Specification of blood meals ingested by female sand flies caught in Palestinian foci and identification of their concomitant leishmanial infections

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Visceral (VL) and cutaneous leishmaniasis (CL) in West Bank
Mission

- Diagnosis
- Disease control
- Diversity
- Epidemiology
This study investigated the Palestinian sand fly fauna, their infection rates and feeding preference determined.
Study area and sample collection
PCR hybridization approach for the identification of the sources of blood imbibed by female sand flies.

This is based on the PCR amplification of vertebrate mitochondrial cytochrome b gene sequences combined with reverse dot blot hybridization as described by Boakye et al., 1999, and Abbasi et al., 2009.
Multiplex PCR amplifying leishmanial kDNA gene sequences (120 bp) and the 12 SL RNA sand fly housekeeping gene (165 bp) in representative sand flies from different districts of the West Bank.
Digestion profile of the ribosomal RNA internal transcribed spacer 1 (ITS1) gene of leishmanial parasites amplified from parasites within sand flies from Palestinian districts, using the restriction enzyme HaeII

Lane 1, a Ph. sergenti (45A: a representative) sand fly PCR positive for Leishmania tropica collected in Jenin District; lane 2, reference strain of L. major MHOM/SU/1973/5ASKH; lane 3, reference strain of L. tropica MHOM/1998/LRC-747; lane 4, reference strain of L. infantum MHOM/TN/1980/IPT1; lanes 5: AQU lab internal marker for 100, 200 and 290bp & 6, 100 bp DNA ladde
The number of sand flies collected, their species diversity, relative abundance, infection rates and positivity rate for the presence of different species of Leishmania.

| Species                     | Total Female SF (%) | Sand flies positive by kDNA PCR (%) | Sand flies positive by ITS1 PCR (%)* | Infected sand flies (%)/Leishmania species**/location sites |
|-----------------------------|---------------------|------------------------------------|-------------------------------------|----------------------------------------------------------|
| Ph. alexandri              | 2 (0.3)             | 1 (50)                             |                                     |                                                          |
| Ph. jacusieli               | 1 (0.1)             | 0 (0)                              |                                     |                                                          |
| Ph. kazeruni                | 2 (0.3)             | 0 (0)                              |                                     |                                                          |
| Ph. major neglectus         | 10 (1.4)            | 3 (30)                             | 2 (20); Blood meal (n=2: human)    |                                                          |
| Ph. major syriacus          | 32 (4.6)            | 0 (0)                              |                                     |                                                          |
| Ph. canaaniticus            | 18 (2.6)            | 1 (5.6)                            |                                     |                                                          |
| Ph. papatasi                | 233 (33.7)          | 19 (8.2)                           | 13 (5.58); Blood meal (n=1: human & avian; n=4: human; n=8: NI) | 2 (0.4%)/ L. major² Blood meal (n=2: human) |
| Ph. perfiliei transcaucasicus| 27 (3.9)           | 5 (18.5)                           | 2 (7.4); Blood meal (n=1: human; n=1: NI) |                                                          |
| Ph. saltiae                 | 2 (0.3)             | 1 (50)                             |                                     |                                                          |
| Ph. sergenti                | 145 (21)            | 47 (32.4)                          | 19 (13.1); Blood meal (n=11: human; n=1: goat; n=1: human & cow; n=1: human, cow & goat; n=5: NI) | 6 (4.1%)/ L. tropica² Blood meal (n=1: human & cow; n=2: human; n=1: avian & cow) |
| Ph. tobbi                   | 86 (12.6)           | 26 (30.2)                          | 3 (3.4); Blood meal (n=2: human; n=1: NI) |                                                          |
| Total Phlebotomus           | 558 (80.8)          | 103 (18.4)                         | 39 (6.9)                            |                                                          |
| Se. africana                | 24 (3.5)            | 4 (16.6)                           |                                     |                                                          |
| Se. antennata               | 3 (0.4)             | 1 (33)                             |                                     |                                                          |
| Se. christophersi           | 6 (0.9)             | 1 (16.6)                           |                                     |                                                          |
| Se. dentata                 | 46 (6.7)            | 6 (13)                             |                                     |                                                          |
| Se. fallax                  | 9 (1.3)             | 0 (0)                              |                                     |                                                          |
| Se. taizi                   | 2 (0.3)             | 1 (50)                             |                                     |                                                          |
| Se. theodori                | 18 (2.6)            | 3 (16.7)                           | 1 (5.5); Blood meal (n=1: NI)       |                                                          |
| Se. tiberiadis              | 13 (1.9)            | 3 (23)                             |                                     |                                                          |
| Se. (Neophlebotomus) sp.    | 12 (1.7)            | 6 (50)                             | 4 (33); Blood meal (n=3: human; n=1: NI) | 2 (0.3%) L. tropica² Blood meal (n=1: human; n=1: NI) |
| Total Sergentomyia          | 133 (19.2)          | 25 (18.8)                          | 5 (3.8)                             |                                                          |
| Total                       | 691 (100)           | 128 (18.5%)                        | 44 (6.4%)                           | 10                                                       |
Sand flies collected inside, outside animal sheds and the rock crevasses inhabited by hyraxes in the vicinity of houses

