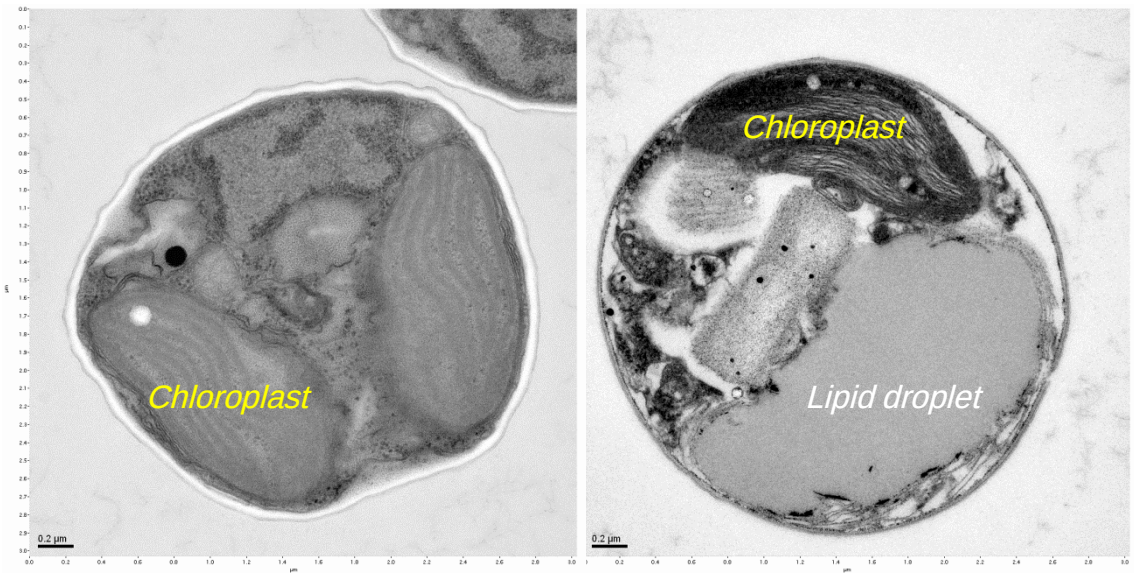

Figure S1. The ultrastructure of *N. oceanica* cells under nitrogen-replete (left) and nitrogen-depleted (right) conditions.

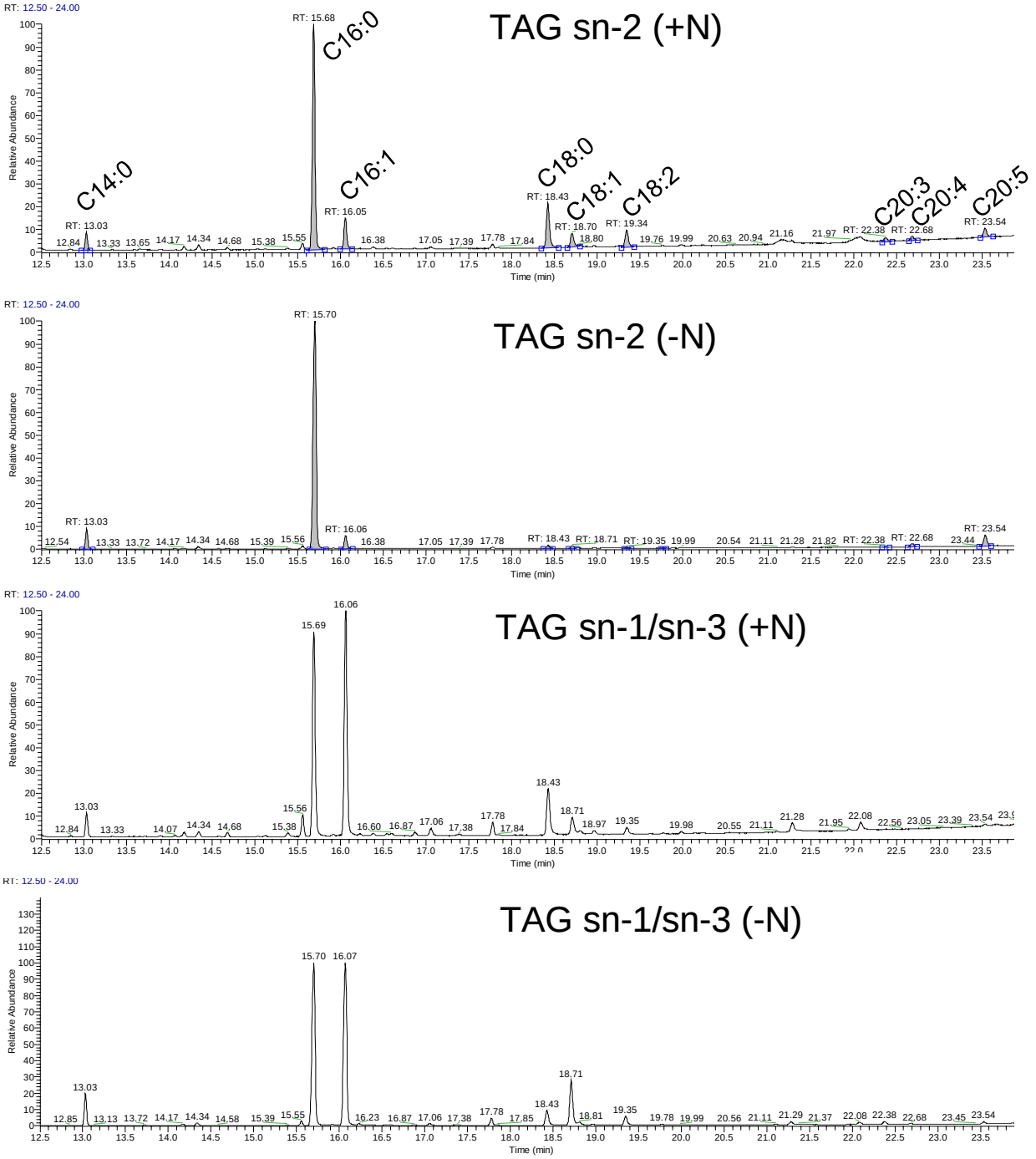

Figure S2. GC-MS chromatogram of fatty acids in the *sn*-2 and *sn*-1/*sn*-3 positions of TAG from *N. oceanica* cells under nitrate-replete (+N) and nitrate-depleted (-N) conditions

