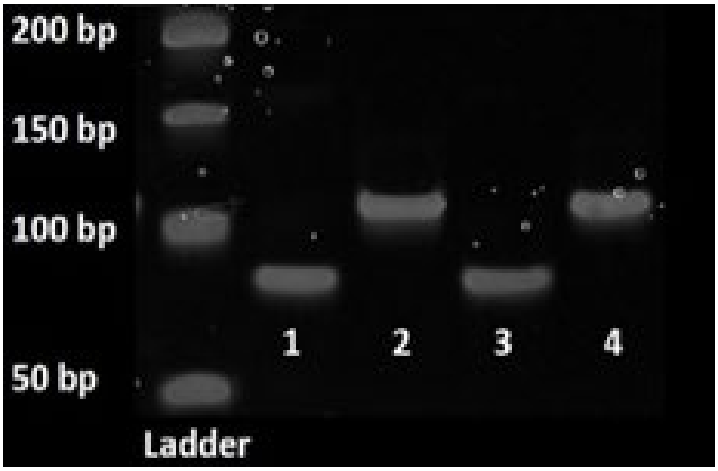
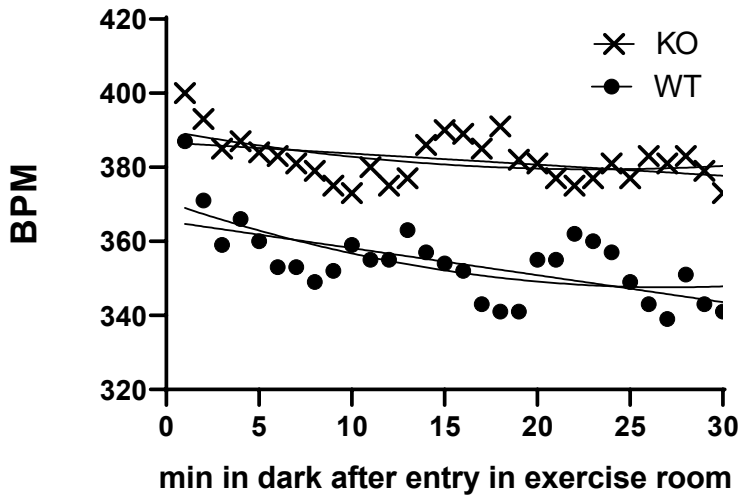
Supplementary Material

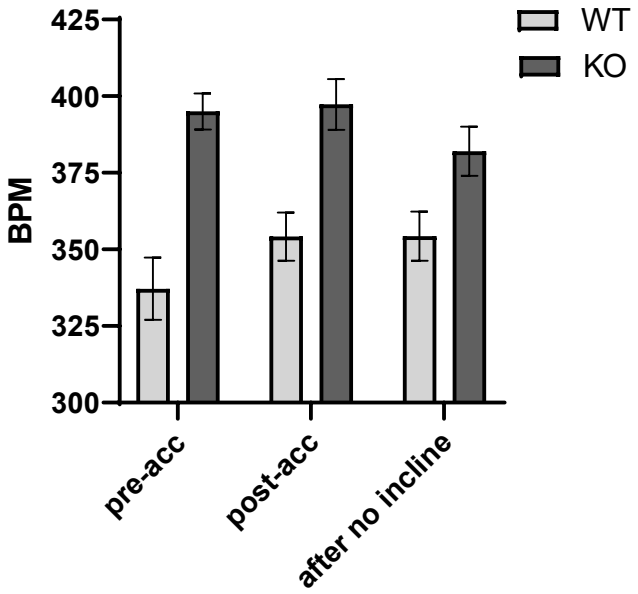
Establishing Equivalent Aerobic Exercise Parameters Between Early-Stage Parkinson's Disease and Pink1 Knockout Rats



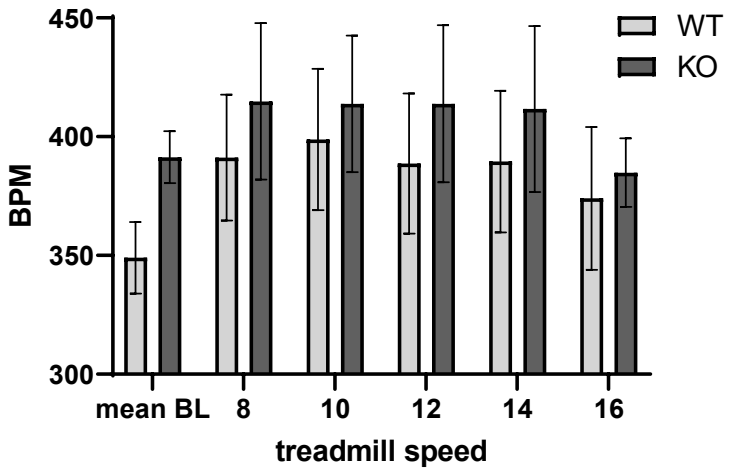
Supplementary Figure 1. Verifying Pink1 KO 5% TBE electrophoresis of *PINK1* and wild-type PCR products of cerebellar extract. Far left lane: DNA Ladder. Lanes 1, 3: Pink1 KO rats, Lanes 2, 4: WT rats. Gene fragment length is 109 bp in the WT and 83 bp in KO, reflecting deletion of 26 bp.



