## Supplemental data

**Supplemental Figure S1.** Effects of extended dark exposure on photosynthetic parameters in Col and *atpc2*, lacking the ATP synthase  $\gamma_2$  protein.

**Supplemental Figure S2.** The effects of reduced ATP synthase activity  $(g_{H+})$  but not redox regulation on the stability of maximal PSII quantum efficiency during extended dark exposure.

**Supplemental Figure S3.** The effects of extended dark exposure on the expression of senescence-related genes and chlorophyll content.

**Supplemental Figure S4.** Expression of representative photosynthesis-related genes under extend dark conditions at 0, 2 and 4 days in wild-type (Ws) and *gamera*-1.

Supplemental Figure S5. Visualization of leaf starch accumulation and breakdown.

Supplemental Table S1. Gene Specific Primer sets used for RT-PCR.



**Supplemental Figure S1.** Effects of extended dark exposure on photosynthetic parameters in Col and *atpc2*, lacking the ATP synthase  $\gamma_2$  protein. Extended dark exposure and photosynthetic measurements were carried as in Main Text Fig. 1, out for 4 days in wild-type (Ws) (black circles), and *atpc2* (white squares). (A) Spectroscopy analysis were performed at different actinic light intensities – dependence of  $F_V/F_M$ , (B) Linear electron flow (LEF), (C) proton conductivity across the thylakoid membrane (g<sub>H+</sub>) based on ECS decay kinetics. Data presented was the average and standard deviation of results from experiments on 4-5 biological replicates.



**Supplemental Figure S2.** The effects of reduced ATP synthase activity ( $g_{H+}$ ) but not redox regulation on the stability of maximal PSII quantum efficiency during extended dark exposure. A construct containing a single point mutation between *ATPC1/ATPC2*, P194M (*minira-2*), was made and used to complement *dpa1*, which lacks *ATPC1*. The steady-state ATP synthase activity as estimated by the dark interval relaxation kinetics of the ECS (Ws-gray bar and *minira-2* white bar) showed a decrease of 41% activity from Ws and *minira-2* at 200 µmol photons m<sup>-2</sup> s<sup>-1</sup> PAR (**A**), similar to the loss of ATP synthase activity in *gamera* (Kohzuma et al. 2012). However, the decay of the flash-induced ECS signals increased upon exposure from 1 (red bar) to 60 (black bar) min of darkness (**B**), indicating wild type-like redox regulation of the ATP synthase (Kramer and Croft 1989). Measurements of F<sub>V</sub>/F<sub>M</sub> were taken once a day during three days of continuous darkness in a DEPI chamber using chlorophyll visual imaging (C) and quantified as described in Figure 1 of the main text (d, n=3). No differences were seen in the loss of F<sub>V</sub>/F<sub>M</sub> values over three days in darkness indicating that loss of ATP synthase activity by itself does not alter the decay of PSII during extended darkness (**D**).



**Supplemental Figure S3.** The effects of extended dark exposure on the expression of senescence-related genes and chlorophyll content. Wild-type (Ws) and *gamera*-1 plants were treated as in Fig. 1 and assayed after 0, 2 and 4 days of darkness. (**A**) RT-PCR was performed to monitor expression levels of two classical senescence marker genes, *SAG12* and *SEN1*, as well as two photosynthetic genes, *RBCS2B* and *CAB2B*. 18s rRNA expression level was used as an internal loading control. For more details regarding RT-PCR conditions see Materials and Methods. (**B**) Chlorophyll content was extracted and determined from 6mm diameter leaf discs derived from wild-type and *gamera* plants at 0 day (gray bar) and after 4 days of dark adaption period (dark bar). Reported are averages and standard deviations of 4 biological replicates.



**Supplemental Figure S4.** Expression of representative photosynthesis-related genes under extend dark conditions at 0, 2 and 4 days in wild-type (Ws) and *gamera*-1. RT-PCR was performed using gene specific primers (Primers sets are listed in Supporting Table S1). PCR amplification conditions were determined empirically for each template-primer pair. 18s rRNA was used as an internal standard. For more details regarding RT-PCR conditions see Materials and Methods. Similar results were obtained among three independent experiments.



