

# On the definition and measurement of pleiotropy

Jianzhi Zhang<sup>1</sup> and Gunter P. Wagner<sup>2</sup>

<sup>1</sup> Department of Ecology and Evolutionary Biology, University of Michigan, 830 North University Avenue, Ann Arbor, MI 48109-1048, USA

<sup>2</sup> Yale Systems Biology Institute and Department of Ecology and Evolutionary Biology, Yale University, 165 Prospect Street, POB 208106, New Haven, CT 06405-8106, USA

Pleiotropy, a 103-year-old concept [1], refers to the phenomenon of one mutation affecting multiple traits. Compared with the fundamental importance of pleiotropy in genetics, development, evolution, and medicine, our empirical knowledge of the prevalence and mechanism of pleiotropy is scarce [2]. Given this context, Paaby and Rockman's recent Opinion article on pleiotropy and its empirical estimation [3] is timely, but several of their claims are debatable.

First, based on the type of traits concerned, Paaby and Rockman proposed three kinds of pleiotropy, namely 'molecular gene pleiotropy', 'developmental pleiotropy', and 'selectional pleiotropy'. Although there is no doubt that the pleiotropic level of a mutation can vary depending on the types of trait considered, this classification scheme is fundamentally a categorization of traits rather than of pleiotropy. As an analogy, consider the common terms 'dominance' and 'recessiveness'. Whether a particular allele is dominant over another depends on the trait considered. For example, missense mutations in the gene encoding hexosaminidase A (*HEXA*) often cause the recessive Tay–Sachs disease (OMIM #272800) [4], but at the level of mRNA, the mutant alleles and wild type alleles are codominant, because the mutations do not affect *HEXA* gene expression. Despite the dependence of dominance on the trait considered, dominance or recessiveness is not classified, as Paaby and Rockman proposed for pleiotropy, as expression dominance or developmental dominance. We believe that, as long as the trait under discussion is clear, there is no need to classify dominance, recessiveness, or pleiotropy.

Second, Paaby and Rockman contended that a previous summary of the reverse genetic data on yeast gene pleiotropy [5] is biased because of the exclusion of essential genes, which are defined as genes that cause lethality or sterility when deleted. Note that gene pleiotropy is a simplified phrase for the pleiotropy of null mutations in a gene, because, strictly speaking, pleiotropy is a property of mutations rather than of genes. Paaby and Rockman considered essential genes to affect all traits of an organism and to be maximally pleiotropic. Although this is a potential way of looking at the situation, in most contexts lethal and/or sterile mutations are not relevant to start with. The pleiotropy of lethal and/or sterile mutations does not inform about the evolvability of a genotype, because such mutations will never compete for representation in the next generation. Furthermore, if null mutations of essential genes are specifically investigated for certain traits, their levels of pleiotropy are not maximally high [5,6].

Corresponding author: Zhang, J. (jianzhi@umich.edu); Wagner, G.P. (gunter.wagner@yale.edu).

Third, Paaby and Rockman argued that the pleiotropic level has been grossly underestimated from recent reverse genetic and quantitative trait locus mapping data [2,5,7–9], because they believe that these methods are not sensitive enough to detect most phenotypic effects of mutations. Although increasing the sample size and the precision of measurement is expected to boost the estimated degree of pleiotropy, the argument misses two critical points. First, the key question is not how many traits are significantly affected by a mutation statistically, but biologically. Of course, we do not have scalable methods to estimate fitness effects of trait differences, but in principle we should not mistake statistical issues for biological insights. Second, even if more phenotypic effects are detected upon experimental improvement, their effect sizes are expected to be smaller than those that have been detected. The majority of these small effects may be biologically irrelevant.

Finally, as mentioned previously [2], a measure of pleiotropy that is not sensitive to the statistical power of the experiment is urgently needed; for instance, methods describing the distribution of estimated effect sizes across traits, rather than only counting the statistically significant effects.

Although the total amount of empirical data for studying pleiotropy is still limited, the past few years have seen a rapid growth of suitable data, which has led to significant expansion of our knowledge of the prevalence and mechanism of pleiotropy [2,10,11]. We believe that the most productive way forward is to expand and improve attempts to estimate pleiotropy empirically. Paaby and Rockman highlighted two pleiotropy-related hypotheses on the genetic mechanisms of phenotypic evolution and aging that they suggest are well suited to exploring this phenomenon [3]. Although they mentioned the argument that *cis*-regulatory mutations are less pleiotropic than are coding sequence mutations as an example of a testable hypothesis, to our knowledge, there is only anecdotal evidence to support this. An empirical demonstration of the lower pleiotropy of *cis*-regulatory mutations than of coding sequence mutations will only be possible if the methodological problems discussed above are addressed. Similarly, the genes and mutations that antagonize development and/or reproduction and life span, and the mechanism of their trade-off have not been identified [12]. Although many basic questions on pleiotropy remain, we are hopeful that, with rapid advancement of genetic and genomic technologies and data-driven theory development, these questions will be addressed in the near future by empirical characterization of pleiotropy at a high resolution, on a large scale, and for many species.