Supplementary Figure 2. Influence of relocation to exercise room on HR. HR was recorded within 5 min after relocation of rats from the home colony to the exercise room to determine if the exercise environment affected HR. Evidence for increased HR was seen in both genotypes. Linear regression analysis indicated significantly non-zero slopes for both genotypes; WT, ($F_{(1,28)}$ =18.79, p=0.0002), Pink1 KO (KO) ($F_{(1,28)}$ =6.01, p=0.021). Similar to the pre- and post-exercise acclimation phases (Fig. 2A), comparison of the mean BPM for the 30-min period revealed greater BPM in the KO compared to WT (t=2.44, p=0.035)

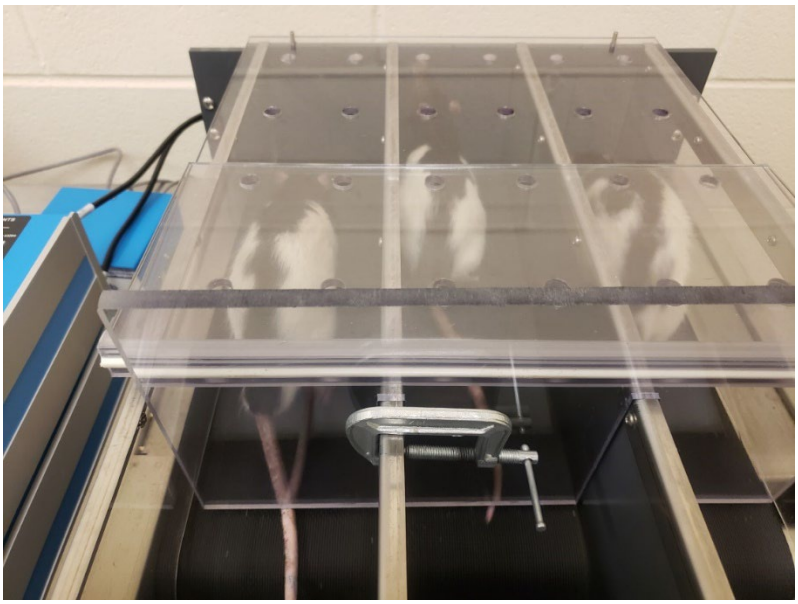


Supplementary Figure 3. Pre-exercise baseline determination. To determine a baseline HR unaffected by anticipation of exercise, we evaluated genotype and across three phases during the study. Whereas there was a strong influence of genotype, there was no significant influence of when the HR was collected across these experimental phases. Genotype ($F_{(1,10)} = 30.51$, p=0.0003); phase ($F_{(2,19)} = 0.99$, ns), genotype x phase ($F_{(2,19)} = 2.11$, ns).

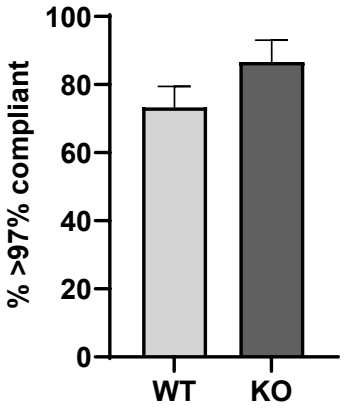


Supplementary Figure 4. Influence of exercise anticipation on HR. Immediately prior to placement on the treadmill at any speed interval, HR was greater than mean HR levels obtained at three different time points temporally dissociated from the exercise sessions (mean BL). Mean BL ($F_{(5,50)} = 9.69$, $p < 0.0001$), genotype ($F_{(1,10)} = 3.08$, *ns*); genotype x exercise session $F_{(5,50)} = 1.29$, ns).