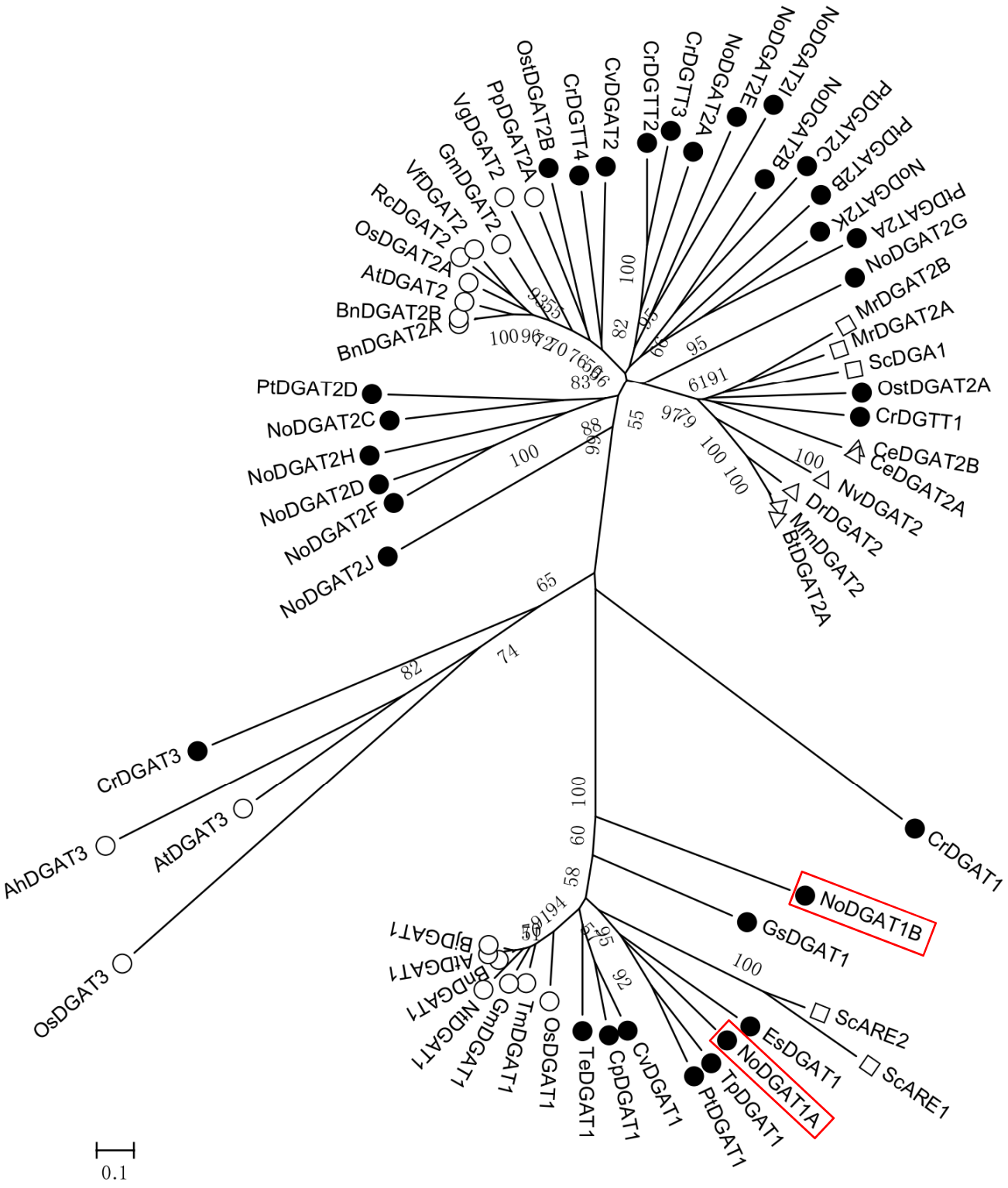

Figure S3. Cladogram of the DGATs from plants, fungi, algae and animals. The neighbor-joining method was used to reconstruct the cladogram using MEGA6 [34], with the bootstrap value (obtained from 1000 replicates) is shown on each node. The scale bar 0.1 represents 10% divergence, calculated as the estimated number of replacement. Circles, plants; Squares, fungi; Filled circles, algae; triangles, animals. Protein sequences used for the cladogram construction see Additional file 2: Table S1.

