Supplementary Materials for "Microfluidic chamber arrays for whole-organism behavior-based chemical screening" by Chung, Zhan et al.



Figure S1. Repeated loading of DI water and blue food coloring into the device using a syringe pump operated at 0.3 ml/min. 100 μ L of solution was injected for each loading cycle. We observed that the chambers reach steady state concentration after 50 μ L injections.

To gauge uniformity over all 48 chambers, we compare chamber 1 (the first chamber in the serpentine channel flow path) and chamber 48 (the last chamber in the serpentine chamber flow path) over all of the repeated loadings. Using a paired t-test between the dataset for chamber 1 and chamber 48, we find no statistical difference (p>.6).



Figure S2. Loading of blue food coloring into a device primed with DI water using a syringe pump injecting at 0.3 ml/min. Red asterisks indicate the chamber intensities for each chamber after 50 μ l of the dye had been injected through the device. Black circles indicate the chamber intensities for each chamber after 250 μ l of the dye had been injected through the device. The mean intensity over all chambers for 50 μ l injection was 2.1107e+004 (a.u.); the mean intensity over all chambers for 250 μ l injection was 2.1077e+004 (a.u.). There is no statistical difference between the data sets (p>0.3), indicating 50 μ l is sufficiently large for exchanging out the existing solution in all chambers.



Figure S3. HPLC-MS ion chromatograms for the ion m/z = 269, corresponding to $[M+Na]^+$ of ascaroside ascr#2. **A**: HPLC-MS ion chromatogram obtained with injection of 1 µL of a 10 µM solution of ascr#2 in water. **B**: HPLC-MS ion chromatogram obtained with injection of 1 µL of the same solution after pass through microfluidic device. Integration of the peaks representing ascr#2 using MassLynx software (Waters) indicated less than 5% loss.

Supplementary Method for HPLC-MS characterization of compound adsorption:

Device was equilibrated with water. Subsequently 100 μ L of a 10 μ M solution of ascr#2 in water was pumped through the device via syringe pump. Collection was started after 50 μ L of solution had passed through the device. HPLC-MS analysis was performed using an Agilent 1100 Series HPLC system equipped with an Agilent Eclipse XDB-C18 column (9.4 x 250 mm, 5 μ m particle diameter). A 0.1% acetic acid – acetonitrile solvent gradient was used, starting with an acetonitrile content of 5% for 5 min which was increased to 100% over a period of 40 min. The HPLC system was connected to a Quattro II mass spectrometer (Micromass/Waters), which was operated in positive-ion electrospray ionization mode.