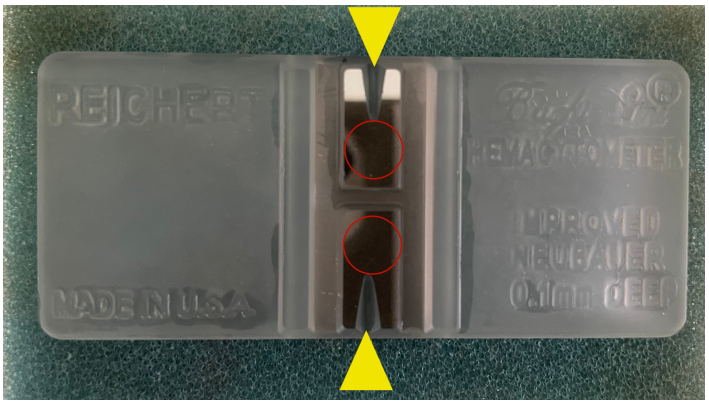
# Symbiodiniaceae counts

## 0.1 Material

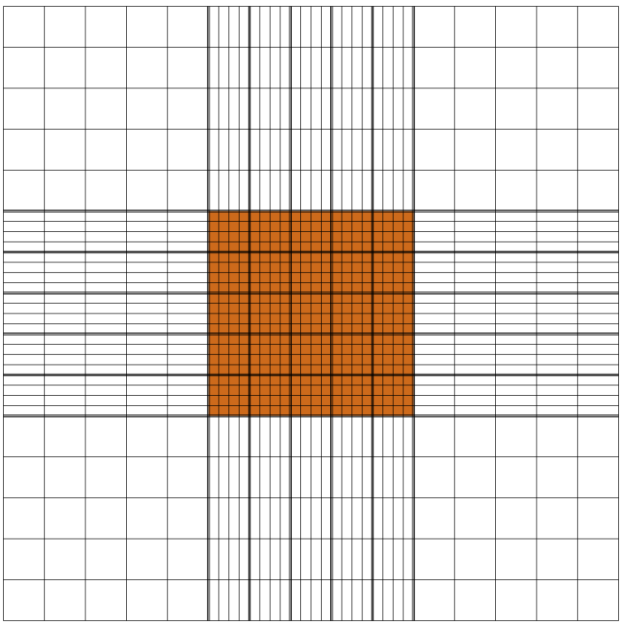
- ☐ Hemacytometer (Neubauer improved - Bright-Line) with glass cover
- ☐ Ethanol 96% for cleaning
- ☐ Kimwipes
- ☐ 10 µL pipette and tips
- ☐ Microscope with objective lens 20X or 40X

## 0.2 Method

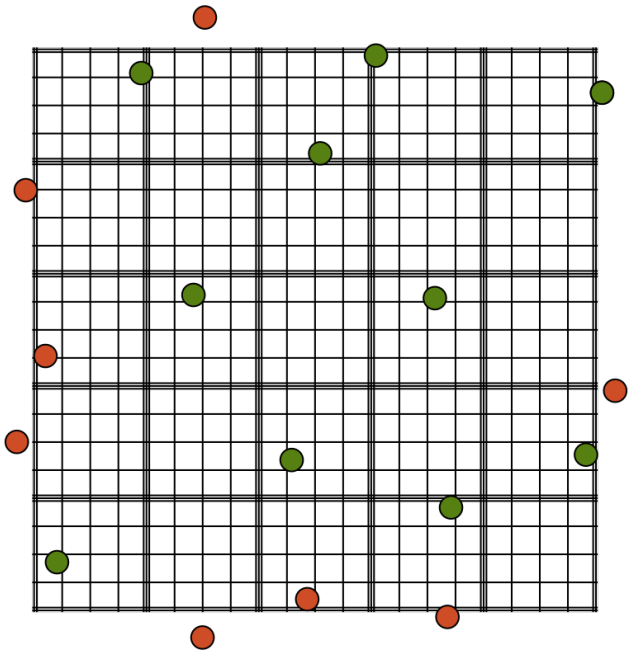
1. Clean hemacytometer with distilled water or ethanol and Kimwipes.
2. Two chambers exist on the hemacytometer (red circle below). Place a coverslip covering both chambers. Vortex sample for 15 s and add 10 µL sample solution at the entry points for each chamber (yellow triangles).




3. Count all Symbiodiniaceae cells in the central grid marked in orange below. Count the cells in both chambers and note in the c# columns in the metadata sheet.



4. If a cell touches the border (3 lines) on the top or right side, include it in the counts (green cells below, only the central grid is shown); if it touches the border at the bottom or left side, do not include it in the counts (red cells below).



5. If you can estimate that more than 100 cells are in the central grid, dilute sample and note dilution, e.g. 200 mL sample (`V_zoox` in metadata sheet) with 800 µL SST (`V_sw`). Vortex the sample before diluting and the diluted sample.
6. Repeat above steps in total three times per samples, yielding 6 count replicates per sample.
7. After each count, clean hemacytometer with ethanol and Kimwipes.