**Supplemental Figure S5.** Visualization of leaf starch accumulation and breakdown. Plants grown under a 16 hr/8 hr light cycle were stained with iodine to visualize starch accumulation 1-hour before the end of the light cycle (the end of the day) in Ws (**A**) and *gamera* (**B**). Starch breakdown was similarly visualized 1-hour before the end of the dark cycle (the end of the night) in Ws (**C**) and *gamera* (**D**). The lack of iodine staining in the end of the night samples is expected due to the normal breakdown of starch reserves by the end of the normal night period.

## Supplemental Table S1. Gene Specific Primer sets used for RT-PCR.

RNA was isolated from leaf tissue derived from wild-type and *gamera* plants incubated for extended dark conditions at 0, 2 and 4 days using an RNeasy Plant Mini kit according to protocol supplied by the manufacturer (Qiagen). Isolated RNA was quantitated using Aligent 2100 bioanalyzer. First-strand cDNA was synthesized from 1µg of total RNA using SuperScript<sup>TM</sup>III Reverse Transcriptase according to the manufacturer's protocol (Invitrogen<sup>TM</sup>). PCR reactions were performed using various gene specific primers (Table S1) while PCR amplification conditions were determined empirically for each template-primer pair (Supplemental Figure. S3 and Supplemental Figure. S5).

Gene Name	At ID#	Primer Direction	Sequence	Fragment Length (bp)
SAG12	At5g45890	Forward	5'-GATGAAGGCAGTGGCACACCAA-3"	333
	-	Reverse	5'-TCCCACACAAACATACACAATTAAAAGC-3'	
SENI	At4g35770	Forward	5'-ATCACGAATTGGAAACTGG-3'	133
	-	Reverse	5'-CTTTCCTCCATCGGAAG-3'	
RBCS2B	At5g38420	Forward	5'-ACCTTCTCCGCAACAAGTGG-3'	256
	C	Reverse	5'-GTGAAGCTTGGGGGGCTTGTAGG-3'	
CAB2B	At1g29920	Forward	5'-TTGAAGGCTACAGAGTCGCAGGAAA-3'	290
	-	Reverse	5'-CACTCACGAAGCAAAGACTGAAGCA-3'	
18srRNA		Forward	5'-AACGGCTACCACATCCAAG-3'	442
		Reverse	5'GAAGCCAACACAATAGGAT-3'	
ATPB	AtCg00480	Forward	5'-GTAGCAAGAGCACGAAAAAT-3'	210
		Reverse	5'-TAAGTTCGTAGCCTTCGCAG-3'	
PSBA	AtCg00020	Forward	5'-CAATATGCTAGTTTCAACAA-3'	200
	-	Reverse	5'-GTTCATGCATAACTTCCATA-3'	
OEC17	At4g05180	Forward	5'-TAGATTTTACATACAACCAT-3'	250
	-	Reverse	5'-GTCCAAGTTGTCAATGGTTT-3'	
OEC23	At2g30790	Forward	5'-AGCAACTGCAAACATTTTGG-3'	150
		Reverse	5'-ATGTAAAGCTTTCCACCATT-3'	
OEC33	At5g66570	Forward	5'-ACTTCCTTCACCGTCAAGGC-3'	200
		Reverse	5'-TGGAAGTTGGACTGTGACTG-3'	
PETA	AtCg00540	Forward	5'-ATGGACGCGAAGTTATTGAT-3'	240
		Reverse	5'-AAGTACTATTTCCGCATCCC-3'	
PETC	At4g03280	Forward	5'-AGGGAGATCCGACTTACCTA-3'	200
	-	Reverse	5'-AGCCAACGCTAGCGACAATG-3'	
PSAF	At1g31330	Forward	5'-GTTTACCGCACTTGATAGTG-3'	200
	-	Reverse	5'-ACCACGGAAGATGATCCGAC-3'	
PSAD2	At1g03130	Forward	5'-AAGTACAAGATCACTTACCA-3'	180
		Reverse	5'-TTGTTTCCCAGTGAATTTAA-3'	