Supplement Methods

Rodent trapping and flea and blood sampling

Over a period of two to three consecutive nights, we trapped rodents in each of ten 1-ha plots located in the Western Negev, Israel (Hevel Shalom, 31° 10' N). The minimum distance between plots was 40 m. In each plot, we uniformly placed 40 live Sherman traps (H.B. Sherman, Tallahasse, FL, USA) in 4 columns and 10 rows per plot. We tagged each captured individual and determined its species, sex, weight, age group (juveniles <18 g; adults >18 g, following Hawlena et al 2006) and, in female individuals, the reproductive status (pregnancy or lactation versus non-reproductive adult).

Within each of the two sampling seasons (i.e., spring and summer), we collected fleas from rodent individuals only following their initial capture to avoid pseudo-replication. We gently held the rodent above a plastic can, blew its fur, and collected and counted jump-off fleas until no fleas were detected on the host body (procedure similar to that in Hawlena et al (2006)). We stored the fleas in 70% ethanol at -20 °C until species identification and DNA extractions.

In addition, under local anesthesia (Localin, Fischer Pharmaceutical Labs, Tel Aviv, Israel), we drew 100-200 µl of blood from the retro-orbital sinus of each rodent with sterile capillaries immersed in 0.15% EDTA, and stored the blood in EDTA blood-collection tubes at -20 °C until DNA extraction. Due to the sensitivity of the eyes, we could not sterilize the local area before drawing the blood. Therefore, to minimize contamination, we performed the bleeding process as quickly as possible. In addition, we always changed capillaries when we had to stab the retro-orbital sinus of a given individual more than once. Such efforts cannot completely prevent surface contamination; however, since all host individuals were subjected to exactly the same sampling procedure, possible surface contaminants should have only increased the number of bacterial phylotypes that are not associated with the flea vectors and

increased the variance among bacterial communities in the blood of different host individuals but not across seasons. At the end of the procedure, we released the individual at the site of capture.

Statistical analyses

Quantification of bacterial phylotype composition: We performed a non-parametric multivariate multiple regression analysis with a stepwise forward-selection procedure to analyze the effects of host characteristics (individual identity, age and sex), sample type (female flea, male flea and rodent blood), sampling season (spring or summer), and second (host age × sampling season, and sample type × host sex) and third (host age × sampling season × sample type, and host age × sampling season × host sex) interaction terms of these variables on bacterial phylotype composition. These analyses were performed by using DISTLM software (McArdle and Anderson 2001). We assessed bacterial community composition with the Bray-Curtis dissimilarity index (Magurran 1988) after a fourth root transformation. We determined the overall contribution of individual phylotypes within a community to the observed differences by using SIMPER, an analysis tool within the PRIMER-E software (Lutton, UK; Clarke and Gorley 2006), where only bacteria phylotypes that contributed consistently to the distinction between communities (i.e., when the ratio between the average and the standard deviation of dissimilarity between two phylotypes was greater than one) were regarded as good discriminating phylotypes (Clarke 1993). To visualize patterns in multivariate data, we used an unconstrained non-metric multi-dimensional scaling (MDS) plot, based on Bray-Curtis similarities on the fourth root transformed assemblage data (by using PRIMER-E).

Model selection approach: We compared models by using model probabilities (w_i, where *i*

corresponds to a specific model), which give a measure of the plausibility (on a scale of 0 to 1) that a particular model is indeed the best model (Burnham and Anderson 2002). We calculated the w_i values based on corrected quasi-independent model criteria (QICc). We used an unstructured correlation matrix for all GEE models since the correlation matrices could not be distinguished based on the QIC values, and since this matrix demands no specific assumption for the data structure.

Controlling for the effect of the total number of sequences: Phylotype composition, richness and diversity may have potentially been affected by the total number of sequences, as the total number of sequences per sample ranged between 138 and 1173 sequences with a mode and a median of 614. Therefore, for the composition and richness analyses, we normalized the number of sequences in each sample by randomly selecting 138 sequences from each sample 1000 times (re-sampling with replacement) and used the average number of sequences detected in each phylotype-sample combination in the subsequent analyses. For the diversity analyses, we employed Fisher's alpha diversity index (Fisher et al 1943), a reliable index of species diversity that is independent of sample size (Hubbell 2001).

Estimation of community stability of bacteria: To characterize the stability of bacterial communities in an individual host (as assessed through the two repeating samples of the rodent blood and of the female and male fleas), we calculated the fraction of phylotypes that were shared between the first and second samples (samples A and B, respectively) in each individual by using the Jaccard index (Equation 1; Jaccard 1912).

$$(1)\ \frac{Sample\ A \cap Sample\ B}{Sample\ A \cup Sample\ B}$$

We then averaged the indices of the three sample types for each host individual, yielding its 'intra-individual stability'. Then, for each individual rodent, we compared the 'intra-

individual stability' to the 'inter-individual stability', calculated as the mean Jaccard index of all possible same-sample type pairs of the specific host individual and its conspecifics. We evaluated the effect of the source of variation (within versus between host individuals), sample type and their interaction (independent variable) on the Jaccard index (dependent variable) by a GEE model and compared each model to an intercept-only model by using the model selection approach.

References

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