Sprinters versus marathon runners – are there differences in how Drosophila respond to exercise induction?

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ABSTRACT
Animal activity is an important trait that influences an organism's fitness. For example, when an organism is active, during the day or the night, is important as this choice greatly impacts its ability to find food or mates and its chances to encounter predators or prey. It is also important how organisms respond to external stimulation, i.e. how much energy to expend on such a stimulation in terms of activity. We have used the DGRP strain collection to investigate variation in activity levels in *Drosophila melanogaster*, both for basal activity levels without stimulation and for activity levels induced by continued rotational stimulation (exercise). We found that within the DGRP strain collection, activity levels varied by approximately 500-fold and that the rotational stimulation is able to increase activity levels in most of the strains. Using a GWAS, we linked the variation in activity levels to over 400 genetic variants present in the DGRP strain collection. Here, we use this dataset to investigate how the various Drosophila strains respond to the rotational stimulation, either by a short burst of activity following the start of the rotation (sprinters) or by continued activity throughout the two-hour exercise period (marathoners). We find that the DGRP strains vary greatly in their response pattern, with some strains exhibiting less than 10% of their activity in the first quarter of the exercise period and other strains exhibiting over 85% in this time interval. The amount of activity performed during the first quarter of the exercise period is strongly dependent on the sex of the animals, their genotype, as well as the interaction of sex and genotype. We will present the results of an ongoing GWAS, which suggests that the response type exhibited by Drosophila, either a short burst or sustained activity, might be under genetic control.

1. Different Drosophila genotypes vary in the amount of activity performed during the first 25% of a 2hr exercise period.

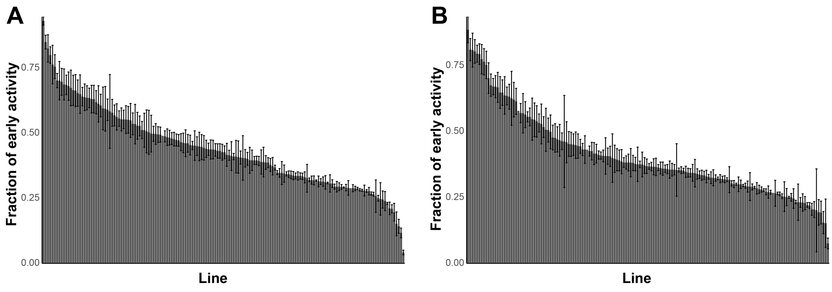

Figure 1. The average fraction of activity performed early (in the first 30 minutes of a 2hr exercise period; Y-axis) is plotted for each of the DGRP strains in this study (X-axis). The strains are ordered by the average fraction of activity performed early, from highest to lowest, and the error bars represent standard errors.
A. Data from females.
B. Data from males.

2. Approx. 50 genetic variants contribute to how animal activity is biased towards the 1st quarter of the exercise period.

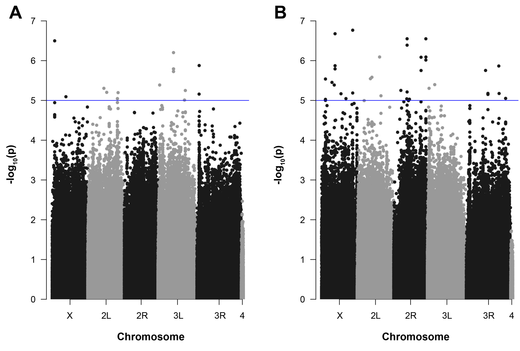

Figure 2. Manhattan plots illustrate the genetic variants that contribute to how much an animal's activity is biased towards the first quarter of the exercise period. X-axis: Location of the genetic variant along the four Drosophila chromosomes. Y-axis: $\log_{10}$ of the p-value of each genetic variant from the regression analysis. Significance line: $p<10^{-5}$.
A. Data from females.
B. Data from males.

3. The candidate variants identified by the GWAS reflect the functional categories of variants in the DGRP population.

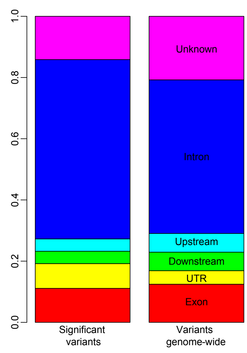

Figure 3. All variants in the DGRP population were classified based on the annotation available. The variants significantly associated with exercise patterns (left) is not significantly different from the variant distribution seen genome-wide (right; p=0.1564, chi-square test). Y-axis: fraction of variants.

4. Functional enrichment analysis reveals links of candidate genes to muscle function and animal activity.

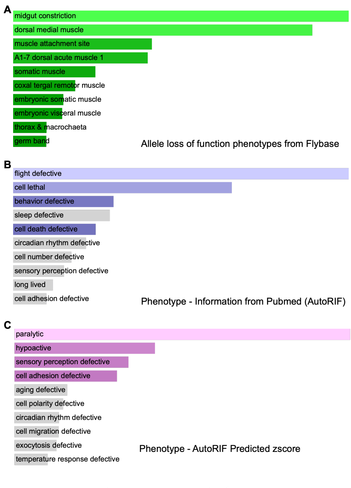

Figure 4. Functional enrichment analysis results from FlyEnrichr. Terms in grey are not significantly enriched and the length of the bars indicate the significance level of enrichment.
A. Enrichment analysis for allele loss of function phenotypes mined from Flybase.
B. Enrichment analysis for phenotype information mined from PubMed (AutoRIF).
C. Enrichment analysis for phenotype information mined from genes related to the gene set based on PubMed and co-expression data (Phenotype AutoRIF Predicted z-score).

5. A split sample validation analysis identifies 10 high confidence candidate genes.

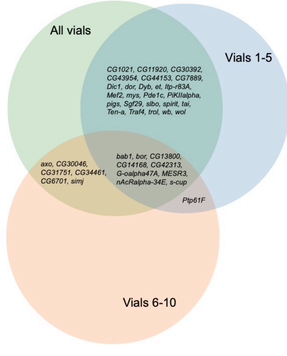

Figure 5. The original GWAS analysis was based on exercise measurements from 10 vials of 10 flies per sex and genotype. To validate this analysis, we split the sample, analyzing data from vials 1-5 and 6-10 separately. Proportional Venn diagram showing the overlap in candidate genes identified in our initial GWAS ("All vials") as well as the two split sample analyses ("Vials 1-5" and "Vials 6-10").

Conclusions

- **There are clear differences in activity patterns in response to rotational exercise stimulation among the DGRP strains, with some strains showing short bouts of activity ("runners") and others showing prolonged activity ("marathoners") over the 2hr exercise period.**
- **The activity patterns are strongly impacted by sex and genotype.**
- **GWAS analysis identifies more than 50 genetic variants linked to the exercise activity, with 10 high confidence candidate genes being confirmed by a split sample validation.**
- **The transcription factor *bab1* is linked to activity in both males and females.**

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