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Response to Comment on “Global Genetic Change Tracks Global Climate Warming in Drosophila subobscura”

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Rodríguez-Trelles and Rodríguez advocate standardizing old and new collections by climate rather than by calendar and also propose that some of our samples were biased by inappropriate timing. Their first suggestion applies to few species, and its implementation alters photoperiodic cues. Their second point is valid, but our conclusions are robust: Observed genetic changes reflect global warming, not sampling artifacts.

In their specific objection to the timing of our samples (4), Rodríguez-Trelles and Rodríguez (1) assert that inversion polymorphisms of D. subobscura undergo “pronounced seasonal cycles” and then argue that “it is conceivable” that our between-sample differences might be a sampling artifact. However, their statement that inversion polymorphisms cycle seasonally in D. subobscura is not universally accepted (13, 14), and the example they cite (15) established cycles only for the O chromosome. To help resolve this controversy, we reanalyzed a large data set (16) for a Catalonian population of D. subobscura. For each of the 11 months, we computed a genome-wide PC1 score (4) and regressed it on month using a third-order polynomial weighted by sample size. PC1 shows temporal variation but no evidence of a sinusoidal trend (all P > 0.24). Thus, genome-wide seasonal cycles are undetectable at this site.

We did not include sampling dates in our study (4), so Rodríguez-Trelles and Rodríguez (1) relied on published dates (17) for the 13 European sites. They objected that four new European sites were collected in a cooler month, while seven of our new North American sites were collected in a cooler month, and all six of our new South American samples were collected in a cooler or the same month. Thus, for most of our sites, seasonal shifts in inversion frequencies would only reduce the magnitude of change over time, not exaggerate it as suggested by Rodríguez-Trelles and Rodríguez (1).

If the four sites did bias the inversion data, then the difference in monthly ambient temperatures should be correlated with the difference in chromosome index (Ch_{PC1}) among sites. This correlation is significant (Spearman rho = 0.452, P = 0.021), and the correlation is eliminated when these four samples are deleted (P = 0.0047, P = 0.834). Thus, the four sites did induce bias. Is the bias sufficient to challenge our conclusions? To evaluate this, we deleted the four problematic samples and re-ran all analyses. The increase in the intercept from old to new samples remains significant for temperature (F_{1,21} = 19.3, P = 0.00025) and for chromosomes (F_{1,21} = 37.6, P = 0.000004). Moreover, chromosomes and temperature still shift in parallel (Rayleigh test: \( \hat{r} = 0.60, P = 0.0002 \)). Therefore, our conclusions are robust.

We concur with Rodríguez-Trelles and Rodríguez (1) that the timing of collections should be considered carefully. However, their suggestion to standardize by climate rather than by calendar date will bias photoperiodic cues. Moreover, the observed growing-season shifts of a few days will generally be too small to have major genetic impact and thus bias results. Our new analyses reject their claim that inversions in this species show pronounced seasonal cycles. Finally, with regard to our own study, 4 of 26 samples are indeed problematic, but our original conclusions hold when those samples are excluded. We stand by our original conclusion that global genetic change in D. subobscura is tracking global warming and is not a sampling artifact.

References and Notes
1. F. Rodríguez-Trelles, M. Á. Rodríguez, Science 315, 1497 (2007); www.sciencemag.org/cgi/content/full/315/5818/1497a.

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