| Species                   | Animal sheds | Hyrax burrows and caves in the vicinity of houses | Indoor | Outdoor | Total |
|---------------------------|--------------|---------------------------------------------------|--------|---------|-------|
| Ph. major syriacus        | 3            | 19                                                | 1      | 9       | 32    |
| Ph. mascittii canaaniticus| 5            | 12                                                | 1      | 0       | 18    |
| Ph. papatasi              | 23           | 12                                                | 191    | 7       | 233   |
| Ph. perfiliewi transcaucasicus | 0     | 6                                                 | 6      | 15      | 27    |
| Ph. sergenti              | 7            | 105                                               | 19     | 14      | 145   |
| Ph. tobbi                 | 24           | 33                                                | 6      | 23      | 86    |
| Se. africana              | 2            | 1                                                 | 21     | 24      |       |
| Se. dentata               | 0            | 28                                                | 7      | 11      | 46    |
| Se. (Neophlebotomus) sp.  | 0            | 12                                                | 0      | 12      |       |
| other Phlebotomus         | 1            | 15                                                | 0      | 1       | 17    |
| other Sergentomyia        | 1            | 27                                                | 8      | 15      | 51    |
| Total                     | 66           | 269                                               | 240    | 116     | 691   |
# Blood fed Sand flies

| Blood fed Sand flies       | Blood meal sources                                                                 |
|----------------------------|-----------------------------------------------------------------------------------|
| **Ph. papatasi (n=90)**    | human (77), human and cow (7), human and avian (2), avian and cow (1), human and goat (1), human, avian and cow (1), human, hyrax, cow & avian (1). |
| **Ph. sergenti (n=104)**   | human (72), human and cow (17), human, cow and goat (5), human and hyrax (4), human and rat (2) hyrax and rat (1), human, cow and avian (1), human and cat (1), goat (1). |
| **Ph. tobbi (n=31)**       | human (22), human, cow and goat (5), human and cow (3), human and avian (1).       |
| **Ph. major syriacus (n=18)** | human (16), human and cow (1), human, cow and goat (1).                           |
| **Other Phlebotomus (n=26)** |                                                                                   |
|                            | human (20), human and cow (3), human, cow and goat (2), human and donkey (1)      |
| **Sergentomyia (n=32)**    |                                                                                   |
|                            | human (28), human and cow (3), human, cow and goat (1)                           |
| **Total**                  | 301                                                                               |
• Twenty phlebotomine species were shown to be circulating in the study area. The most abundant species was Ph. papatasi (33.7%), followed by Ph. sergenti (21%).
• Infection by parasites of L. tropica was confirmed in 6.4%, using an ITS1-PCR.
• RDB blood meal assay proved efficient in analyzing blood meals imbibed by phlebotomine sand flies and those of the genus Sergetomyia in identifying the animal hosts from which the blood meals came.
• Meals of human blood were the most abundant (78.1%) and those of hyrax blood were identified in the cases of six sand flies, five Ph. sergenti and one Ph. papatasi
Thank you