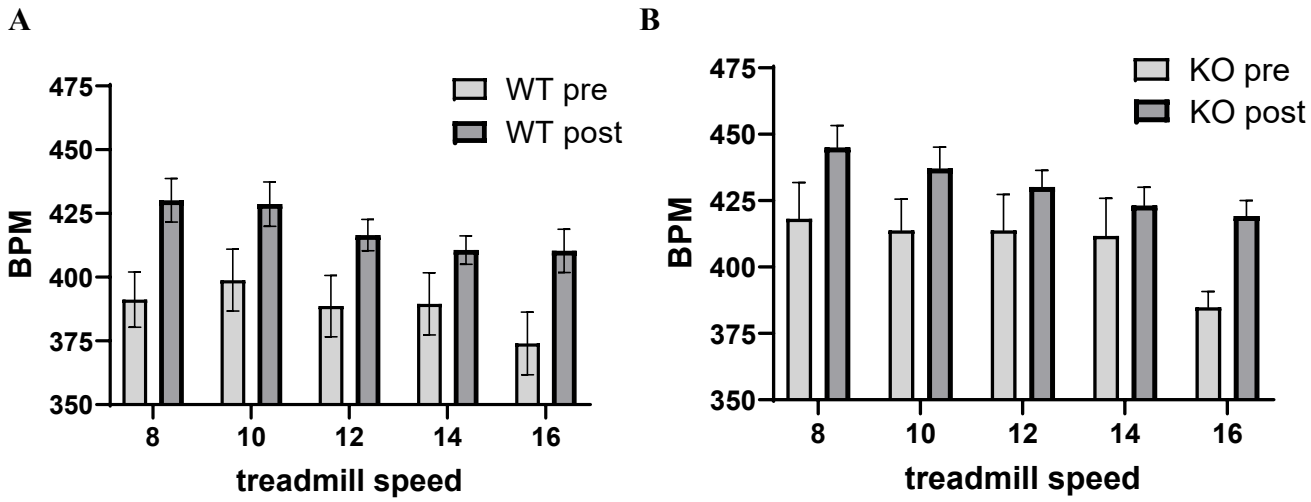
A



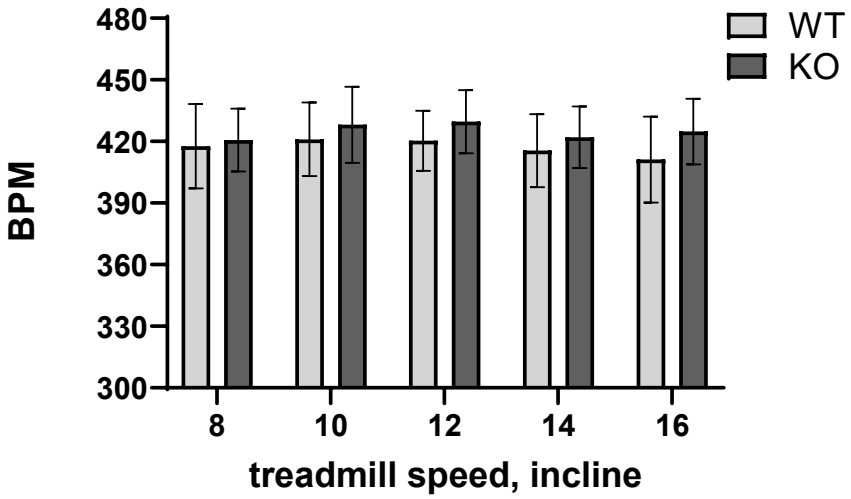
B



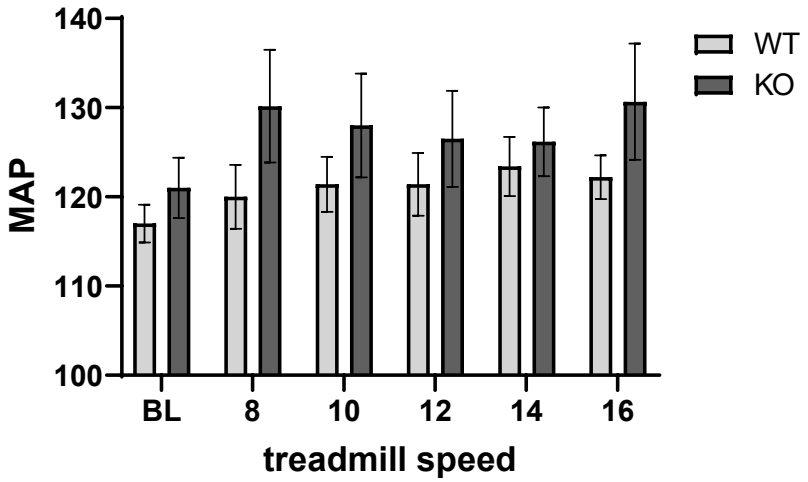
Supplementary Figure 5. Exercise compliance. A) Pink1 KO and Long-Evans WT rats exhibit compliance by maintaining movement on the treadmill with little or no contact with the backstop located at the rear of the treadmill and maintaining proximity toward the front of the treadmill. B) Compliance following completion of entire exercise regimen was observed >97% of the time in >75% of rats from both genotypes. There was no significant difference in compliance between the genotypes ($t = 1.49$, $p = 0.15$, $df = 18$).



