(1) Primary Screen
via Nile red coupled to flow cytometry

- Cells being stained with Nile red
- Detection of Nile red fluorescence with flow cytometer

Culture of *C. reinhardtii* in 96 deep-well plates

- Chlorophyll fluorescence (FL3)
- Nile red fluorescence (neutral lipids) (FL2)

Wild-type phenotype

A high-oil phenotype

Positive clones

(2) Confirmation Step

- Repeat the same procedure as above
- Biological triplicates

Confirmed clones

(3) Validation Step
via TAG quantification based on TLC

Oil mutants

Quantification of triacylglycerols (TAGs) (TLC, densitometry)

C17:0 TAG standard

Loading origin

insertional mutants

Culture of *C. reinhardtii* in 96 deep-well plates

Lipid extraction

Positive clones

Wild-type phenotype

A high-oil phenotype

- Repeat the same procedure as above
- Biological triplicates

Confirmed clones