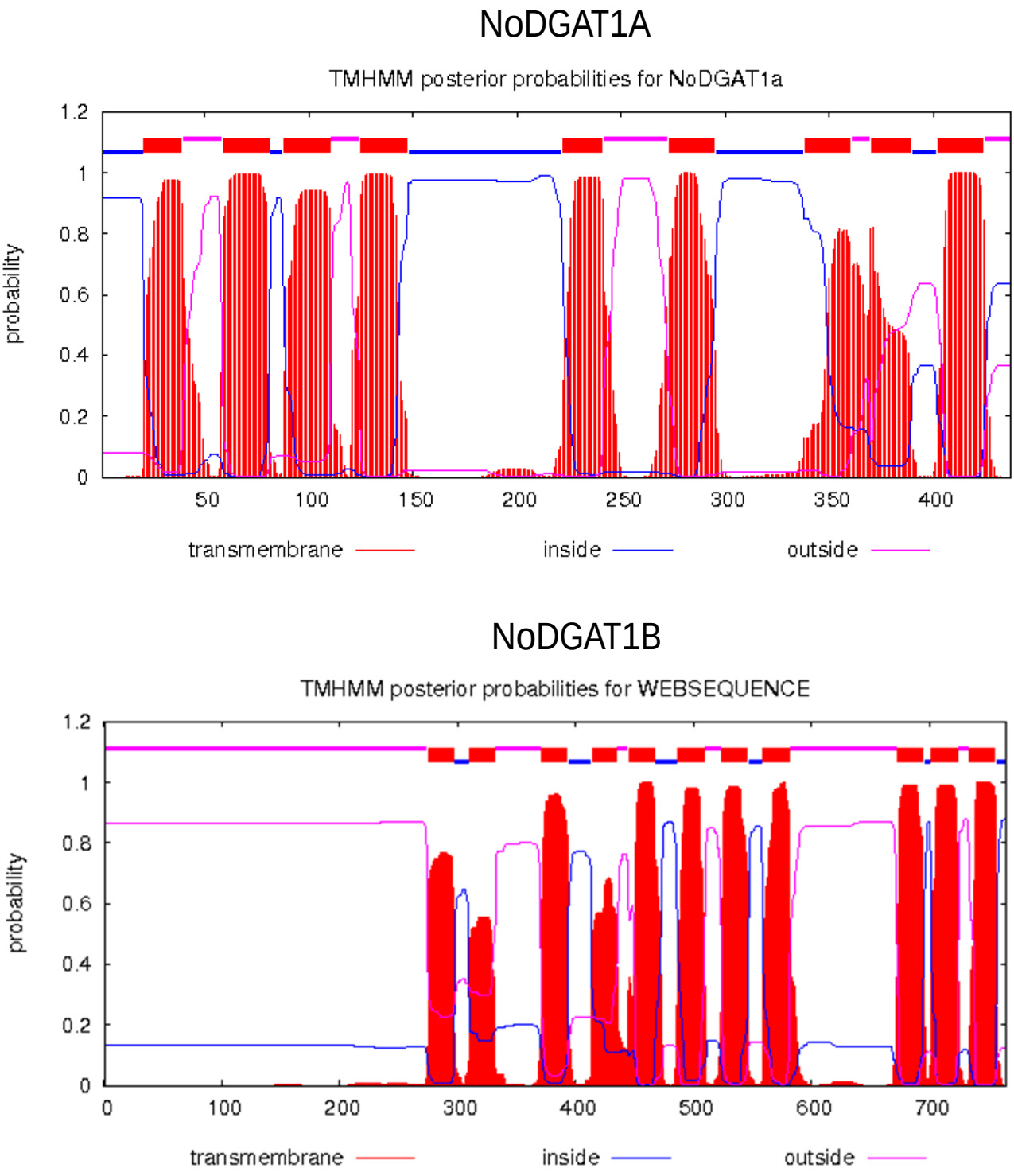

Figure S4 Predicated transmembrane domains for NoDGAT1A NoDGAT1B by TMHMM (V2.0, http://www.cbs.dtu.dk/services/TMHMM/).

