Natural hosts of the larvae of *Nuttalliella* sp. (*N. namaqua*?) (Acari: Nuttalliellidae)

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The first collection of unengorged and fully engorged larvae of *Nuttalliella* sp. (*N. namaqua*?) from the murid rodents *Micaelamys namaquensis*, *Aethomys chrysophilus* and *Acomys spinosissimus* in Limpopo Province and from *M. namaquensis* in the Northern Cape Province, South Africa, is documented. A total of nine larvae were collected from two *M. namaquensis* in the Soutpansberg mountain range in the Limpopo Province during April 2009. During the last week of September 2011, 221 larvae were collected from rodents at the same locality and 10 of 48 *M. namaquensis*, 6 of 12 *Ae. chrysophilus* and 3 of 14 *Ac. spinosissimus* were infested. One of the *M. namaquensis* harboured 53 larvae. Five larvae were collected from two *M. namaquensis* in the Northern Cape Province. Total genomic DNA was extracted from two larvae and a region of the 18S rRNA gene was sequenced for these. BLASTn searches revealed similarity between these specimens and the *Nuttalliella* sequences published on GenBank.

Communication

The collection of larvae, nymphs and females of *Nuttalliella namaqua* during 2011, after many years in which none had been detected, has revived interest in the morphology, hosts and biology of this unusual tick (Mans *et al*. 2011). Nymphs and females from this collection have successfully been fed on laboratory-infested lizards (Mans *et al*. 2011).

Various papers on the role of murid rodents as hosts of immature ixodid ticks in South Africa have recently been published (Matthee *et al*. 2007; Matthee *et al*. 2010; Petney *et al*. 2004). The present communication records their role as hosts of the larvae of *Nuttalliella* sp. (*N. namaqua*?). Two studies were conducted in Goro Game Reserve (22°58′S, 29°25′E) located in the Soutpansberg mountain range, Limpopo Province, South Africa. Both studies focused on various aspects of the biology of rock elephant shrews (*Elephantulus myurus*), and included collecting ticks from these small mammals. Animals were trapped with baited Sherman live traps set overnight on rocky outcrops. In addition to the elephant shrews, a number of murid rodents were trapped and also examined for ticks. Animals were collected during April and May 2009, and during the last week of September 2011. The animals were collected under permit number CPM-333-00002 issued by the Department of Environmental Affairs, Limpopo Province, South Africa, and all procedures were approved by the Animal Ethics Committee of the University of Pretoria (EC028-07, V052-11).

A third study was conducted during 2009 on the farm Plaatfontein (31°02′S, 23°46′E) in the eastern region of the Northern Cape Province. It focused on collecting biological data and also ticks from Southern African hedgehogs, (*Atelerix frontalis)* (Horak *et al*. 2011), under permit FLORA 036/2009 issued by the Northern Cape Department of Tourism, Environment and Conservation. However, a number of murid rodents were also trapped and examined for ticks.

After removal from the traps the body of each animal was searched for ticks, and particular attention was given to the ear margins, legs and the base of the tail, where ticks aggregated. The rest of the body was searched by back-combing the fur. All ticks were removed using fine-tipped forceps and placed in 70% ethanol, after which the host animals were released.

In the first study, five *Nuttalliella* sp. larvae were collected during April 2009 from a Namaqua rock mouse (*Micaelamys namaquensis*) and four from another. Two of the nine larvae were engorged. These are the first recorded collections of the larvae of this tick from host animals. Not one of the 58 rock elephant shrews, or remaining 141 rock mice or 8 spiny mice (*Acomys spinosissimus*) examined at the same time was infested. In the second study, 221 unengorged and engorged *Nuttalliella* sp. larvae were collected. Of these, 154 larvae were recovered from 10 of 48 *M. namaquensis* examined, 58 from 6 of 12 red veld rats (*Aethomys chrysophilus*), and 9 larvae from 3 of 14 *Ac. spinosissimus* (Table 1). One of the *M. namaquensis* harboured a total of 53 larvae,

whilst the comparatively large number of larvae collected from the 6 *Ae. chrysophilus* and the fact that 50% of the 12 examined were infested could imply that they are preferred hosts in Limpopo Province. Alternatively, but not mutually exclusive, this may be attributed to the greater body size of the *Ae. chrysophilus* (72.7 g ± 3.3, *n* = 12) compared to the *M. namaquensis* (39.0 g ± 0.9, *n* = 48). None of the 8 rock elephant shrews captured at the same time was infested. In the third study a total of five unengorged larvae were collected from two of an undisclosed number of *M. namaquensis* in the Northern Cape Province. Not one of the 12 hedgehogs examined at the same locality was infested.

