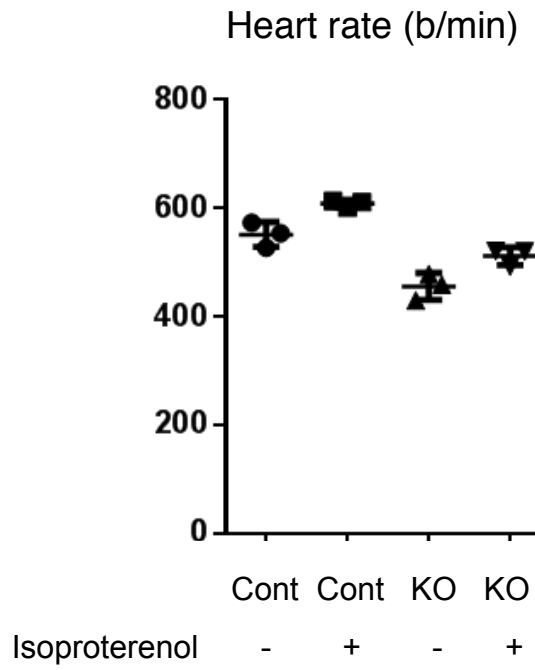
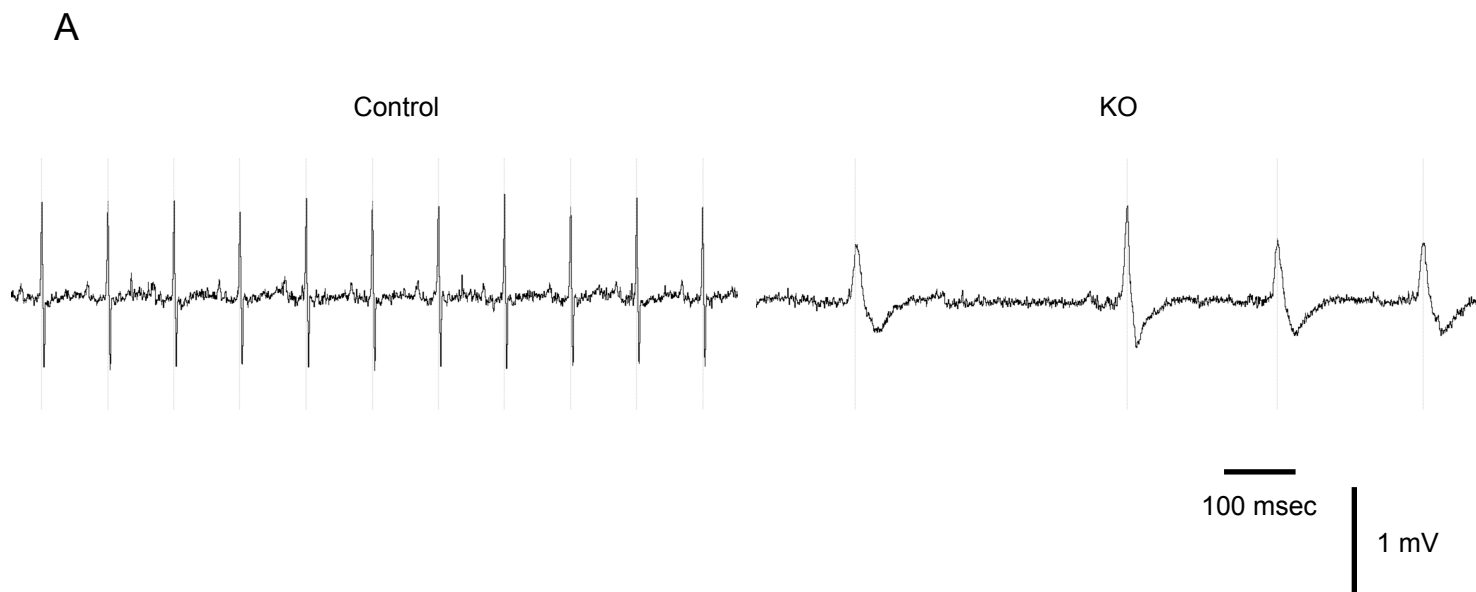


Supplementary Figure 1. The levels of Drp1 in P1 hearts

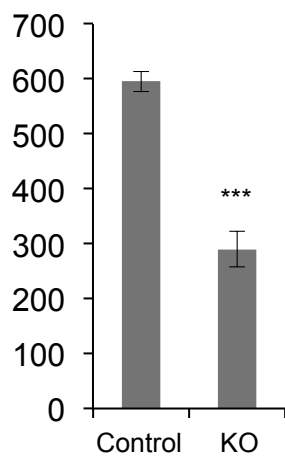
Immunoblotting of hearts isolated from P1 control, Het and Myh6-Drp1KO (KO) mice was performed using antibodies to Drp1 and actin.



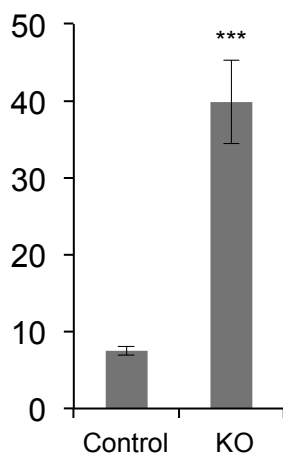
Supplementary Figure 2. Isoproterenol (4mg/kg of body weight) was administered with an intraperitoneal injection to control and Myh6-Drp1KO mice at P7. Electrocardiography was performed before and after the isoproterenol injection.



