Molecular markers for Phlebotomus papatasi (Diptera: Psychodidae) and their usefulness for population genetic analysis

Article in Transactions of the Royal Society of Tropical Medicine and Hygiene · April 2009
DOI: 10.1016/j.trstmh.2009.02.011 · Source: PubMed

6 authors, including:

Omar Hamarsheh
Al-Quds University
41 PUBLICATIONS 382 CITATIONS
See profile

Amer Al-Jawabreh
LRU-Jericho
42 PUBLICATIONS 623 CITATIONS
See profile

Ziad Abdul Muhsen Abdeen
Al-Quds University
237 PUBLICATIONS 3,053 CITATIONS
See profile

Ahmad Amro
Al-Quds University
26 PUBLICATIONS 289 CITATIONS
See profile

Some of the authors of this publication are also working on these related projects:

Surface active germanium complexes View project

Leishmania project View project
MINI-REVIEW

Molecular markers for *Phlebotomus papatasi* (Diptera: Psychodidae) and their usefulness for population genetic analysis

Omar Hamarsheh a,*, Wolfgang Presber b, Amer Al-Jawabreh c, Ziad Abdeen d, Ahmad Amro b, d, Gabriele Schönian b

a Department of Biological Sciences, Al-Quds University, P.O. Box 51000, Jerusalem, Palestine
b Institut für Mikrobiologie und Hygiene, Humboldt Universität, Charité Universitätsmedizin Berlin, Dorotheenstraße 96, 10117 Berlin, Germany
c Islah Medical Laboratory, Islah Charitable Social Society, Jericho, Palestine
d Laboratory of Leishmaniasis, Faculty of Medicine, Al-Quds University, Jerusalem, Palestine

KEYWORDS
Phlebotomus papatasi; Sand flies; Genetic markers; Population genetics; rDNA; Phylogeny

Summary
Three molecular typing tools: multilocus microsatellite typing, cytochrome *b* sequence analysis and internal transcribed spacer 2 (ITS2) sequence analysis, were evaluated for their usefulness in inferring the population structure of *Phlebotomus papatasi* sand flies. ITS2 sequence analysis did not prove suitable for inferring phylogenetic and population genetic relationships across *P. papatasi* sand flies. Microsatellite markers showed high resolution in differentiating globally distributed *P. papatasi* populations, whereas cytochrome *b* sequence analysis provided insight into the relationships between closely related populations from the Mediterranean. Population structure, differentiation and demographic history among *P. papatasi* are important for understanding patterns of dispersal in this species and for planning appropriate control measures.

© 2009 Royal Society of Tropical Medicine and Hygiene. Published by Elsevier Ltd. All rights reserved.

*Phlebotomus papatasi* is the main vector of *Leishmania major*, which is one of the causative agents of Old World zoonotic cutaneous leishmaniasis. It has a large geographical distribution, ranging from Morocco to the Indian subcontinent and from southern Europe to central and eastern Africa. The geographical range of *P. papatasi* includes varying climatic and ecological discontinuities. Therefore, knowledge of the genetic relationships and biogeography among *P. papatasi* populations would enable better understanding of the current geographic distribution and improve the design of appropriate control measures.

Population genetic studies should rely on markers that have appropriate discriminatory power, depending on the question being asked. Markers that are neutral, not under selective pressure and are co-dominant are highly significant in population studies. Multilocus enzyme electrophoresis and several approaches based on DNA sequence comparisons fulfil these requirements, for example, the sequencing...
of mitochondrial genes such as cytochrome b (cyt b) and of different housekeeping genes by multilocus sequence typing, the typing of single nucleotide polymorphisms and the exploration of variation in microsatellite sequences with multilocus microsatellite typing (MLMT).

Genetic studies on P. papatasi sand flies are still very scarce. Various cyt b haplotypes have been identified in P. papatasi populations from different Mediterranean areas. Depaquit studied the relationships between different sand fly species by analysing rDNA sequences. This mini-review highlights recent findings obtained by analyzing large numbers of P. papatasi individuals originating from widely separated populations, allowing the re-assessment and re-evaluation of well-known genetic typing techniques as well as the newly developed multilocus microsatellite typing tool.

The sequence typing of rDNA spacers has many advantages, such as enormous variability due to high mutation rates and high sensitivity due to their multi-copy character. Analysis of internal transcribed spacer 2 (ITS2) sequences revealed significant intra-specific variation for the P. papatasi individuals studied and even intra-individual variation in the multiple copies of the ribosomal spacer. It seemed that variant copies of ITS2 within individual sand flies often differed as much as between flies from different localities. This resulted in poor geographical structuring. Thus, ITS2 sequences were found to be too variable to resolve the genetic relationships between different populations of P. papatasi. Therefore, ITS2 does not represent a suitable marker for inferring phylogeny and population genetics in P. papatasi sand flies.

MLMT and comparisons of mitochondrial cyt b sequences have proven useful for the detection of intra-species variation in P. papatasi. Consistently significant levels of genetic differentiation have been observed using both tools. MLMT revealed the existence of population structuring and sub-structuring and allowed globally isolated populations to be distinguished. The comparison of cyt b gene sequences provided important information concerning P. papatasi populations and their demographic history. The technique was useful in resolving the genetic relationships of closely related populations of P. papatasi; five populations were distinguished in the Mediterranean area and the Middle East, and two populations were found to co-exist in Palestinian and Israeli foci. There are many reasons why cyt b is often used for phylogenetic analysis in sand flies. The high discriminatory power of the cyt b marker is based on the existence of discrete character classes (i.e. the three codon positions) that exhibit mutation rates reliable for phylogenetic analysis, plus the fact that the gene is maternally inherited and, thus, free of recombination. Furthermore, the gene evolves slowly in terms of non-synonymous substitutions, but the rate of silent mutations is relatively high. Sequence variation in cyt b was due only to single point mutations in all P. papatasi individuals studied.

The slow mutation rate and absence of recombination of the cyt b marker seems to make it better suited than microsatellites for differentiating closely related populations and distinguishing populations in close geographical proximity. Using MLMT in conjunction with comparison of cyt b sequences is highly recommended to provide dual insights into the population structure of P. papatasi.

Using more microsatellite markers could be promising for better resolution of closely related populations. Sequencing the P. papatasi genome is of great significance to map the whole genome for the existence of potential markers on both population and specific levels.

Funding: We are grateful to Deutscher Akademischer Austausch Dienst (DAAD) for providing partial financial support.

Conflicts of interest: None declared.

Ethical approval: Not required.

References


