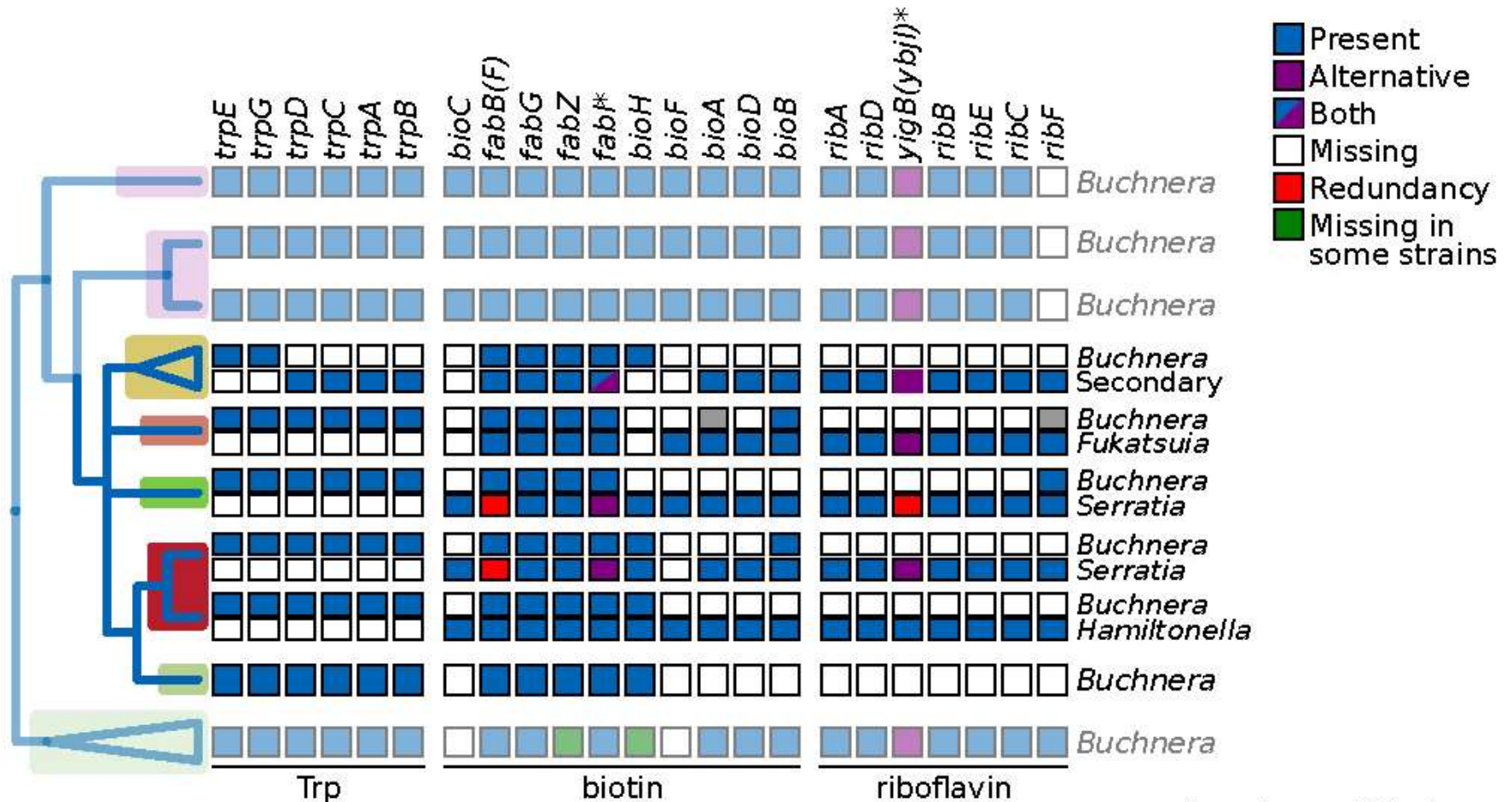
Phylogenetic tree and gene presence matrix for *Buchnera* strains. The tree on the left shows relationships between 10 strains, with color-coded clades: pink (top), yellow (middle), red (bottom), and green (bottom-most). The matrix to the right shows the presence (blue) or absence (white) of 21 genes across these strains. Genes are grouped by function: Trp (trpE, trpG, trpD, trpC, trpA, trpB), biotin (bioC, fabB(F), fabG, fabZ, fabI*, bioH, bioF, bioA, bioD, bioB), and riboflavin (ribA, ribD, yigB(ybjI)*, ribB, ribE, ribC, ribF). A legend indicates: blue = Present, purple = Alternative, pink = Both, white = Missing, red = Redundancy, green = Missing in some strains.