Because the larvae of *N. namaqua* have not previously been collected from natural hosts, nor been described, we relied on morphological features resembling those described for the females to arrive at a diagnosis. As an adjunct to identification of the larvae on taxonomic features, total genomic DNA from two larvae collected from *M. namaquensis* was extracted using the Qiagen, DNeasy® Blood and Tissue kit. Polymerase chain reaction and sequencing was performed on the 18S rRNA gene fragment using conserved universal primers. Amplifications were performed following standard PCR protocols and cycle-sequencing reactions were performed using BigDye Chemistry. Products were analysed on an automated sequencer (ABI 3730 XL DNA Analyzer, Applied Biosystems).

A 466 base-pair region of the 18S rRNA gene was obtained for both larvae, and these sequences were aligned to the *N. namaqua* sequence on GenBank (Mans *et al*. 2011; JF751071.1). The region sequenced corresponds to positions 1093–1571 of the published sequence. The sequences generated for the two larvae in our study were identical. BLASTn searches on GenBank revealed 99.58% similarity (476/478) between the newly sampled individuals (GenBank Access Number JQ424828) and the 18S rRNA *Nuttalliella* sequence published on GenBank. Further support for the authenticity of our identification of the larvae was obtained by doing a GenBank BLASTn search against the entire database. The next closest alternative match to *Nuttalliella* would be the genus *Ixodes*, but in this instance the level of differentiation is more than 4% (614/640 identities between the larval sequences and that of *Ixodes persulcatus*: AY274888.1).

Conclusion

The larvae of a nuttalliellid tick, *Nuttalliella* sp. (*N. namaqua*?) can now be added to the immature stages of a large variety of ixodid tick species, which by preference feed on murid rodents.

TABLE 1: Larvae of *Nuttalliella* sp. (*N. namaqua*?) collected from small mammals in Limpopo Province, South Africa.

Hosts	*Micaelamys namaquensis* (*n* = 48)	*Elephantulus myurus* (*n* = 8)	*Aethomys chrysophilus* (*n* = 12)	*Acomys spinosissimus* (*n* = 14)
% infested	20.8	0	50.0	21.4
Total larvae	154	0	58	9
Mean ± SD	3.21 ± 10.089	0	4.83 ± 7.998	0.64 ± 1.865

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Competing interests

The authors declare that they have no financial or personal relationship(s) which may have inappropriately influenced them in writing this paper.

Authors' contributions

I.G.H. (University of Pretoria) identified the ticks with the assistance of D.A.A. (Georgia Southern University), and also compiled the manuscript. H.L. (University of Pretoria) and K.M. (University of Pretoria) collected the ticks and did the statistical work, and C.A.M. (Stellenbosch University) was responsible for the molecular analysis.

References

Horak, I.G., Welman, S., Hallam, S.L., Lutermann, H. & Mzilikazi, N., 2011, 'Ticks of four-toed elephant shrews and Southern African hedgehogs', *Onderstepoort Journal of Veterinary Research* 78(1), Art. #243, 3 pages. http://dx.doi.org/10.4102/ojvr.v78i1.243

Mans, B.J., De Klerk, D., Pienaar, R. & Latif, A.A., 2011, '*Nuttalliella namaqua*: a living fossil and closest relative to the ancestral tick lineage: implications for the evolution of blood-feeding in ticks', *PLoS ONE* 6(8): e23675. http://dx.doi.org/10.1371/journal pone.0023675

Matthee, S., Horak, I.G., Beacournu, J.C., Durden, L.A., Ueckermann, E.A. & McGeoch, M.A., 2007, 'Epifaunistic arthropod parasites of the four-striped mouse, *Rhabdomys pumilio*, in the Western Cape Province, South Africa', *Journal of Parasitology* 93, 47–59. http://dx.doi.org/10.1645/GE-819R2.1, PMid:17436941.

Matthee, S., Horak, I.G., Van der Mescht, L., Ueckermann, E.A. & Radloff, F.G.T., 2010, 'Ectoparasite diversity on rodents at De Hoop Nature Reserve, Western Cape Province', *African Zoology* 45, 213–224. http://dx.doi.org/10.3377/004.045.0222

Petney, T.N., Horak, I.G., Howell, D.J. & Meyer, S., 2004, 'Striped mice, *Rhabdomys pumilio*, and other murid rodents as hosts for immature ixodid ticks', *Onderstepoort Journal of Veterinary Research* 71, 313–318. PMid:15732458.