DNA barcoding: A rapid and accurate tool for identifying aphid (Hemiptera: Aphididae) species of economic importance in Kenya

G. Kinyanjui, F. M. Khamis, S. Mohamed, L. O. Ombura and S. Ekesi
International Centre of Insect Physiology and Ecology (*icipe*), Kenya
gkinyanjui@icipe.org

INTRODUCTION

- Aphids are among the most serious pests of economic importance throughout the world.
- Rapid and accurate identification of aphid species is crucial for effective pest management strategies (Miller and Foottit, 2009).
- Identification of aphids is done using morphological characters, but due to their small size, cryptic speciation, polymorphism, and reduction in morphological characters, identifying aphids can be difficult (Miller and Foottit, 2009).
- Availability of an accurate and rapid tool for identifying aphid species will facilitate quicker and effective implementation of pest management programmes, and enhance plant quarantine systems.

OBJECTIVE

- To characterise aphid species in Kenya using DNA barcoding and generate a reference DNA barcode library.

METHODS

- Aphid samples were collected from 13 counties across Kenya (Bungoma, Embu, Kajiado, Kiambu, Kwale, Laikipia, Makueni, Migori, Nairobi, Nakuru, Nyandarua, Nyeri, and Taita Taveta).
- DNA was extracted from 175 aphid samples using proteinase K buffer.
- The DNA barcoding region of the *COI* gene was PCR amplified using universal primers LCO1490/HCO2198 (Folmer *et al*., 1994), and sequenced.
- Sequence data were analysed using the following software: Chromas 2.1.1, BLAST, MUSCLE 3.8.31, Jalview 2.8.2, jModel test 2.1.7, RAxML 8.2.0, MEGA 6.0, GenAlEx 6.41, DnaSP 5.0, and Network 4.6.1.1.

CONCLUSION

- DNA barcoding is a quick and precise tool for identifying aphid species.
- It is effective in distinguishing morphologically inseparable species (*Aphis craccivora* and *A. fabae)* and cryptic species.
- The two clusters of *A. fabae* confirms the existence of subspecies that cannot be distinguished morphologically.
- DNA barcodes also provide a reliable means of identifying damaged specimens and immature stages of aphids.

IMPACT

- The study provides DNA barcodes of aphid species (175 sequences for 7 species) that will be a reference for identifying them in the Barcode of Life Database (BOLD).
- The study confirms DNA barcoding as an accurate and rapid tool for identifying species as well as genetic variability studies of aphid species, thus contributing to proper decision-making for their effective management, and for phytosanitary measures to be put in place in Kenya and beyond.

RESULTS

- Neighbour-joining tree (Fig. 1) clustered the species populations into 7 distinct clusters irrespective of their geographical locations, and revealed two sub-species of *Aphis fabae*.
- Genetic distances between species ranged from 0.052 (*Lipaphis pseudobrassicae* and *Brevicoryne brassicae)* to 0.098 (*Myzus persicae* and *Aphis craccivora*) (Table 1).
- Principal Component Analysis (Fig. 2) confirmed the results of the neighbour-joining tree, and the haplotype network further supported the results (Fig. 3).