Manzano-Marín *et al.* Unpublished

Exploring *Buchnera* across aphids

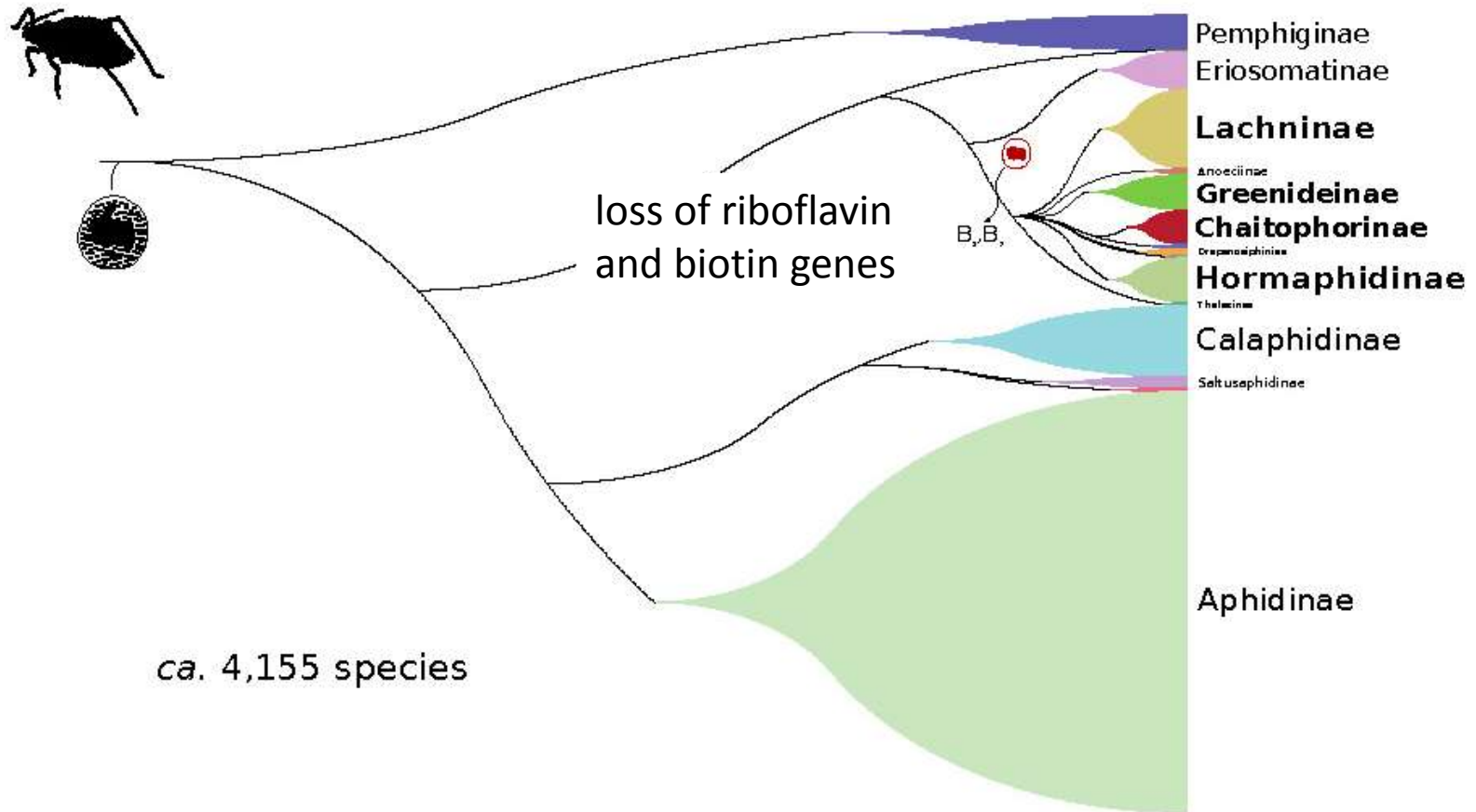
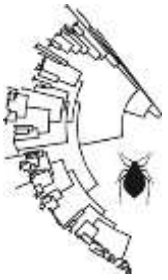


« Deficient » *Buchnera* rely on co-obligate endosymbiont in 5 subfamilies

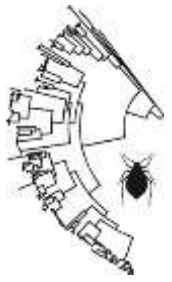


Manzano-Marín *et al.* Unpublished

Exploring *Buchnera* across aphids



Conclusions



Diversity and history of the associations between *aphids* and their co-obligate symbionts:

Di-symbiotic systems occur repeatedly in aphids

The partners may diversify through cospeciation until the co-symbiont is replaced, recurrent replacement of the “new-comer”

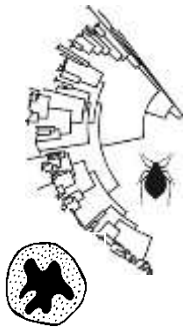
Rates and patterns of evolution in bacterial endosymbionts:

Chimeric endosymbionts: HGT between symbionts occur and could play an important role in symbiont replacement.

Role of bacterial associations in aphids evolutionary transitions:

New co-symbionts carry essential metabolic functions, no specific functions associated with aphids ecological niche so far...

Co-symbiont dynamics could be the result of competitive interactions between symbionts that carry similar functions (*& not selective pressures on the aphid host*).



no doubt that in many lineages foreign microbes have repeatedly invaded the established symbiotic associations, coexisting or replacing the original endosymbionts [19, 37]. Moran and Baumann [37] suggested that such replacement events could be governed by host adaptations favoring one or the other microbial type and/or by evolution of the microbes themselves. We agree with them, but emphasize that many of the invasion and replacement events may be neutral rather than advantageous to the hosts, and that the neutrality should be responsible for a large part of the conspicuous diversity of endosymbiotic systems of Homoptera we can observe now.

Fukatsu, 1994

Ongoing work :

- Evolutionary history of associations in *Cinara*
- Comparative genomics of endosymbionts
- Estimate rates of genome evolution with the calibrated phylogeny of the aphid

My obligate symbionts



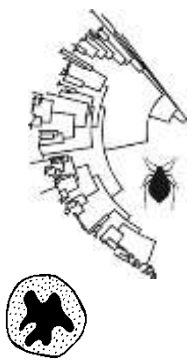
Alejandro Manzano-Marin (post-doc)



Armelle Cœur d'acier (aphid systematics)



Anne-Laure Clamens (Molecular biology)



Valerie Barbe
Céline Orvain
Corinne Cruaud



Andrea Sanchez-Meseguer (post-doc)



Marianne Annonier (M2)