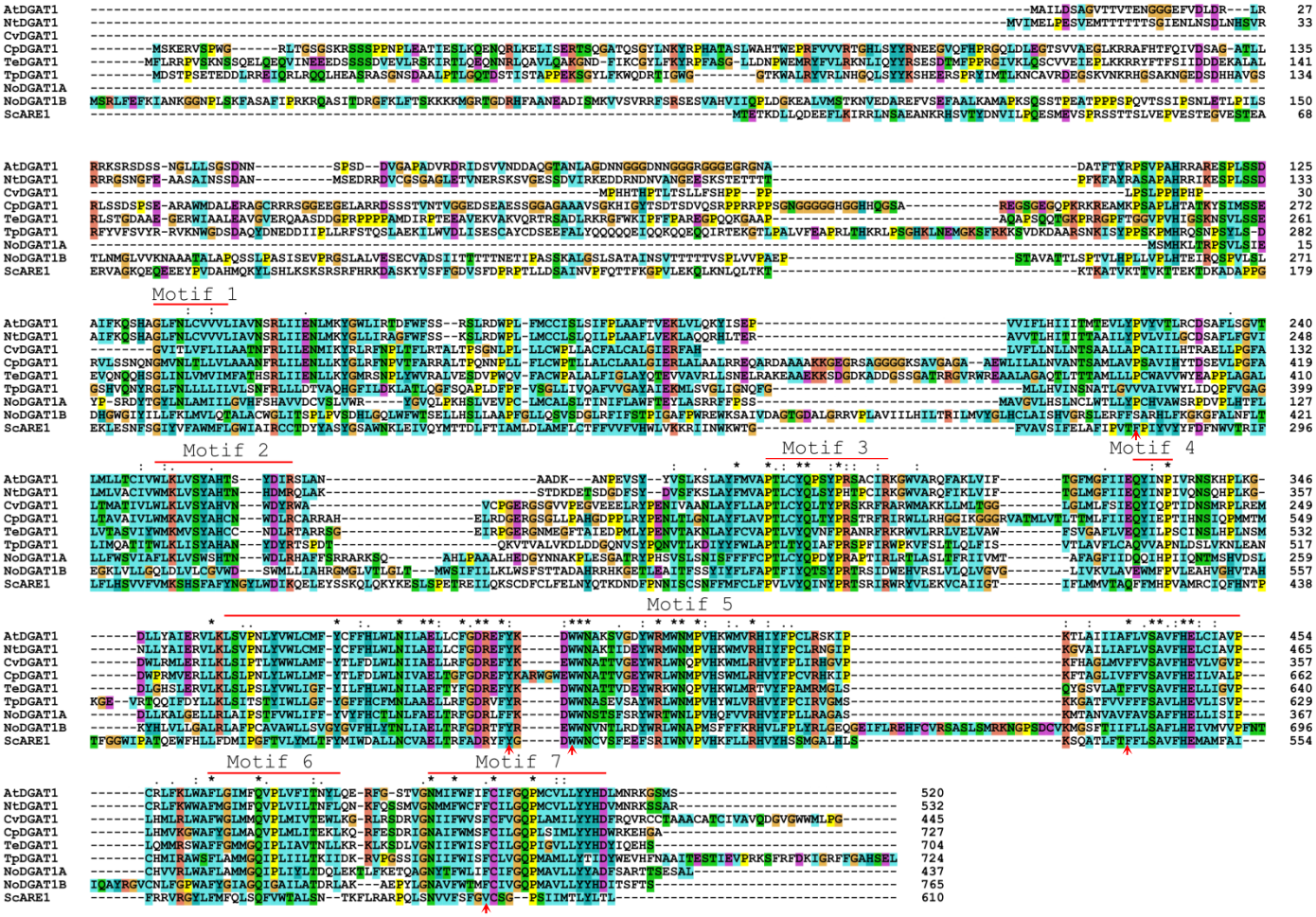

Figure S5 Protein sequence alignment of putative DGAT1s. The alignment was conducted using ClustalX2.1. The sequences used see Additional file 2: Table S1. Red arrows indicate the key amino acid residues identified by previous studies.

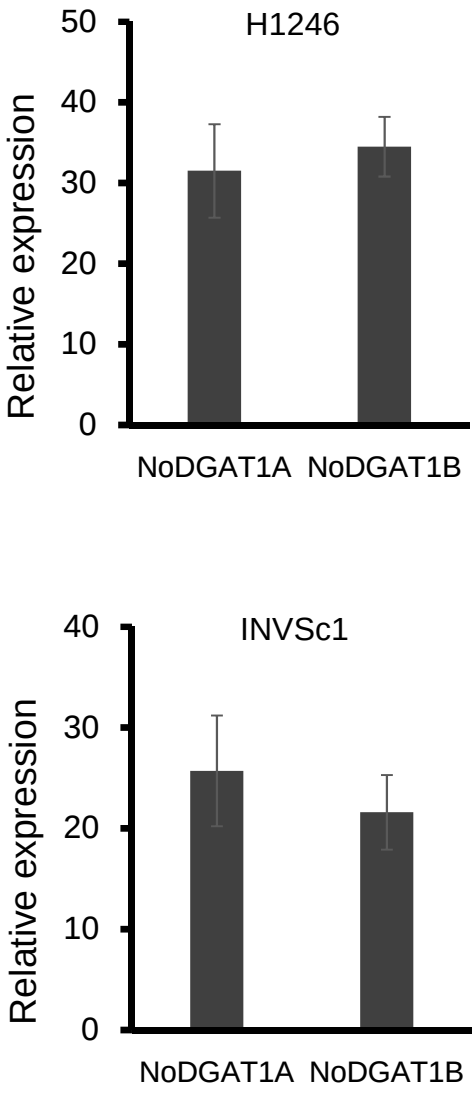

Figure S6. The transcriptional expression levels of *NoDGAT1A and NoDGAT1B* in H1246 (upper) and INVSc1 (lower), as determined by quantitative real-time PCR. The gene expression levels were normalized to the endogenous *ACT1* gene.

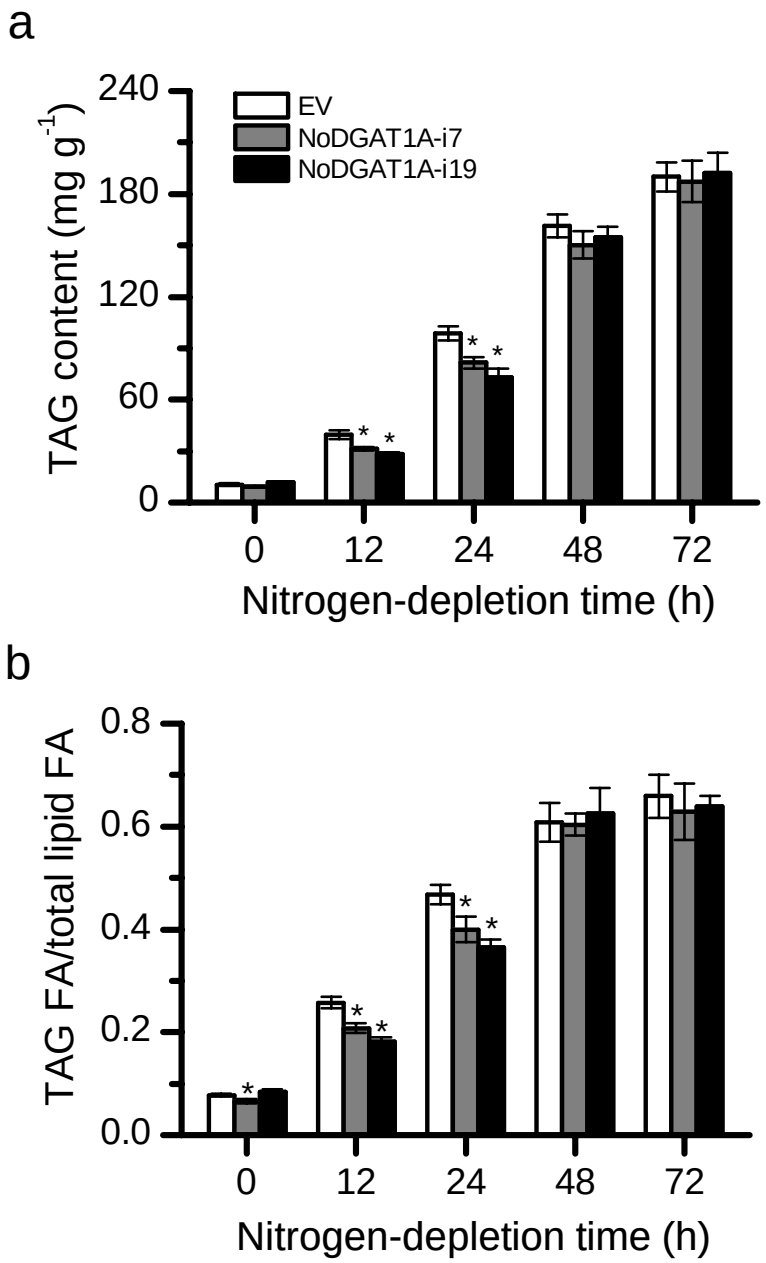

Figure S7. TAG content per dry weight (**a**) and the ratio of fatty acids in TAG over TFA (**b**) in *NoDGAT1A* knockdown lines and EV. Algal cells grown in nitrogen-replete medium for 4 days (considered as 0 h of nitrogen depletion) were used for nitrogen depletion experiment. Data are expressed as mean ± SD (n=3). Asterisks indicate the significant difference compared with EV (t-test, P<0.05).

