Supplementary Information

A MICROFLUIDIC DEVICE FOR PARTIAL IMMOBILIZATION, CHEMICAL

EXPOSURE AND BEHAVIOURAL SCREENING OF ZEBRAFISH LARVAE

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1. Zebrafish Larva's Body Size



Figure S1. (a) Shows the zebrafish larvae and the different anatomical regions highlighted. (b) Shows the size of 5-7 dpf TL strain larvae for different anatomical regions. N=10 larvae.

2. Removal of an Air Bubble from the Assay Chamber



Figure S2. Extraction of an air bubble trapped within the microfluidic chamber. Sequential images (i-iii) captured 2 minutes apart show the air bubble diminishing in size until it vanishes completely (iv).

3. Zebrafish Larva's Body Size

The narrowing channel used for immobilization and the wide chamber designed for allowing sharp tail movements by larvae did not create ideal conditions for the water flow. In the absence of the C-shape PDMS, water took a path similar to that illustrated in Fig. S3a. Water could not reach the corners of the chamber and the presence of air in the system introduced air bubbles during unloading and reloading of larvae. Therefore, we implemented the C-shape PDMS to help direct water to the corners and fill the chamber evenly (Fig. S3b).



Figure S3. (a) Illustration of the path water takes in the absence of C-shaped PDMS after travelling through the narrowing channel. (b) i-iv show that in the presence of a C-shaped PDMS the flow of water towards the chamber edges becomes more favourable.

Video S1. Loading of a 7 dpf TL strain zebrafish larvae into the trap of the microfluidic device followed by the actuation of an orthogonal microfluidic valve for head-immobilization.

Video S2. A head-immobilized 6 dpf TL strain zebrafish larvae at the trap of the microfluidic device displaying rapid and sharp C-turns while exposed to mM level of L-arginine.