

Figure 1. Partial physical map of the 15.8-kb mitochondrial genome of *C. reinhardtii*

The rectangles represent protein-coding genes: *cob*, gene encoding apocytochrome b of complex III; *nd1*, 2, 4, 5, and 6, genes encoding the corresponding subunits of complex I; *cox1*, gene encoding the subunit 1 of complex IV; *rtl*: reverse transcriptase-like protein. The position of the three mitochondria-encoded tRNA genes (W: tRNA^{Trp}; Q: tRNA^{Gln}; M: tRNA^{Met}) are indicated in red. The bidirectional origin of transcription between *nd5* and *cox1* by is represented in arrows. Positions of the *dum11* and *dum22* deletions and of the *dum18* point mutations is indicated.

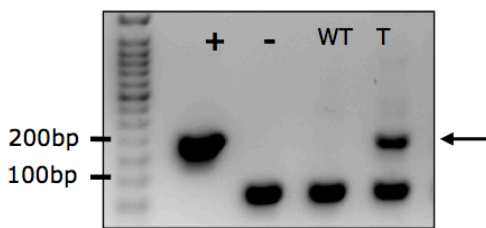


Figure 2. Screening of transformants

Detection of the tagged tRNA^{Val}(UAC) by PCR amplification was performed with primers upstream and downstream of the insertion, (+) and (-) represent PCR positive and negative controls respectively. (WT) corresponds to wild-type strain and (T) corresponds to the transformant with the inserted tRNA.

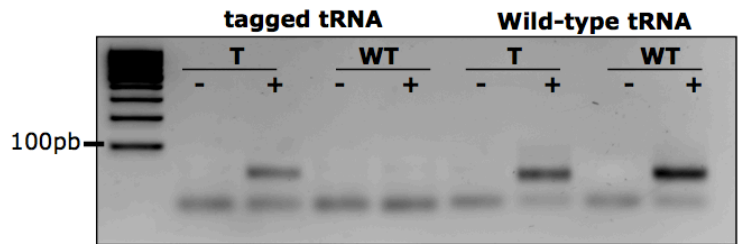


Figure 3. The inserted tagged tRNA^{Val}(UAC) is expressed

Total tRNAs were extracted and ligated. Circularized products were then analyzed by cRT-PCR. The presence (+) or the absence (-) of reverse transcriptase during the cDNA synthesis in the presence of the primer specific to the tagged tRNA or wild-type tRNA are indicated.

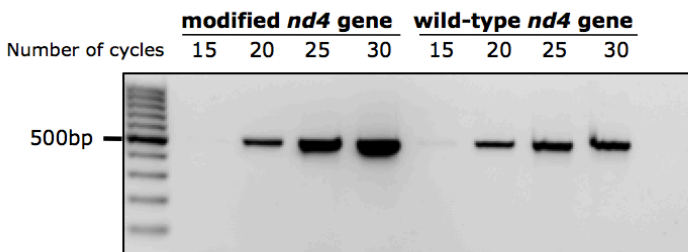


Figure 4. Heteroplasmic mutant

An example of semi-quantitative PCR performed with primer specific to the modified *nd4* gene and to the *nd4* gene on the mutant with 6 changes in the *nd4* gene. The number of cycles of PCR are indicated.

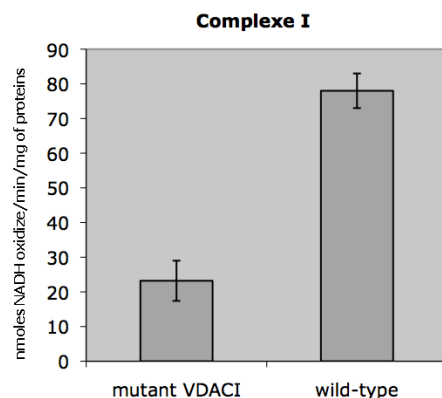


Figure 5. Mitochondrial specific respiratory enzyme activities of mutant VDAC1 and wild-type

Specific activities from three experiments were measured in crude membrane fractions.