

Fly Resveratrol Treatment Protocol

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Protocol reference: Brown JB et al. 2014 (PMID: 24670639)

Treatment feeding schedule for Larvae:

For each treatment, approximately 50 (mixed sex) young mated adults were transferred to each fresh food vials and maintained for 12 hours. Vials were cleared and allowed to age 3.5 to 4 days. Vials were then rinsed into a series of sieves using tepid water; feeding third instar larvae were collected from the #40 sieve and transferred to a hard agar plate with a pot of yeast to induce crawling. Prior to reaching the yeast, larvae were captured and 50 larvae were transferred to new food vials containing the treatment of interest (details below), and larvae were allowed to feed for 4 hours. Treated larvae were captured and transferred to 2 ml vials, flash frozen in liquid nitrogen and stored at -80° C prior to RNA preparations. The number of survivors was recorded and the mean lethality calculated for each treatment.

Treatment feeding schedule for Adults:

For each treatment, 40 newly eclosed males and females (1:1) were transferred to fresh food (BDSC corn meal agar) vials and maintained at 25°C for two days. To treat flies, two Kimwipes were folded into a square and put in the bottom of a one-pint glass bottle. Kimwipes were saturated with 4 ml of the treatment solution, (10% sucrose solution and one drop of green vegetable coloring per 50 ml solution, plus the treatment of interest). Harvesting time for adults varied by treatment. Upon harvesting, flies were placed in 2 ml tubes, flash frozen in liquid nitrogen and stored at -80°C prior to RNA preparations.

Resveratrol treatment:

Two-day-old adults were fed 100 µM resveratrol in 10% sucrose continuously and samples were harvested at 10 days. Adult flies were flash frozen in liquid nitrogen and stored at -80°C.