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Historic DNA reveals Anthropocene threat to a tropical urban fruit bat

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Anthropogenic activities have propelled the Earth into a crisis characterized by unprecedented levels of environmental degradation and habitat loss, generating changes in global climatic regimes and initiating the planet's Sixth Extinction Catastrophe [1]. Loss of population genetic diversity is known to be a harbinger of local and global extinction events [2]. However, there is a lack of direct empirical evidence of historic losses of genetic diversity through periods of anthropogenically linked environmental degradation. We present genomic DNA information from a population of Sunda fruit bats (Cynopterus brachyotis) from Singapore, an exceptionally well-studied tropical rainforest island that has undergone substantial environmental degradation and fragmentation through the

Anthropocene of the 1930–1950s [3]. As an effective pollinator and seed disperser, C. brachyotis represents an important keystone species in Singapore's ecosystem [4]. Here we show that comparison of historic DNA from individuals collected in 1931 with modern specimens reveals a nearly 30fold reduction in effective population size and corresponding levels of decline in genetic diversity estimates. Coalescent population models indicate that Singapore's C. brachyotis bats underwent a continuous decline in genetic diversity followed by a stark bottleneck in approximately the 1940s, consistent with the estimated onset of the Anthropocene [5]. C. brachyotis continues to be considered common across Singapore [4], yet our results reveal large-scale impacts of the Anthropocene on biotic communities, even in those species thought to be tolerant to the effects of environmental degradation.

C. brachyotis (Figure 1A) is widely distributed in human-dominated landscapes across Southeast Asia and is common in Singapore. We hypothesized that this bat has remained largely insensitive to urbanization and deforestation on this island. However, Singapore's urban matrix has affected population demography even in edge species accustomed to disturbed habitats [6], raising the possibility of declining population genetic diversity during the last century. We directly tested this hypothesis using a temporal sampling approach. If there is indeed a change in population demography and diversity in this common species, the general environmental effects of the Anthropocene on natural communities may be much more sweeping than previously appreciated.

Using DNA extracted from 41 individuals (Data S1A) across time (both contemporary and museum samples dating back to 1931), we sequenced approximately 2 Mbp of the *C. brachyotis* genome through sequence capture of 1,184 loci mined specifically for this study based on a comparison of an in-house generated *C. brachyotis* genome (accession ID SSHV0000000) with other bat genomes (see Supplemental Information).

We sequenced over 634 million 150-bp paired-end reads and generated multiple datasets: sequencebased (separate phased alignments of 874 and 184 loci) and SNP-based (one set of 24,782 genome-wide SNPs, and two subsets of transversions only – 7,646 and 22,314 SNPs – to account for ancient DNA based damage). Our observations clearly indicate an overall reduction in genetic diversity in Singapore's population of *C. brachyotis* over the past ~90 years (Figure 1B). Most summary statistics

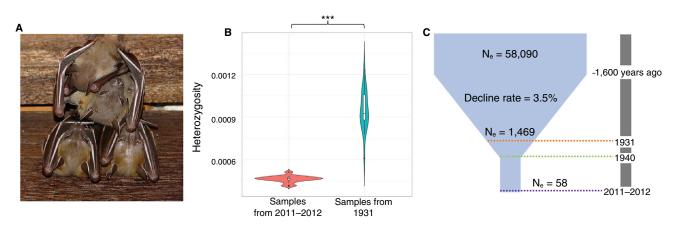


Figure 1. Decrease in genetic diversity and population size during the Anthropocene within a Singaporean population of Cynopterus brachyotis.

(A) Cynopterus brachyotis individuals at Singapore Zoo. Image courtesy of Wikimedia commons: Simon J. Tonge. (B) Loss in genetic diversity of Singapore's population of *C. brachyotis* as illustrated by a significant reduction in genome-wide probability of heterozygosity (estimated in ANGSD considering only transversions) between samples from 1931 and 2012; *** denotes p value < 0.001. (C) Illustration of recent demographic decline in *C. brachyotis* as observed from historical demographic modelling. Orange stippled line depicts the time of historical sampling (11 generations ago = ~1931), green stippled line depicts time to bottleneck (9 generations ago = ~1940) and purple stippled line depicts current sampling time (2011–2012). N_e: effective population size.

displayed a statistically significant difference between historic and contemporary diversity across datasets (Data S1B and S1C).

C. brachyotis is a generalist fruit bat that tolerates urbanized settings. As such, it is an unlikely victim of habitat degradation and fragmentation. The unexpected Anthropocene loss in genetic diversity in this common species indicates that the modern environmental crisis can generate silent adverse effects that only become apparent much later, when the impact of low genetic diversity may take hold in a population [7]. This phenomenon has been characterized as extinction debt, when actual extinction occurs with a time lag, long after the critical damage was done. Hence, an increased understanding of baseline levels and rates of loss of genetic diversity across organismic groups and habitats may - in the future - become imperative for informed conservation action.

