

# Reconstructing *Sordida* subcomplex (Triatominae, Triatoma) phylogeny across species distribution range



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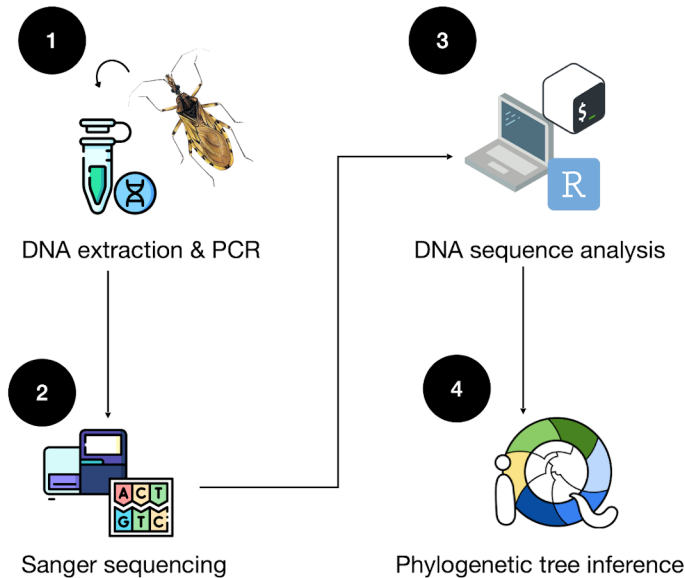
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The conformation of the *Sordida* subcomplex (Hemiptera, Reduviidae, Triatominae) has been a topic of prolonged debate, with diverse methodological approaches employed to discern its constituent species. Up to now, *Triatoma sordida*, *T. garciabesi* and *T. rosai* comprise part of this subcomplex. Distinguishing and identifying these three species poses significant challenges due to their pronounced morphological similarity, overlapping distributions, and presence of natural hybrids. This study aims to uncover the genetic diversity and geographic spread of these three species.

## Methods



1: DNA extraction was performed employing the conventional phenol-chloroform technique. PCR to isolate *cytochrome b* mitochondrial gene fragment was carried out as Gomez-Palacio et al. 2023.

2: Sanger sequencing and purification was performed in MACROGEN Inc. in both strands.

3: Sequence quality was evaluated with R package *sangeranalyseR* (Chao et al. 2021). Mafft (Katoh and Standley 2013) was used for alignment. Basic population statistics, haplotype determination and genetic distance were calculated with R packages *vegan* (<https://CRAN.R-project.org/package=vegan>), *ape*, *pegas* (Paradis 2004, 2010). Species delimitation was determined with GMYC using R packages *splits* (Ezard et al. 2009), and Hierarchical Bayesian Analysis of Population Structure (HierBAPS) (Cheng et al. 2013).

4: Phylogenetic trees were calculated under Bayesian inference (BI) and Maximum likelihood (ML). BI was implemented in MrBayes3.2.7 (Ronquist et al. 2012). ML was calculated using IQtree (Minh et al., 2020)

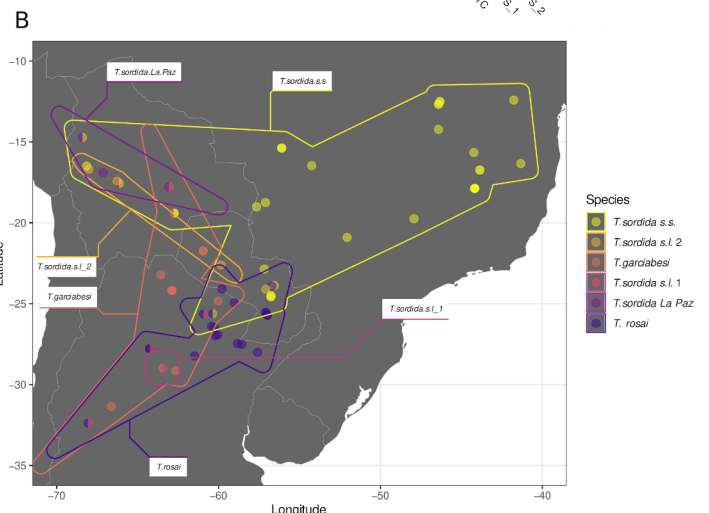
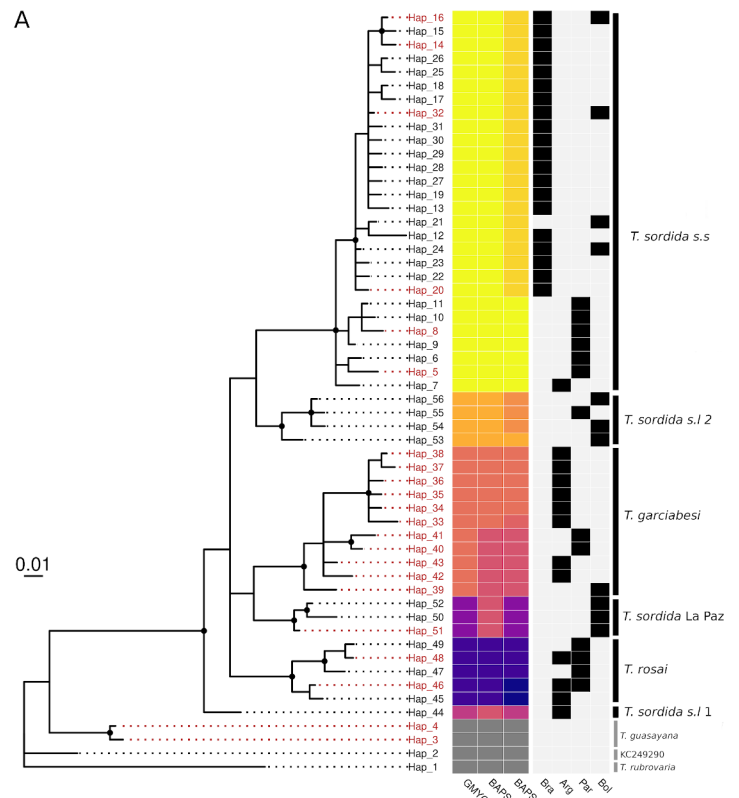
## Conclusions

Phylogenetic analyses suggest the presence of at least six putative species, rather than the three currently recognized. The present findings underscore the potency and significance of molecular analyses from natural populations for species identification and highlight the importance complementing morphology and molecular data when classifying Triatominae species. Whether vectorial importance is different for each of the proposed species is yet to be evaluated.

### References:

Chao et al. 2021, doi:10.1093/gbe/evab028; Cheng et al. 2013, doi: 10.1093/molbev/mst028  
 Ezard et al. 2009, <https://r-forge.r-project.org/projects/splits/>  
 Gomez-Palacio et al. 2023, doi: 10.1111/mve.12633; Katoh and Standley 2013, doi: 10.1093/molbev/mst010  
 Minh et al. 2020, doi: 10.1093/molbev/msaa015; Paradis 2004, doi: 10.1093/bioinformatics/btg412  
 Paradis 2010, doi: 10.1093/bioinformatics/btp696; Ronquist et al. 2012, doi: 10.1093/sysbio/sys029  
 The *T. sordida* drawing in Methods is from Costa et al. 2008: 10.13140/2.1.1578.9449

## Results



A: BI tree posterior probabilities support is depicted with black dots over the nodes when above 0.95. Colors on the GMYC column are defining the mitochondrial lineages and used in part B. The two columns on the right represent results from species delimitation algorithms. The country distribution is represented by black squares on the light gray columns. B: Map from the Southern Cone in South America, indicating the geographical distribution of six clades.