

## **UVB EXPOSURE PROTOCOL**

**UV treated Animal:** Hybrid fish are placed in the dark 24 hrs before first treatment. Fish are treated on day 5 post parturition for 5 consecutive days for a total of 5 treatments. Groups of fry are exposed to a total dose of 6.4 kJ/m<sup>2</sup> at a fluence rate of 12.2 J/m<sup>2</sup>/sec (Mitchell et al. 2010) from two unfiltered Philips TL01 bulbs mounted on either side of the irradiation chamber (four bulbs total). The total UVB exposure time is 8 min 45 sec daily for 5 days. Fish are kept in the dark after each exposure and for 24 hrs after the final exposure before being returned to their tank and normal rearing conditions. The fish will be reared for one year (or until tumor develops) under normal XGSC conditions & then dissected.

**Control Animal:** Control Animals will not receive UV treatment. These fish will be reared under normal XGSC conditions for one year and then dissected to establish/confirm base line tumor formation data.

**Dissections:** Once all fish reach one year of age they will be dissected to observe any internal tumor formations or other abnormalities and to collect tissue samples as well as data. Also if a fish develops tumors or illness that indicate the fish is suffering or dying then it will be dissected sooner to prevent sample loss. Fish will be photographed, measured (SL) and scored just prior to dissection. Normal tissues to collect include brain, eyes, gill, gonad, liver, muscle, skin and tailfin. All tissues will be stored at -80C in RNA later. If the fish has any tumor formation or similar abnormality the sample will be dissected and preserved in RNA later and stored in -80C.