# Preliminary results in the taxonomy of the cryptic group *Aphis frangulae/gossypii* obtained from mitochondrial DNA sequence

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#### **Abstract**

Phylogenetic relationships within members of the cryptic group *Aphis frangulae/gossypii* (Rhynchota Aphididae) were inferred from partial sequence of mitochondrial cytochrome C oxidase (COI). Sixteen postulated species, with several specimens per species, were included, representing part of recognized species inside the group. Results show that the monophyly of the *frangulae/gossypii* group is well supported. In the phylogenetic tree the two main clusters represented by *A. gossypii* and *A. frangulae* are clearly evident, each one associated with other more strictly allied species. Inside the *A. gossypii* (sensu stricto) group there is very little if any genetic difference within other postulated species, i.e. *A. sedi, A. catalpae, A. myopori*, and *A. capsellae*; the latter might represent therefore populations of the former species rather than separate specific taxa in the traditional sense.

**Key words:** Aphididae, *Aphis*, taxonomic relationships, molecular analyses, phylogeny.

#### Introduction

The genus *Aphis* L. is undoubtedly the largest worldwide taxon of viviparous aphids (Rhynchota Aphididae), being it represented by nearly 600 valid species (Remaudière and Remaudière, 1997). The genus includes several species which are major crop pests, as well as many others which live on a wide range of wild host plants of different families, mostly belonging to the Dycotiledones. The obvious consequence is the difficulty for a comprehensive taxonomy of the genus itself which, moreover, includes several cryptic groups to investigate whose molecular analyses have only recently been attempted (Coeur d'acier *et al.*, 2007).

One cryptic group is the Aphis frangulae/gossypii complex, to which a number of specific or subspecific taxa belong, characterized by reduced or absent (at least in apterae) dorsal body sclerifications, short antennal and crural hairs, sclerified medium-sized siphunculi, cauda usually a little paler than the latter and bearing a reduced number of hairs. To it belong a few well known cosmopolitan aphids, such as Aphis gossypii Glover (sensu stricto) and several other palaearctic members living on several host plants and particularly on Labiatae. The group or, at least, its main components have been extensively investigated, either biologically and morphologically by different authors (Thomas, 1968; Stroyan, 1984; Heie, 1996). Their results have always highlighted the difficulty of efficiently solving the taxonomic relationships between the different postulated species, some of which (i.e., gossypii, capsellae, lamiorum, etc.) have sometimes been considered at a subspecific level of Aphis frangulae Kaltenbach.

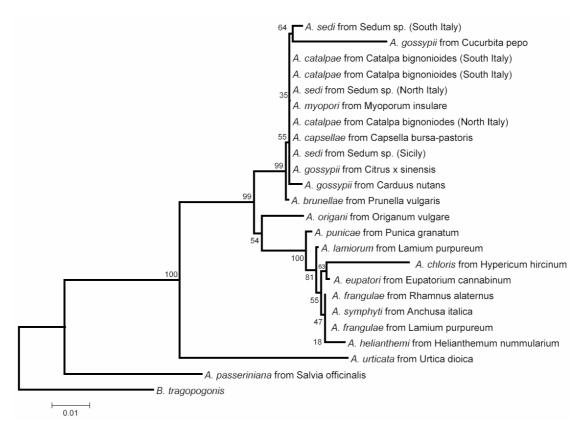
In order to contribute to the taxonomy of that aphidcomplex, an attempt has been developed on a first group of nearly twenty postulated species, by sequencing of mitochondrial cytochrome oxidase subunit I (COI).

## Materials and methods

All samples analyzed were collected in different regional territories in Italy. Genomic DNA was extracted by using Chelex-100 resine. Amplifications of 720 bp of COI were done using primers C1-J-2195 and TL2-N-3014 (Simon *et al.*, 1994), and PCR products were sent to a sequencing service (BMR Genomic sequencing service of Padova). *Brachycaudus tragopogonis* (Kaltenbach) was used as outgroup. After initial inspection of the sequences, average pairwise distances were calculated using Kimura's two parameter model as implemented in Mega3. Phylogenetic reconstructions were obtained by maximum-parsimony criterion employing PAUP\* 4.0b10 (Swofford, 2001).

### Results and discussion

The phylogenetic reconstruction of species or their populations examined to date is shown in figure 1. Here, it is possible to distinguish two main clusters: a clade with A. frangulae associated to other allied species, and a sister clade including A. gossypii and its comparatively more similar taxa. Consequently, the two aphids can be confidently considered as genetically separate taxa at a specific level. A sample of the former species, collected on Lamium (which can be regarded as one of the aphid's secondary hosts) shows no genetic distance with a sample of the same aphid on primary host. Quite interestingly, though difficult to explain at the moment, is the node containing A. gossypii, which includes a number of strictly allied taxa or their populations. Aphis brunellae Schouteden represents the sister group of this clade. The group undoubtedly needs to be more deeply investigated genetically to try to solve the relationship among the different components involved within the group itself. It is evident the strict genetic link (ge-



**Figure 1.** Phylogenetic reconstruction of *Aphis* spp. obtained from the analysis of COI. Numbers above branches of bootstrap replicates (10000) that recovered this branch.

netic distance range from 0 to 0.03) among samples postulated as *Aphis sedi* Kaltenbach (collected on *Sedum*), *Aphis catalpae* Mamontova (from *Catalpa*) and *Aphis myopori* Macchiati (from *Myoporum*). Among the samples examined, *Aphis capsellae* Kaltenbach – holocyclic on *Capsella* and traditionally considered as a full species or, alternatively, subspecies of *A. frangulae* or of *A. gossypii*, as quoted by different authors – stands closely related from the other postulated taxa of the group. They all differ very little, if any, from *A. gossypii* and it may be that they or part of them might represent populations of the latter species rather than separate specific taxa in the traditional sense.

# **Conclusions**

Results to date should be considered as preliminary, because investigation is still in progress, aiming to enlarge the number of species considered, their host plant association and to add further sequences of DNA fragments. However, molecular data shown here may represents a first step to improve the knowledge about relationships among taxa of *Aphis frangulae/gossypii* complex.

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