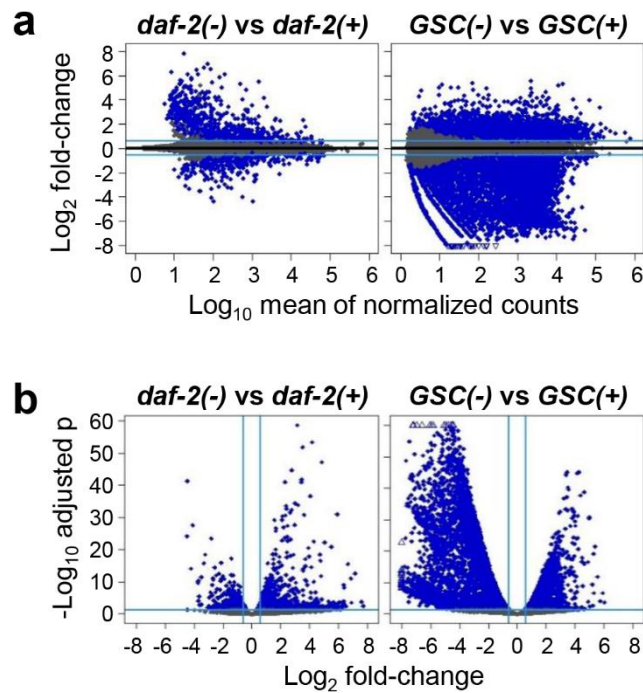
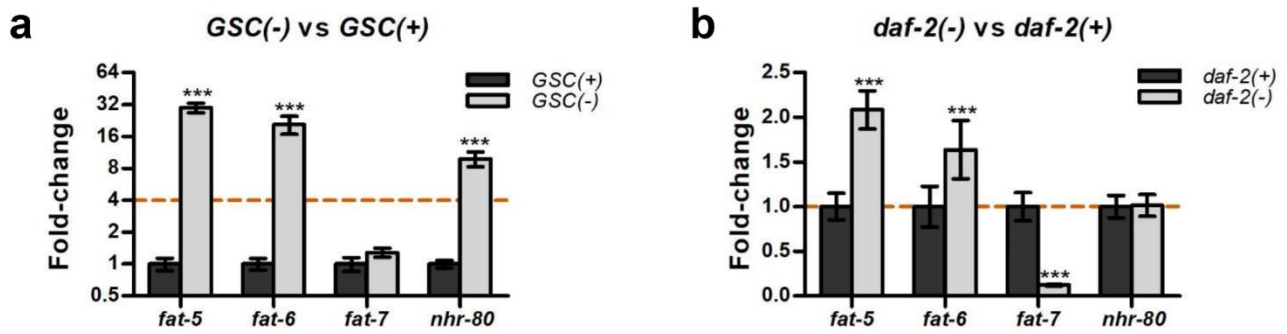


## Additional file 3

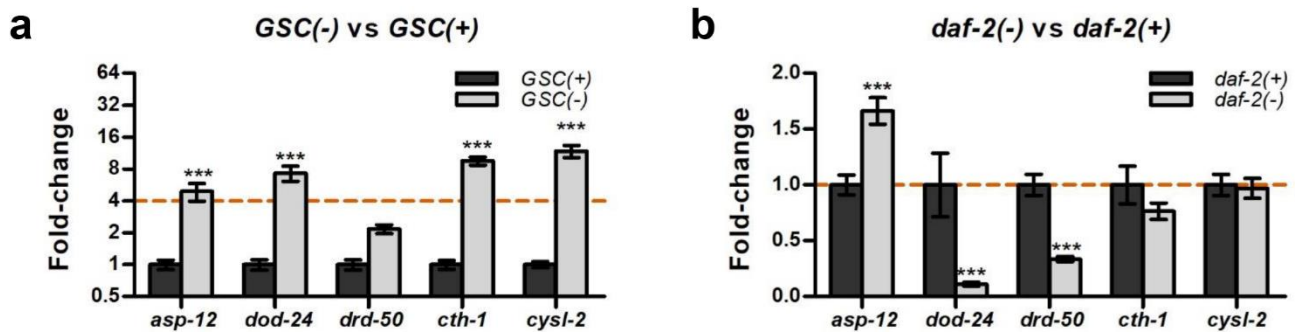
See main text for references.



**Fig. S1** Global changes in gene expression upon *daf-2* or GSC loss. Accompanies Fig. 2. **(a and b)** Differential gene expression between the strains indicated was determined by RNA-seq and visualized in MA-plots **(a)** and Volcano plots **(b)**. Light blue lines indicate  $|\text{Log}_2\text{FC}| > 0.58$  and  $p_{\text{adj}} < 0.05$ , open triangles indicate data points beyond axis limits. See Additional file 2: Table S9 for complete *DESeq2*-analysis results.



**Fig. S2** Influence of GSC and *daf-2* status on fatty acid desaturase gene-expression. Accompanies Fig. 3. Expression levels of the genes indicated were determined in GSC(-) relative to GSC(+) (**a**), and in *daf-2*(-) relative to *daf-2*(+) worms (**b**) by qPCR in day 2 adults. Data shown represent mean fold-changes  $\pm$  SEM from four biological replicates. Statistical significance of expression differences was determined by two-way ANOVA with Bonferroni post tests (\*  $p < 0.05$ , \*\*  $p < 0.01$ , \*\*\*  $p < 0.001$ ). Note that other authors [25] have proposed a fold-change of  $\sim 4$  for detecting genes differentially expressed between GSC(+) and GSC(-) worms, as they differ in the presence of a germline. The following gene expression changes (or lack of change) relative to the corresponding GSC(+)/*daf-2*(+) strain were also observed in published studies: GSC(-): induction of *fat-5*/*nhr-80* [25, 35], induction of *fat-6* [35], no upregulation of *fat-7* (our qPCR, [25]) or downregulation of *fat-7* (our RNA-seq, [35]); *daf-2*(-): induction of *fat-5* [26, 27] and *fat-6* [26], repression of *fat-7* [26, 27], no differential expression of *nhr-80* [26, 27].



**Fig. S3** Influence of GSC and *daf-2* status on the expression of genes involved in pathogen defense and H<sub>2</sub>S/HCN metabolism. Accompanies Fig. 4. Expression levels of the genes indicated were determined in GSC(-) relative to GSC(+) (a), and in *daf-2*(-) relative to *daf-2*(+) worms (b) by qPCR in day 2 adults. Data shown represent mean fold-changes  $\pm$  SEM from 3-4 biological replicates. Statistical significance of expression differences was determined by two-way ANOVA with Bonferroni post tests (\*  $p < 0.05$ , \*\*  $p < 0.01$ , \*\*\*  $p < 0.001$ ). Note that other authors [25] have proposed a fold-change of  $\sim 4$  for detecting genes differentially expressed between GSC(+) and GSC(-) worms, as they differ in the presence of a germline. The following gene expression changes (or lack of change) relative to the corresponding GSC(+)/*daf-2*(+) strain were also observed in published studies: GSC(-): significant induction of *asp-12* [25] (GSC(-) repressed genes not published in this study, thus it is unclear whether *dod-24*, *drd-50*, *cth-1* and *cysl-2* were not induced, induced  $< 4$ -fold, or even repressed by GSC loss); *daf-2*(-): significant induction of *asp-12*, significant repression of *dod-24* and *drd-50*, no differential expression of *cysl-2* [26, 27]; no differential expression of *cth-1* in [27], but repression in [26].