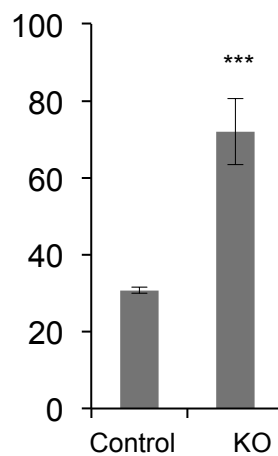
B
Heart rate (b/min)



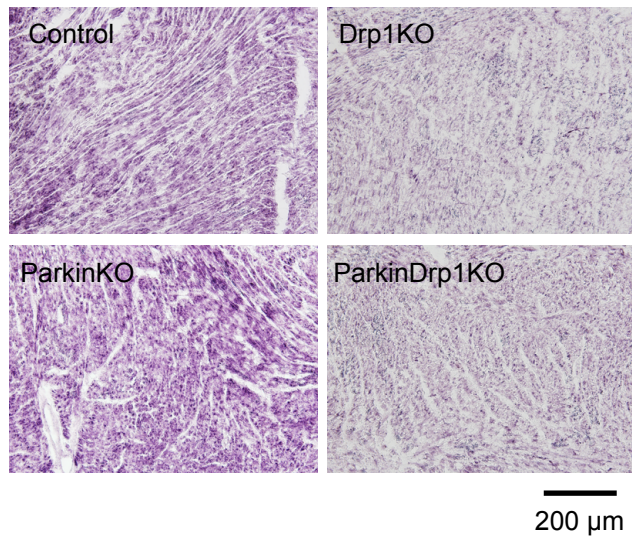
C
QRS complex (msec)



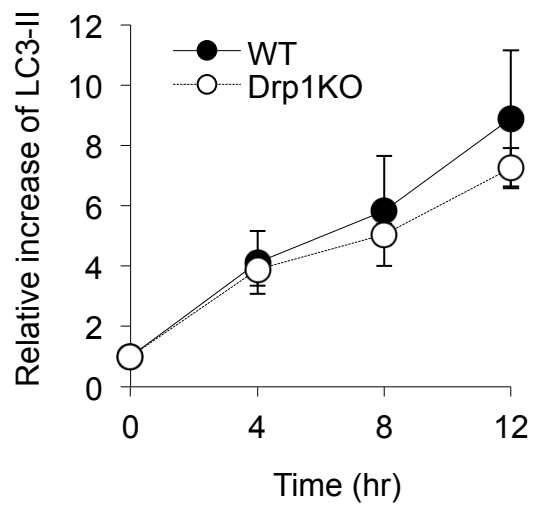
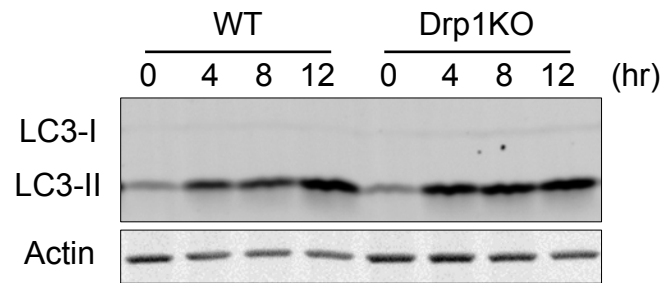
D
PR interval (msec)



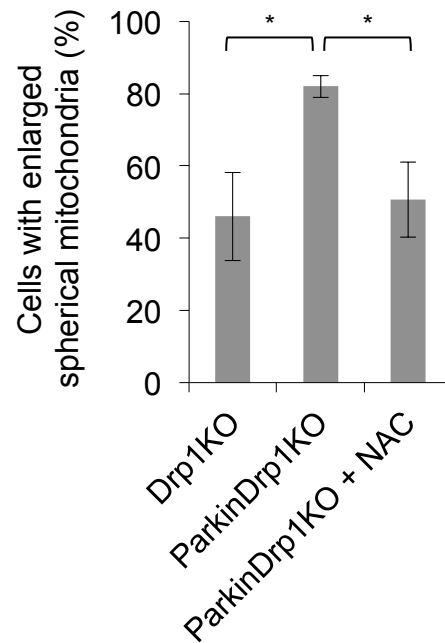
Supplementary Figure 3. Electrocardiogram (EKG) was performed in awake control and Myh6-Drp1KO mice around P11. Representative EKG tracings for each genotype are shown. P wave was not detected in most Myh6-Drp1KO mice (5 out of 8 animals examined). (B and C) EKG was used for calculating the heart rate (B) QRS complex (C) and PR interval (D). Values are mean \pm SEM. 7 control and 8 KO mice were analyzed.



Supplementary Figure 4. Fresh frozen sections of heart from P7 control, Myh6-Drp1KO (Drp1KO), ParkinKO and ParkinDrp1KO mice were histologically stained for the activity of NADH dehydrogenase (complex I).



Supplementary Figure 5. WT and Drp1KO MEFs were incubated with 20 nM bafilomycin A for the indicated amount of time. Whole-cell lysates were analyzed by immunoblotting with antibodies to LC3 and actin. Band intensity of LC3-II was quantified and normalized to that of actin. Values represent the mean \pm SEM (n=3).



Supplementary Figure 6. Drp1KO and ParkinDrp1KO MEFs were generated by infecting Drp1flox/flox and Drp1flox/flox::ParkinKO MEFs with lentiviruses carrying Cre recombinase. We confirmed that the efficiency of transduction is virtually 100%. At the time of infection, 1 mM N-acetylcysteine was added to the culture medium. The culture medium, supplemented with 1 mM N-acetylcysteine, was changed each day for 1 week. MEFs that contained enlarged mitochondria were scored using immunofluorescence microscopy with antibodies to PDH and Tom20. Values represent the mean \pm SEM (n=3). Fifty cells were examined in each experiment.