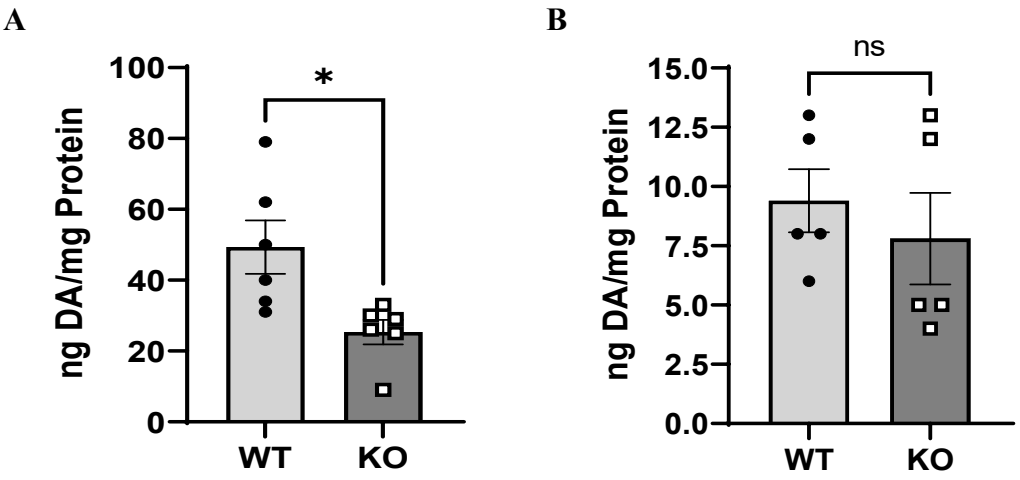
Supplementary Figure 6. Exercise impact on pre-exercise HR. A) WT. There was a highly significant increase in HR in the WT rats within the first 2 min of exercise cessation compared to pre-exercise levels. Post-exercise HR ($F_{(1,25)} = 19.47$, $p = 0.0002$). There was no effect of treadmill speed on the HR ($F_{(4,25)} = 1.92$, ns), nor interaction of treadmill speed interaction with post-exercise HR ($F_{(4,25)} = 0.21$, ns). B) KO. There was a highly significant increase in HR in the KO rats within the first 2 min of exercise cessation compared to pre-exercise levels. Post-exercise HR ($F_{(1,25)} = 25.25$, $p < 0.0001$). There was no effect of treadmill speed on the HR ($F_{(4,25)} = 1.67$, ns), nor interaction of treadmill speed interaction with post-exercise HR ($F_{(4,25)} = 0.80$, ns).



Supplementary Figure 7. Progressive treadmill exercise on 5-degree incline impact on HR. Increased exertion, as would be expected with progressively increased speed on an incline, did not further increase the HR following exercise. There was also no further decrease in the HR following exercise at the greater treadmill speeds as observed under the no incline conditions first imposed, suggesting maximal conditioning from the regimen during the sessions with no incline. Number of sessions/treadmill speed ($F_{(4,37)} = 0.75$, ns). Genotype ($F_{(1,10)} = 1.29$, ns); genotype x exercise session $F_{(4,37)} = 0.31$, ns).



Supplementary Figure 8. Influence of exercise anticipation on MAP. Placement on the treadmill affected MAP during the no incline phase of the treadmill regimen, without any genotype difference or interaction. Length of exercise session/treadmill speed ($F_{(5,46)} = 3.44$, $p = 0.01$), genotype ($F_{(1,10)} = 1.14$, *ns*); genotype x exercise session $F_{(5,46)} = 0.99$, ns).



Supplementary Figure 9. Dopamine-regulation in the mesoaccumbens pathway. Dopamine (DA) tissue content in the nucleus accumbens (NAc) or ventral tregmental area (VTA). A) NAc. DA levels were significantly less in the KO rat ($t = 2.90$, $*p = 0.016$, $df = 10$). B) VTA. In the same rats, DA levels were similar between WT and the KO rat ($t = 0.68$, ns, $df = 8$).