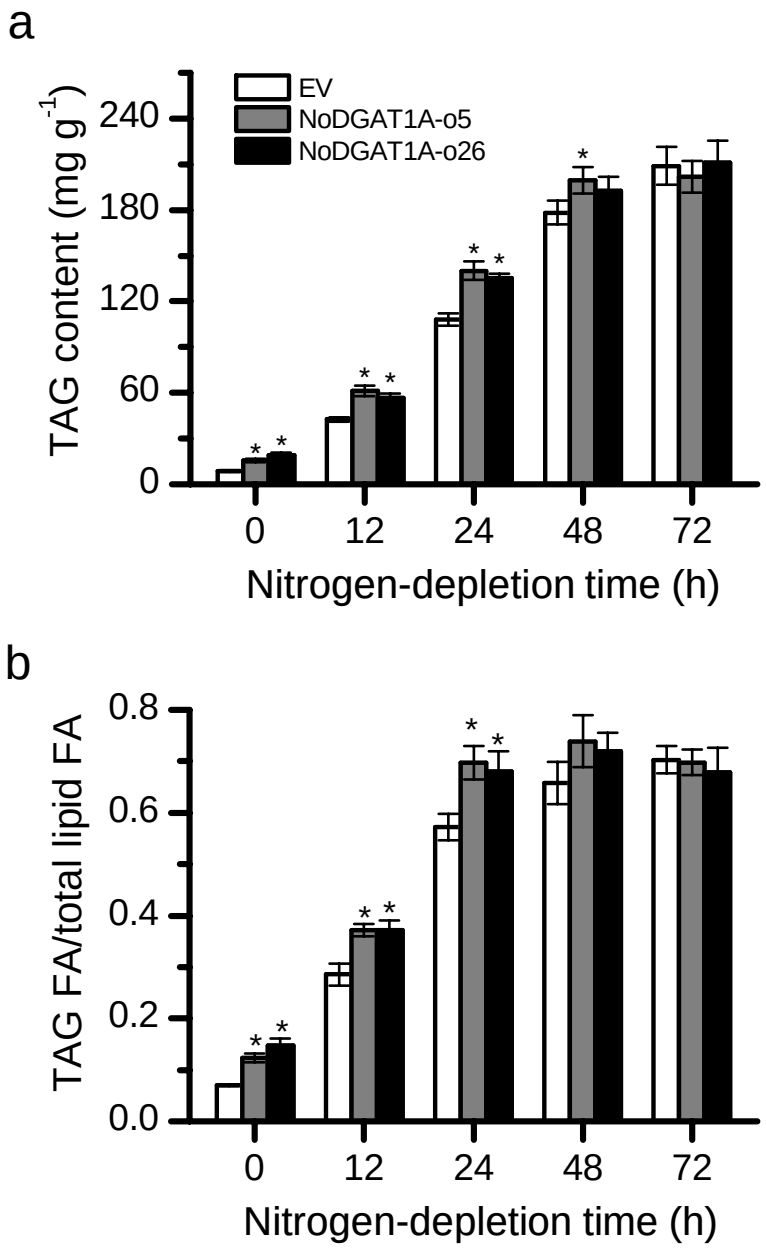

Figure S8. TAG content per dry weight (**a**) and the ratio of fatty acids in TAG over TFA (**b**) in *NoDGAT1A* overexpression lines and EV. Algal cells grown in nitrogen-replete medium for 4 days (considered as 0 h of nitrogen depletion) were used for nitrogen depletion experiment. Data are expressed as mean ± SD (n=3). Asterisks indicate the significant difference compared with EV (t-test, P<0.05).