## References

- 1 Plate, L. (1910) Genetics and evolution. In *Festschrift zum sechzigsten Geburtstag Richard Hertwigs*. pp. 536–610, (in German), Fischer
- 2 Wagner, G.P. and Zhang, J. (2011) The pleiotropic structure of the genotype–phenotype map: the evolvability of complex organisms. *Nat. Rev. Genet.* 12, 204–213
- 3 Paaby, A.B. and Rockman, M.V. (2013) The many faces of pleiotropy. *Trends Genet.* 29, 66–73
- 4 Myerowitz, R. (1997) Tay–Sachs disease-causing mutations and neutral polymorphisms in the Hex A gene. *Hum. Mutat.* 9, 195–208
- 5 Wang, Z. et al. (2010) Genomic patterns of pleiotropy and the evolution of complexity. *Proc. Natl. Acad. Sci. U.S.A.* 107, 18034–18039
- 6 Liao, B.Y. et al. (2010) Contrasting genetic paths to morphological and physiological evolution. *Proc. Natl. Acad. Sci. U.S.A.* 107, 7353–7358
- 7 Wagner, G.P. et al. (2008) Pleiotropic scaling of gene effects and the ‘cost of complexity’. *Nature* 452, 470–472
- 8 Albert, A.Y. et al. (2008) The genetics of adaptive shape shift in stickleback: pleiotropy and effect size. *Evolution* 62, 76–85
- 9 Dudley, A.M. et al. (2005) A global view of pleiotropy and phenotypically derived gene function in yeast. *Mol. Syst. Biol.* 1, 2005.0001, (<http://dx.doi.org/10.1038/msb4100004>)
- 10 He, X. and Zhang, J. (2006) Toward a molecular understanding of pleiotropy. *Genetics* 173, 1885–1891
- 11 Qian, W. et al. (2012) The genomic landscape and evolutionary resolution of antagonistic pleiotropy in yeast. *Cell Rep.* 2, 1399–1410
- 12 Flatt, T. (2011) Survival costs of reproduction in *Drosophila*. *Exp. Gerontol.* 46, 369–375

0168-9525/\$ – see front matter © 2013 Elsevier Ltd. All rights reserved.  
<http://dx.doi.org/10.1016/j.tig.2013.05.002> Trends in Genetics, July 2013, Vol. 29, No. 7

## Pleiotropy: what do you mean? Reply to Zhang and Wagner

Annalise B. Paaby and Matthew V. Rockman

Department of Biology and Center for Genomics and Systems Biology, New York University, New York, NY, USA

It is a testament to the conceptual slipperiness of pleiotropy that thoughtful people can reasonably disagree about its meaning and extent. Zhang and Wagner [1] raise important issues in response to our recent Opinion article [2], and readers will do well to consider their critique. Here, we offer a brief counterpoint to the objections that they raise.

First, Zhang and Wagner argue against our categorization of pleiotropy into three distinct forms. At some level, this point is semantic, because we could as well have described a single kind of pleiotropy in three contexts: molecular genetics, developmental genetics, and evolutionary genetics. Our central goal, however, is to discourage inappropriate applications of data from one context to theory from another. Zhang and Wagner’s analogy to dominance illustrates our concern. As they note, a single mutation can have different dominance with respect to different phenotypes. Where the term ‘dominance’ appears in evolutionary population genetics, however, it refers exclusively to dominance with respect to fitness. Given that phenotypes may vary nonlinearly with fitness, data on dominance in the context of development may be misleading about the role of dominance in the evolutionary fate of mutations. The same holds for pleiotropy.

Next, Zhang and Wagner argue that lethal gene deletions are uninformative because they cannot contribute to evolution. This is our point. If the goal is to estimate the pleiotropy of null mutations to answer questions about evolution, then those mutations too pleiotropic to ever contribute to evolution are exceedingly relevant. They demonstrate that pleiotropy constrains evolution. Zhang and Wagner suggest that the patterns of pre-lethality

developmental pleiotropy exhibited by such mutations are informative, but we reiterate the importance of distinguishing developmental from selectional pleiotropy in such cases. Embryonic lethals all have the same pattern of selectional pleiotropy.

The third point of disagreement is whether subtle allelic effects that lie below thresholds for statistical detection are biologically important. We believe that the weight of empirical data decisively favors an affirmative answer, and we invite readers to revisit our notes on this point in the original Opinion [2]. We share with Zhang and Wagner the view that the development of methods to measure pleiotropy without dependence on statistical thresholds is an important goal.

Finally, Zhang and Wagner suggest that the way forward is more data. As empiricists, we echo this point. However, data alone are not sufficient. We need data that are relevant to our hypotheses. Zhang and Wagner suggest that the examples we offer of successes in the study of pleiotropy (the *cis*-regulatory theory of morphological evolution and the antagonistic pleiotropy model of aging) are themselves in dire need of such data, and we do not doubt that they remain hypotheses. Their virtue, from our perspective, is that they are well-posed hypotheses, such that the relevant data are precisely defined. In our Opinion, we argue that many questions about pleiotropy are ill posed, and that greater attention to conceptual foundations will improve our empirical progress.

### References

- 1 Zhang, J. and Wagner, G.P. (2013) On the definition and measurement of pleiotropy. *Trends Genet.* 29, 383–384
- 2 Paaby, A.B. and Rockman, M.V. (2013) The many faces of pleiotropy. *Trends Genet.* 29, 66–73

DOI of original article: <http://dx.doi.org/10.1016/j.tig.2013.05.002>  
 Corresponding author: Paaby, A.B. ([aapaaby@nyu.edu](mailto:aapaaby@nyu.edu)); Rockman, M.V. ([mrockman@nyu.edu](mailto:mrockman@nyu.edu)).

0168-9525/\$ – see front matter © 2013 Elsevier Ltd. All rights reserved.  
<http://dx.doi.org/10.1016/j.tig.2013.05.003> Trends in Genetics, July 2013, Vol. 29, No. 7