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Maternally inherited bacterial symbionts infecting arthropods have major implications on host ecology and evolution. Among them, the genus *Arsenophonus* has particular characteristics (large host spectrum, wide range of symbiotic relationships), making it a good model to study the evolution of host-symbiont associations. We realized a survey of *Arsenophonus* diversity in whitefly species, especially the *Bemisia tabaci* species complex. This polyphagous pest, composed of genetic groups that differ in many ecological aspects, harbor several lineages of *Arsenophonus*. The evolutionary history of these bacteria at a fine host taxonomic level was studied in association to host geographical range and ecology.

- Samples were screened for *Arsenophonus* infection by amplification of the 23s *rRNA* gene. *B. tabaci* genetic groups were identified based on the *mtCOI* gene.
- *Arsenophonus* diversity was investigated on three genes, *ftsK*, *yaeT* and *FbaA*.
- Phylogenetic analyses were performed using maximum-likelihood (ML) and Bayesian inferences (BI) for the **concatenated data set**.
- Putative recombinant regions were searched using methods available in RDP3.

B1-47 (Burkina Faso)

B1-42 (Burkina Faso)

Recombinants

100/1

100/1

86/1

92/1

98/1

96/1

100/1

99/1

0.05

Burkina Faso
Togo
Burkina Faso
Togo
Burkina Faso
Burkina Faso
Réunion
Israel
France
Spain
Grande Comore
Tanzania
Madagascar
Réunion
Mayotte
Mauritius
Seychelles
Tanzania (Ms)
Réunion (Ms)
Réunion (*T. vaporariorum*)
Tanzania (Ms)
Madagascar (Ms)
Réunion

ASL + AnSL
Q3
ASL
Q2
Ms
T. vaporariorum + Ms
B. afer

- Among **152 individuals**, 19 allelic profiles and **6 phylogenetic groups** of *Arsenophonus* have been identified which **correlated with *B. tabaci* genetic groups** (Fig. 2).
- There is no evidence of intragenic recombination within the three genes but **intergenic recombination events** were detected within 2 individuals belonging to the ASL genetic group from Burkina Faso (Fig. 2).
- Phylogenetic analyses revealed the existence of **2 clades of *Arsenophonus***. One is composed of strains found in other Hemiptera and the other of strains found in Hymenoptera and Diptera (Fig. 3).



Phylogenetic tree showing relationships between *Bemisia tabaci* and related species. The tree is rooted at the bottom with *Hippobosca equina*. The main clade splits into two major groups. The left group contains *Bemisia tabaci* (Tanzania, Réunion), *T. vaporariorum* (Réunion, Seychelles, Réunion, Madagascar, Madagascar), and a clade of *B. afer* (Burkina Faso, Burkina Faso, Burkina Faso, Spain, Spain). The right group contains *Aphis spiraecola*, *Aphis gossypii*, *Cacopsylla alaterni*, and a clade of *B. afer* (Burkina Faso, Burkina Faso, Burkina Faso, Burkina Faso). Bootstrap values are shown at the nodes. Scale bar = 0.02.

Species and their locations/hosts are listed on the right side of the tree:

- Bemisia tabaci* (Tanzania)
- T. vaporariorum* (Réunion, Seychelles, Réunion, Madagascar, Madagascar)
- B. afer* (Burkina Faso, Burkina Faso, Burkina Faso, Spain, Spain)
- Aphis spiraecola*
- Aphis gossypii*
- Cacopsylla alaterni*
- B. afer* (Burkina Faso, Burkina Faso, Burkina Faso, Burkina Faso)
- Hippobosca equina*

*Duron, O., T. Wilkes, and G. Hurst. 2010. Interspecific transmission of a male-killing bacterium on an ecological timescale. *Ecology Letters* 13: 1139-1148.

- **High diversity** of *Arsenophonus* symbionts in *B. tabaci*.
- **Intergenic recombination** events suggested that *Arsenophonus* is not a strict clonal bacterium.
- Some strains were found in other Hemiptera and could be the ancestral clade in whiteflies while others were found in Hymenoptera and Diptera and may have been acquired more recently by whiteflies through **horizontal transfers**.