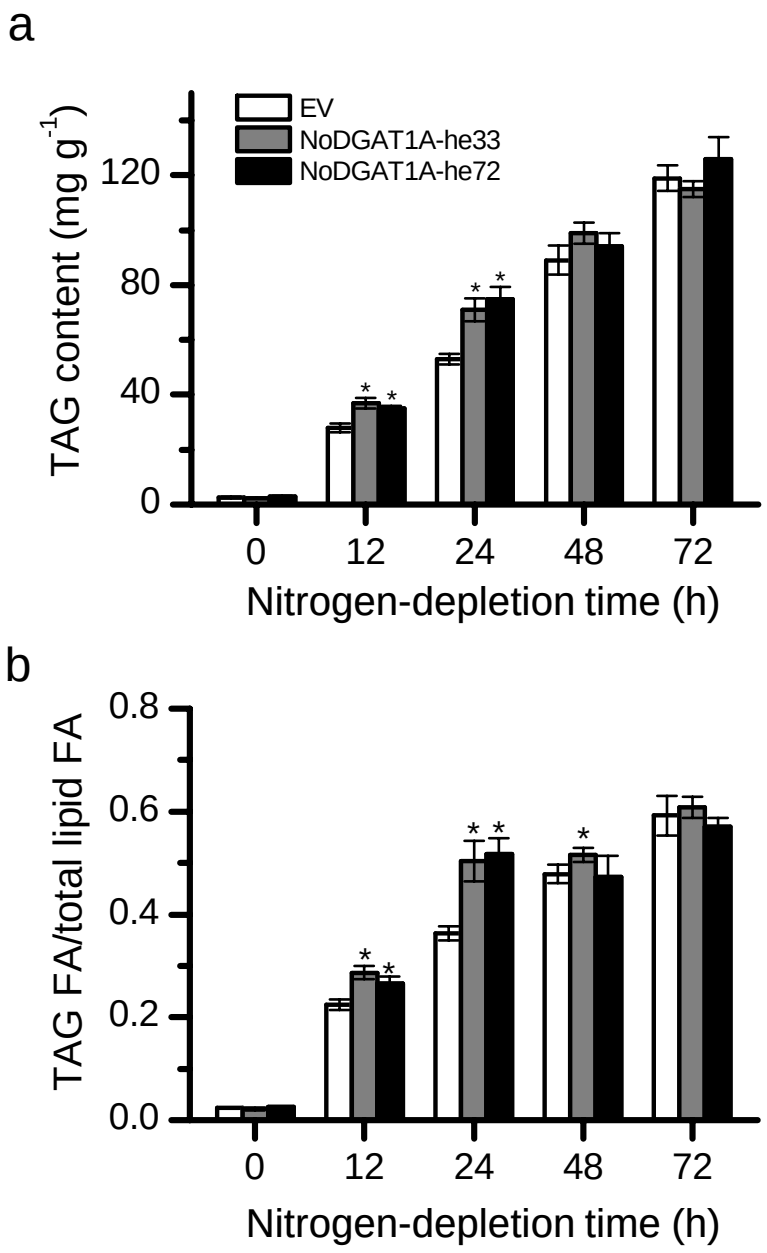

Figure S9. TAG content per dry weight (**a**) and the ratio of fatty acids in TAG over TFA (**b**) in *NoDGAT1A* heterologous expression lines of *Chlamydomonas* and EV. Algal cells grown in nitrogen-replete medium for 4 days (considered as 0 h of nitrogen depletion) were used for nitrogen depletion experiment. Data are expressed as mean ± SD (*n*=3). Asterisks indicate the significant difference compared with EV (*t*-test, P<0.05).

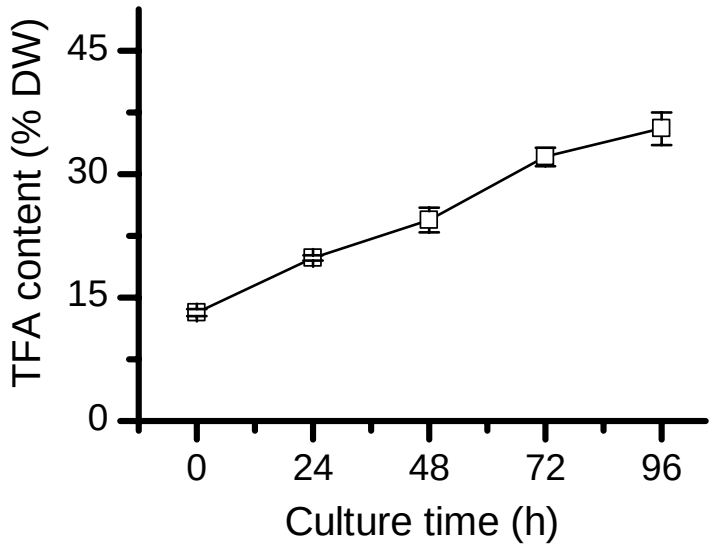

Figure S10. Time course of total fatty acid (TFA) content of *N. oceanica* in response to nitrogen depletion.

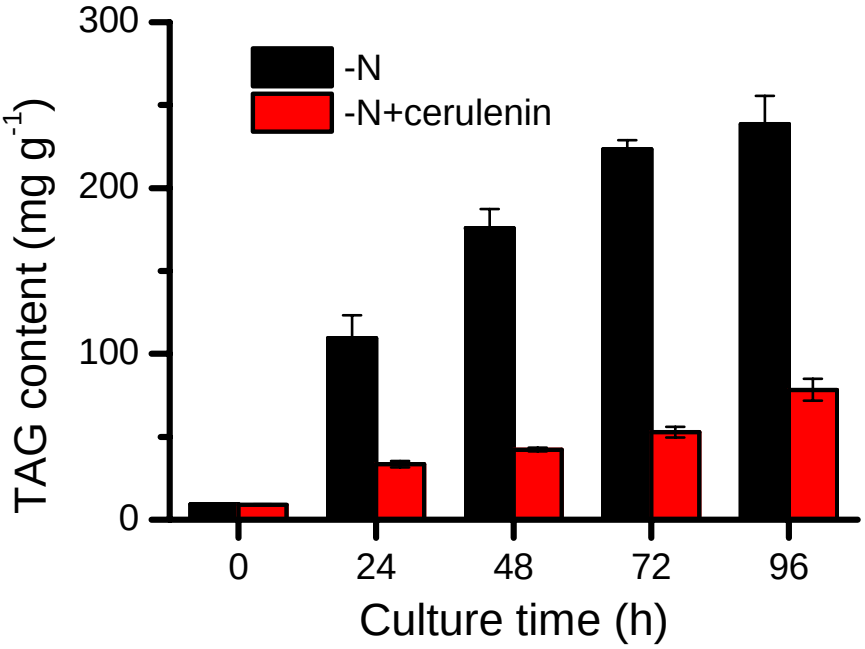

Figure S11. Effect of cerulenin on TAG content of *N. oceanica* in response to nitrogen depletion. The cerulenin concentration used was 10 μM.

