

# DNA barcoding and the effect of spatial distance on the genetic variation of African Noctuoidea (Insecta; Lepidoptera)

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## Introduction

The Noctuoidea constitute the most cosmopolitan and speciose lineage of Lepidoptera. Many noctuid species are notoriously difficult to identify using morphology. DNA barcoding allows to identify some noctuids, although the success of this technique has not been evaluated for the African noctuid representatives. It is also unclear how spatial distance relates to genetic divergence of COI in this group and how this may affect identification success.

## Aims

- (1) To test if DNA barcoding can be used to identify African noctuid species.
- (2) To test if spatial distance is correlated with intraspecific sequence divergence.
- (3) To assess how well the Barcode of Life Database Systems (BOLD) and GenBank perform in identifying African noctuids.

## Materials and Methods

One hundred and twenty three specimens of 66 noctuid species, caught at four localities in Tanzania (fig. 1), were identified using morphology and DNA barcoded. This dataset was supplemented with noctuid barcode sequences from BOLD so that the final dataset comprised 627 COI haplotypes of 115 species. Kimura-2-parameter distances were calculated and visualized in neighbour-joining trees. The spatial distance between observations was plotted against the genetic distances between observations for every species that occurred in five or more countries.

## Results and Discussion

One third of the species could not be unambiguously identified. Hence, DNA barcoding alone should not be relied upon to identify these African noctuids.

The low identification success rate could be due to the presence of cryptic species, but is also likely a result of mistakes in the molecular reference databases. For example, in the genus *Spodoptera* (fig. 2) two haplotypes of two species (*S. littoralis* and *S. umbraculata*) were grouped with a different reference species (*S. litura* and *S. pecten*, respectively). Considering the morphological similarity between these species, their geographical distribution and the grouping of the other haplotypes, it is likely that the reference material was erroneously identified.

Many pest species are well represented in BOLD and GenBank, and therefore provide reliable identifications. However, lesser-known noctuid species are far less presented and caution should be taken when using these barcode entries for identifications.

Intraspecific sequence divergence only increased with spatial distance in a few species. The influence of spatial distance was often very pronounced between continents, but almost absent within continents (fig. 3). This may indicate that specimens from different continents represent putative different species.

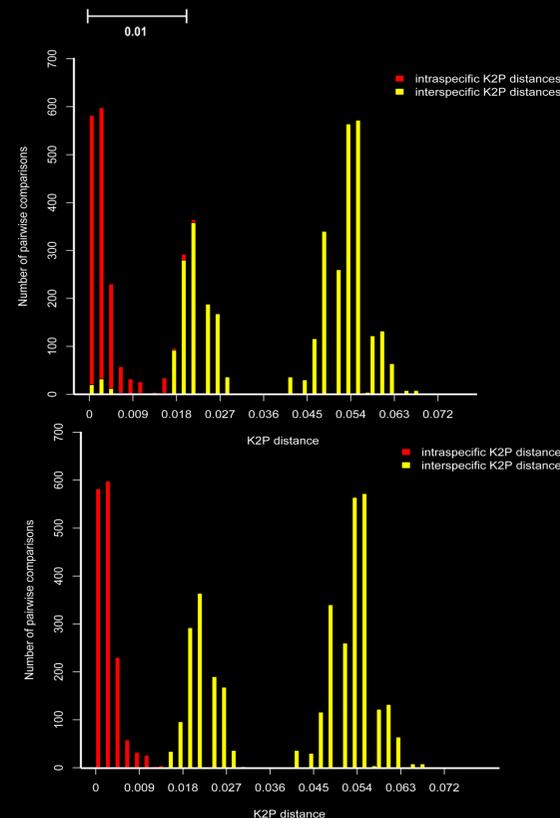


Fig. 2 (from top to bottom): Neighbour-joining tree of the genus *Spodoptera*, with bootstrap values higher than 60% and possible false identifications highlighted by blue arrows; distributions of intra- and interspecific distances with false identifications and after correction; and a specimen of *Spodoptera littoralis*.



Fig 1: sampling method and map with sampling localities.

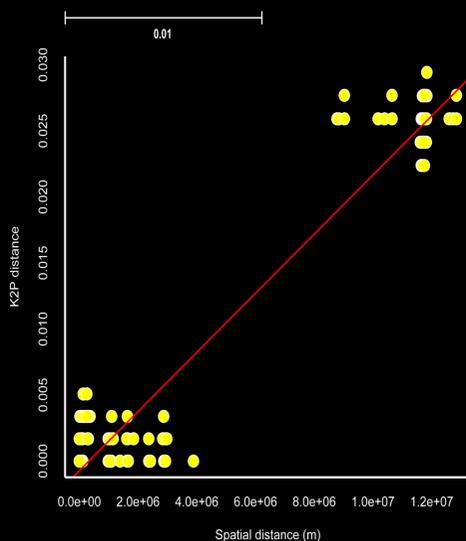
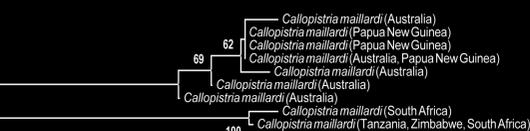


Fig. 3 (from top to bottom): Neighbour-joining tree of *Callopietria maillardi* with bootstrap values higher than 60%; graph showing the influence of spatial distance on the intraspecific genetic distance; and a specimen of *Callopietria maillardi*.