

Supplementary information

Supplementary Figure 1

Flow cytometry of fixed Nile red stained worms reveals fat content per volume in mutants *glp-1* (*e2141*), *eat-2* (*ad465*) and *daf-7* (*e1372*) relative to wild type N2. The same staining protocol as for BODIPY 493/503 staining was used. Nile red fluorescence as function of extinction is a proxy for fat content per volume of *eat-2*, *daf-7*, *glp-1* (25 °C) relative to wild type.

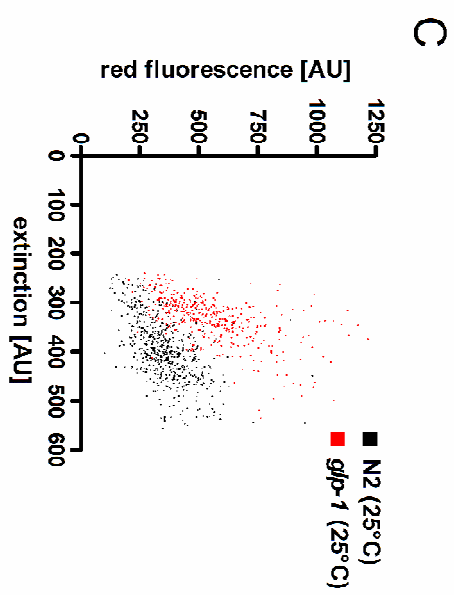
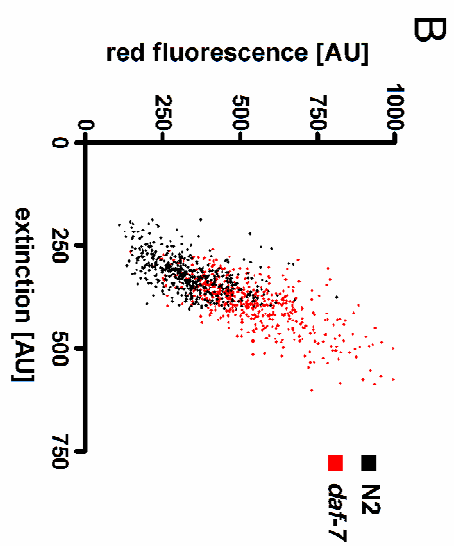
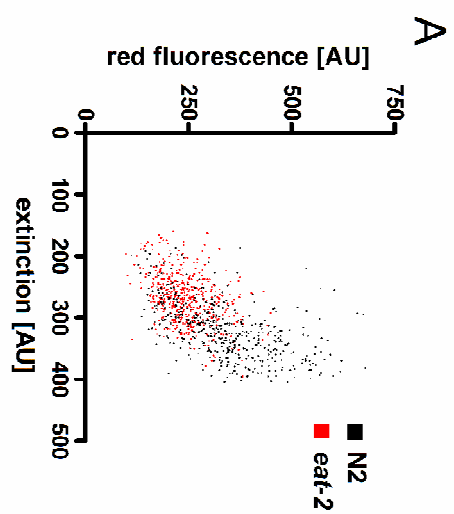
Supplementary Figure 2

Conventional microscopy shows: (A) Vital staining of worms with BODIPY 493/503 with an incubation time of 20 min stains lipid droplets, as proven by co-localisation with CARS signals (Fig.10 C), distinct from LysoTracker Red positive, autofluorescent LROs. (B) However, long term feeding of the worms with BODIPY 493/503 on NGM plates over 18 h leads to accumulation of the dye in the autofluorescent LROs.

