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# Startle induced negative geotaxis (SING)

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**We use this protocol and it's working**

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## Abstract

This protocol describes how to perform the Startle induced negative geotaxis (SING) assay. SING defects in parkinsonism fly models have previously been shown to be caused by dopaminergic impairments.

## Startle induced negative geotaxis (SING)

30s

1 for the describtion of the SING apparatus please refer to:
Benzer, Seymour. 1967. “BEHAVIORAL MUTANTS OF *Drosophila* ISOLATED BY COUNTERCURRENT DISTRIBUTION.” *Proceedings of the National Academy of Sciences* 58 (3): 1112–19. https://doi.org/10.1073/pnas.58.3.1112.

Inagaki, Hidehiko K, Azusa Kamikouchi, and Kei Ito. 2010. “Methods for Quantifying Simple Gravity Sensing in Drosophila Melanogaster.” *Nature Protocols* 5 (1): 20–25. https://doi.org/10.1038/nprot.2009.196.

2 set a specific time period during the day (e.g. Zeitgeber 3-5) to assess negative geotaxis behavior, which will be kept the same for all following experiments

3 flies are transferred into the first tube of the apparatus without prior anesthesia and allowed to adjust
for some minutes to experimental environment (24±1°C, 40-60% humidity in behavior chamber, note down the environmental condition and time for every genotype)

4 tap the flies down five times and allowed to climb up for 00:00:30

30s

5 after these 30 s the flies that reached the upper tube are moved to the next tube

6 repeat the procedure 4 times

7 count the number N of (male/female) flies with the corresponding genotype per tube to calculate the SING score, which is normalized to the mean SING score of the control.

8 SING score = $((N_1 *0) + (N_2 *1) + (N_3 *2) + (N_4 *3) + (N_5 *4))/ 4(N_1 + N_2 + N_3 + N_4 + N_5)$
$N_k$ = number of flies in the *k*th tube

## Protocol references

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