Given the potential for large-scale fluctuations in demographic history [8], the collection of time series is a crucial prerequisite to establishing temporal trends in genetic diversity. Studies involving time series have demonstrated a reduction of genetic diversity and threat to the survival of wild populations specifically following industrialization [9]. But despite the immense utility of this comparative approach, few studies have addressed the loss of population-genomic diversity during the Anthropocene of the last few decades. Our investigation is an important addition in this regard.

In order to understand the effects of human-mediated changes on the evolutionary trajectory of Singapore's population of C. brachyotis, we reconstructed and compared diverse models of historical demography using the site frequency spectrum (see Supplemental Information). Across datasets, we recovered a strong signal of population decline following our historic sampling. The most complex model performed best for the sequence data (Data S1D), suggesting that Singapore's C. brachyotis population underwent a continuous decline that started ~195 generations ago (~1,600 years ago), and experienced a recent bottleneck nine generations ago (roughly in 1940) (Figure 1C, Data S1E).

For the SNP data, with much fewer loci, a simpler model of continuous decline following historic sampling was observed. Parameters estimated from the largest dataset (sequencebased, see Supplemental Information) were within the bounds obtained from parametric bootstraps, lending confidence to parameter estimation (Data S1E).

Our results have critical implications in two respects. First, the timing of the sudden bottleneck of this bat population coincides with the advent of the global Anthropocene [5], a period of rapid urbanization and drastic decline of forest cover in Singapore [3]. In contrast, the more gradual decline in effective population size of C. brachyotis over the past ~1,600 years may be a delayed response to the increasing isolation of the Singapore population from peninsular Malaysian populations during rising sea levels of the Holocene. Low plant diversity and the lack of certain megafauna in Singapore have been attributed to this isolation from the mainland [3]. Second, the Anthropocene bottleneck is characterized by a nearly thirtyfold reduction in effective population size and must be considered severe (Data S1E). Bottlenecks increase the vulnerability of a population to stochastic events and can accelerate extinction of small populations [2]. Bottlenecks following human interference have been documented in many endangered vertebrates [10], but our study suggests that even common species may not be immune to the effects of bottlenecks.

Our study demonstrates that recent advances in sequencing technology combined with historic sampling and population genetic tools can greatly aid in understanding patterns and processes of biodiversity loss. Humanmediated environmental fragmentation and degradation is not only detrimental to the survival of threatened species but may also affect and endanger common species cohabiting closely with humans.

SUPPLEMENTAL INFORMATION

Supplemental Information includes one figure, one excel data file and experimental procedures and can be found with this article online at https://doi.org/10.1016/j. cub.2019.11.013.

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Current Biology

Magazine

AUTHOR CONTRIBUTIONS

B.C and F.E.R conceived the idea; B.C., I.H.M. and K.M.G. collected all samples; K.M.G. and B.C. performed all lab protocols; B.C. and K.M.G. performed all analyses with critical input from F.E.R.; all authors contributed to the writing of the manuscript.

DECLARATION OF INTERESTS

The authors declare no competing interests.

REFERENCES

- Dirzo, R., Young, H.S., Galetti, M., Ceballos, G., Isaac, N.J., and Collen, B. (2014). Defaunation in the Anthropocene. Science 345, 401–406.
- Frankham, R., Briscoe, D.A., and Ballou, J.D. (2002). Introduction to Conservation Genetics. (Cambridge, UK: Cambridge University Press).
- Corlett, R.T. (1992). The ecological transformation of Singapore, 1819–1990. J. Biogeogr. 19, 411–420.
- Ming, L.T., and Wai, C.K. (2011). Bats in Singapore – ecological roles and conservation needs. In NSS Symposium 2011: Nature Conservation for a Sustainable Singapore. pp. 41–64.
- Corlett, R.T. (2015). The Anthropocene concept in ecology and conservation. Trends Ecol. Evol. 30, 36–41.
- Tan, D.J., Chattopadhyay, B., Garg, K.M., Cros, E., Ericson, P.G., Irestedt, M., and Rheindt, F.E. (2018). Novel genome and genomewide SNPs reveal early fragmentation effects in an edge-tolerant songbird population across an urbanized tropical metropolis. Sci. Rep. 8, 12804.
- Kuussaari, M., Bommarco, R., Heikkinen, R.K., Helm, A., Krauss, J., Lindborg, R., Öckinger, E., Pärtel, M., Pino, J., and Rodà, F. (2009).
 Extinction debt: a challenge for biodiversity conservation. Trends Ecol. Evol. 24, 564–571.
- Ramakrishnan, U., Hadly, E.A., and Mountain, J.L. (2005). Detecting past population bottlenecks using temporal genetic data. Mol. Ecol. 14, 2915–2922.
- Leigh, D.M., Hendry, A.P., Vázquez-Domínguez, E., and Friesen, V.L. (2019). Estimated six percent loss of genetic variation in wild populations since the industrial revolution. Evol. Appl. 12, 1505–1512.
- Chattopadhyay, B., Garg, K.M., Yun Jing, S., Low, G.W., Frechette, J., and Rheindt, F.E. (2019). Conservation genomics in the fight to help the recovery of the critically endangered Siamese crocodile *Crocodylus siamensis*. Mol. Ecol. 28, 936–950.

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