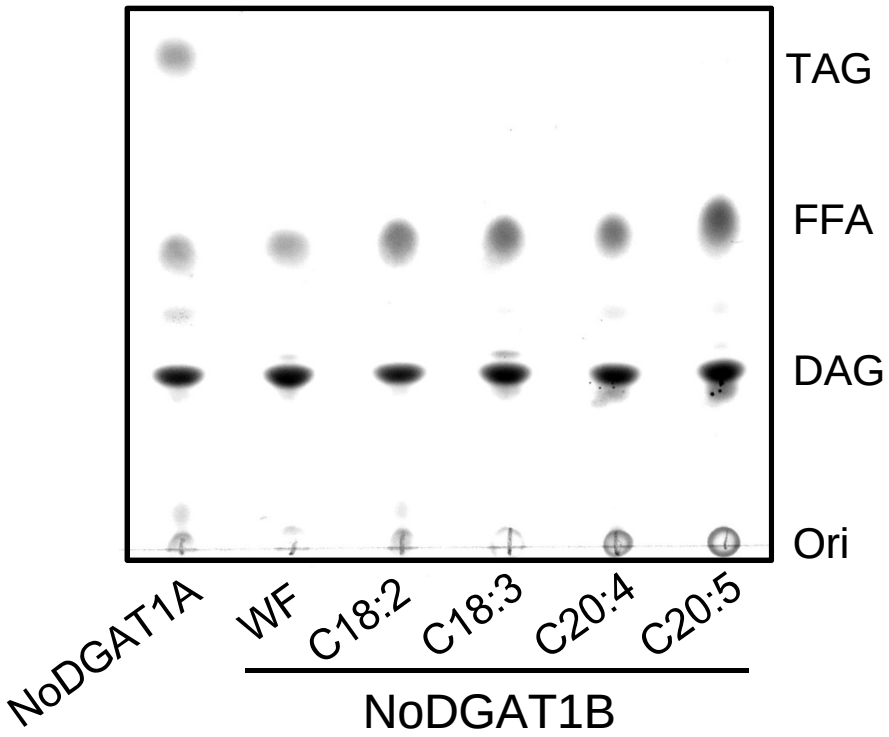

Figure S12. TLC analysis of lipids extracted from *NoDGAT1B*-carrying H1246 cells without feeding (WF) or fed with free fatty acids of C18:2, C18:3, C20:4, or C20:5 (125 μM). *NoDGAT1A*-carrying H1246 cells were used as the positive control.

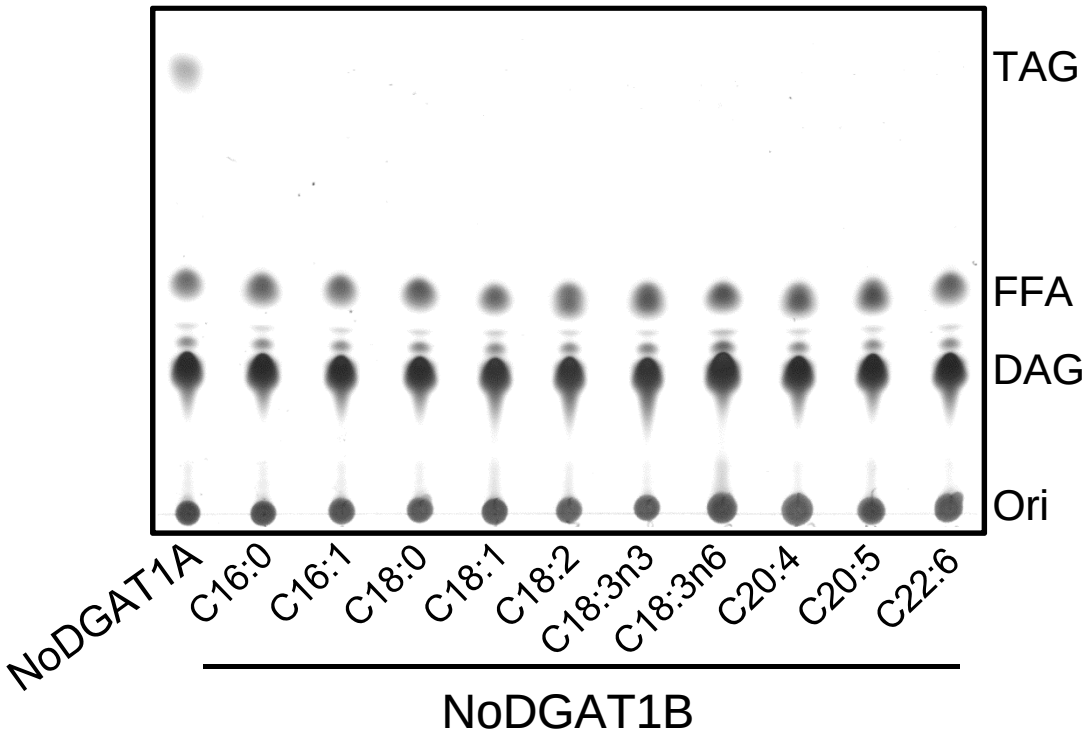

Figure S13. TLC analysis of lipids resulting from *in vitro* enzymatic reactions of NoDGAT1B with various acyl-CoAs. C18:1/C16:0-DAG was used as the acyl acceptor; NoDGAT1A was used as the positive control (C16:0 as the acyl donor).

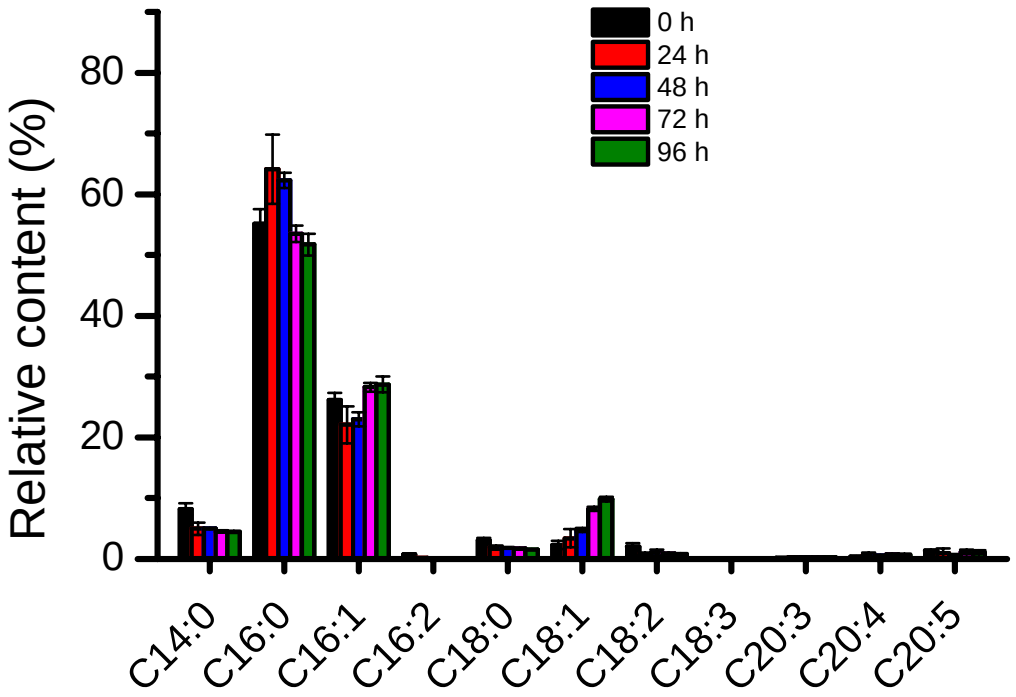

Figure S14. Fatty acid composition of TAG in *N. oceanica* upon nitrogen depletion

