

The role of feral pigeons as potential disease vectors in dairy farms: Association between movement patterns, pathogen presence and microbiome diversity

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The increasing prevalence of zoonotic diseases and the growing global trend of "One Health" highlights the importance of linking vector behaviors with the pathogens they carry. Pigeons are common synanthropic species occupying urban and agricultural sites worldwide and are also a known vector of various human, poultry, and livestock-relevant pathogens (e.g. *Campylobacter*, *Salmonella*). Yet, despite their relevance for zoonotic diseases, little is known about their movement and microbiome (or pathogen) diversity. Because animal movement can both determine pathogens' spread (e.g., range and rate of contacts and transmissions) and be influenced by their load (e.g. sickness), the ecology of disease and host (i.e. pigeons) movement are inherently linked. Understanding this feedback between host behavior and their pathogenic or non-pathogenic microbes (namely microbiome) can facilitate prediction of wildlife disease outbreaks, and their spillovers to livestock and humans especially for pigeons that are found in close proximity to humans and dairy farms.

Here we aim to quantify movements of pigeons living in dairy farms, their microbiome diversity, and prevalence of pathogens, as well as their co-variation, affecting the dynamic of potential pathogen spread. Over the past three years we captured 328 pigeons around three dairy farms in central Israel (Maale Hahmisha, Mevo horon and Mikver Israel), collected pathogen samples from each, and fitted 44 pigeons with GPS tracking devices to monitor their movement. Using a combination of Next Generation Sequencing (Anicon and Base2Bio bioinformatics), rtPCR, and bacterial plate methods we assessed pathogen presence and microbiome composition. Using this datasets we attempted to answer questions about pigeon's role as a (zoonotic as well as animal) disease vector, and how movement at both the population and individual levels affect the microbial profile and potential to spread pathogens.

We have found that pigeon populations in central Israel carry poultry, cattle, and human-relevant pathogens, with some - like *Campylobacter* and Newcastle Disease Virus - at relatively high prevalence. In contrast to our expectations, no *Salmonella* presence was detected. By tracking pigeons' daily movement patterns, we found that individuals roost almost entirely in urban settlements (thus emphasizing the risk of zoonotic transmission), with regular visits to dairy farms and various other agricultural sites. Pigeons in the most urban site moved shorter distances than the other two sites and carried a larger diversity of bacteria. Individuals within each site also differed in their movement patterns, with a positive association between exploration (namely the number of stops at unique locations, i.e., sampling the environment more) and the diversity of microbiome bacteria. According to our investigation of pigeon microbial profiles, we found that microbiota diversity differed between capture locations. Alpha-diversity is also affected by individuals' exploration, where pigeons that make more daily stops (explorers) have a higher diversity of bacteria.

We may conclude that pigeons directly connect urban and agricultural land uses, potentially serving as transmission vectors among wildlife, domestic animals, and humans. Given their large numbers, high proximity to both cattle and humans, and diversity of known pathogens these risks should not be taken lightly. A side tracking effort also shows that regular hunting effort have almost no influence on the behavior of non-targeted pigeons, pointing out the futility of this intervention in preventing these interactions. Furthermore, the significant co-variation of individual movement and microbial diversity suggests that the feedback between host movement and pathogens load can shed light on transmission dynamics, highlighting the relevance of wildlife movement analyses for disease ecology and One Health. We recommend establishing a constant effort monitoring of disease prevalence in feral pigeon populations.

Submitted manuscripts (as of Jan 2024 not yet published beyond initial archive)

- Crafton M, Cahani S, Lublin A, Rauer L, Spiegel O. Association between movement patterns, microbiome diversity, and potential pathogen presence in free-ranging feral pigeons foraging in dairy farms. bioRxiv. 2023; Available from: <https://www.biorxiv.org/content/10.1101/2023.10.11.561861v1>
- Lazebnik T, Spiegel O. Individual Variation Affects Outbreak Magnitude and Predictability in an Extended Multi-Pathogen SIR Model of Pigeons Visiting Dairy Farms. arXiv. 2023; 1–22. Available from: <http://arxiv.org/abs/2310.08613>