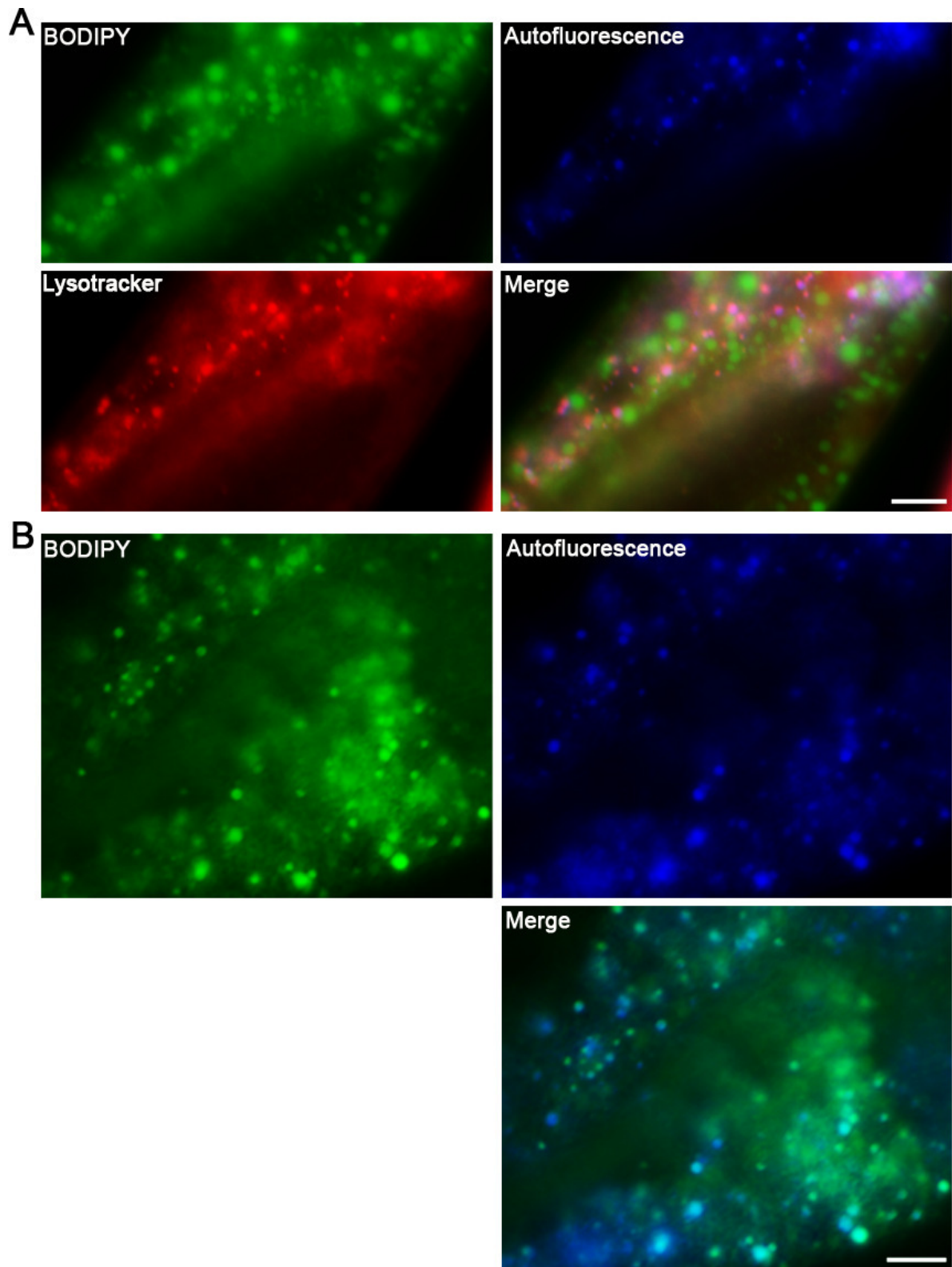
Supplementary Figure 3

Data are shown that that the triacylglycerol assay is suitable for the determination of TAGs in *C. elegans*. (A) Triglyceride measurements using worm extracts with the assigned numbers of worms/70 μ l lysis buffer revealed similar triglyceride contents per worm (ng/ worm) independent of the worm number. (B) The triolein standard curve -■- if spiked with worm homogenate --□-- shows a nearly identical slope, analyzed by linear regression. (A,B) 8 μ l worm extract were used for each reaction. Assays were carried out in duplicates. Data are presented as mean \pm SD. These results demonstrate, that the TAG assay is not interfered by any components in the worm extract, at least in this range.

Suppl. Figure 1



Suppl. Figure 2



Suppl. Figure 3