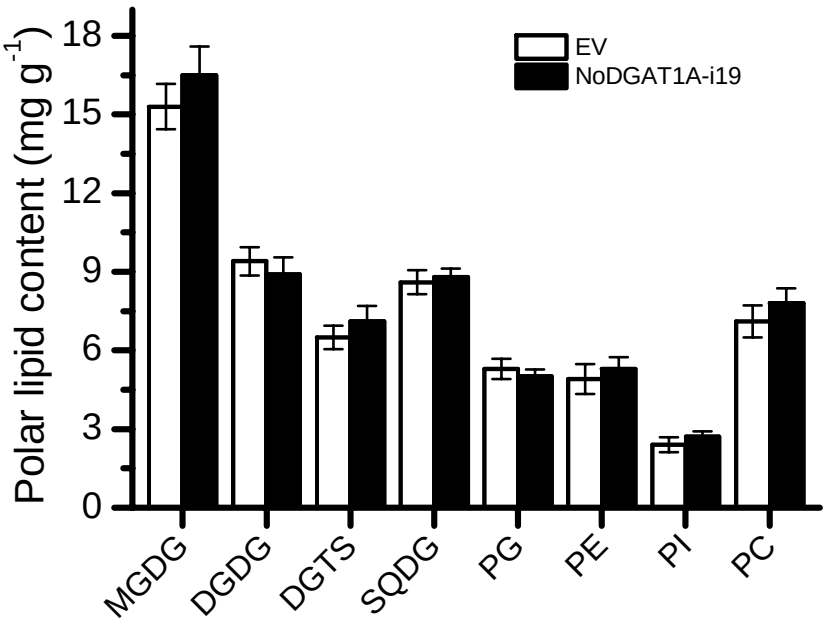

Figure S15 The contents of polar lipids in EV and NoDGAT1A-i19 under nitrogen-depleted conditions (24 h).

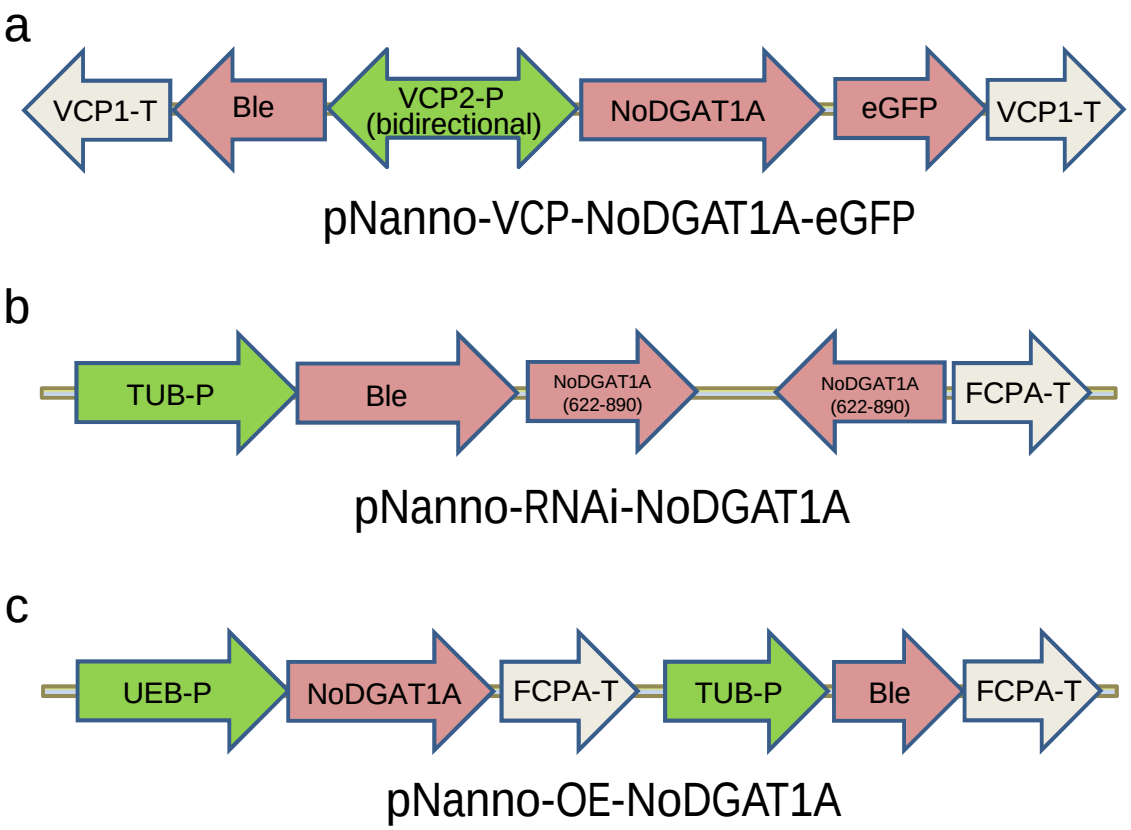

Figure S16 Schematic illustration of constructs for subcellular localization (**a**), knockdown (**b**), and overexpression (**c**) of NoDGAT1A in *N. oceanica* cells.