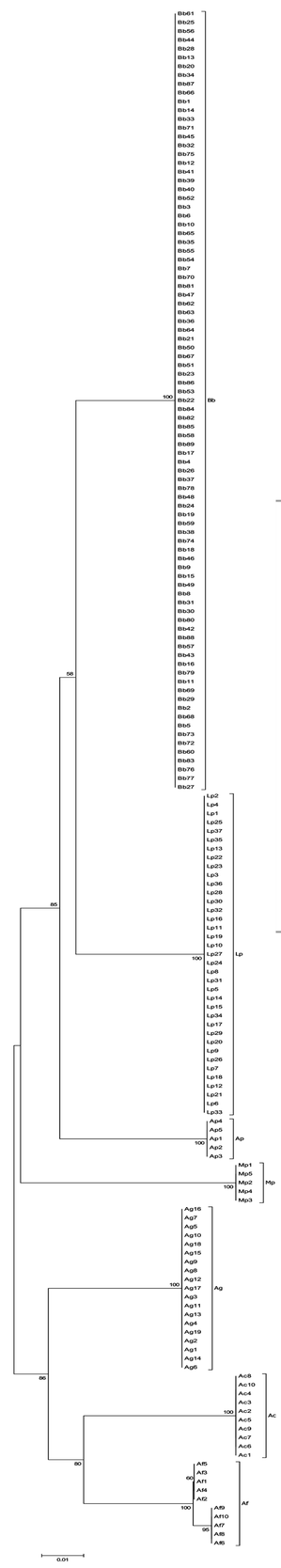

Fig. 1: Neighbour-joining tree of 7 aphid species

Table 1: Genetic distances between species

	Ag	Ac	Af	Bb	Lp	Ap	Mp
Ag	0						
Ac	0.077	0					
Af	0.061	0.062	0				
Bb	0.075	0.086	0.077	0			
Lp	0.074	0.088	0.085	**0.052**	0		
Ap	0.089	0.078	0.085	0.060	0.071	0	
Mp	0.089	**0.098**	0.088	0.083	0.085	0.089	0

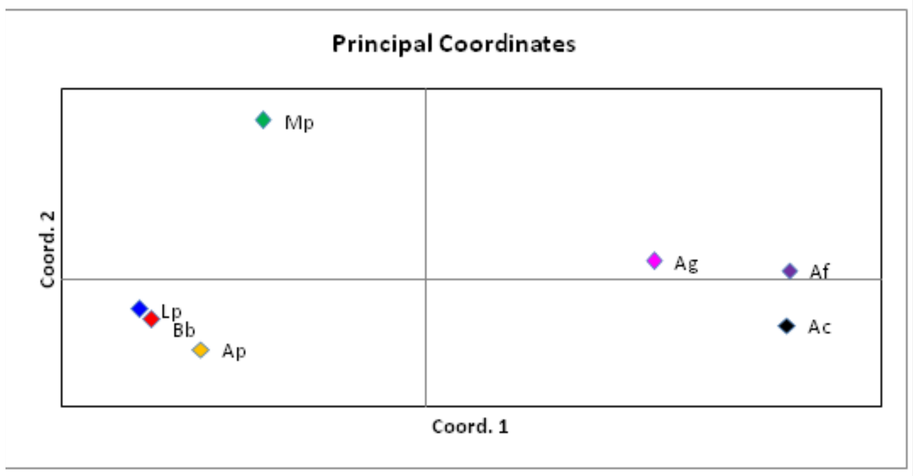

Fig. 2: Principal component plot showing aphid clusters separated by first and second principal components

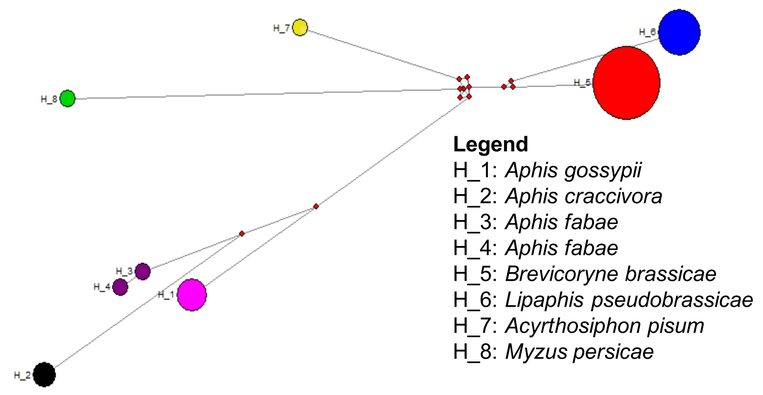

Fig. 3: Phylogenetic network showing evolutionary relationships among 7 aphid species

REFERENCES

Folmer O., Black M., Hoeh W., Lutz R. and Vrijenhoek R. (1994) DNA primers for amplification of mitochondrial *cytochrome c oxidase* subunit 1 from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* 3, 294–297.

Miller G.L. and Foottit R.G. (2009) The taxonomy of crop pests: The aphids, pp. 463-473. In *Insect Biodiversity: Science and Society* (edited by R.G. Foottit and P.H. Adler). Wiley-Blackwell, Oxford.

icipe
African Insect Science for Food and Health
International Centre of Insect Physiology and Ecology (*icipe*)
P.O. Box 30772-00100, Nairobi, Kenya
Tel: +254 (20) 8632000. E-mail: icipe@icipe.org

Sida