

KO tero- k zygote

	Primer used
Genotyping-nNOS	5'-TCA GAT CTG ATC CGA GGA GG-3'
Genotyping-nNOS-reverse	5'-TTC CAG AGC GCT GTC ATA GC-3'
Genotyping-neomycin	5'-CTT GGG TGG AGA GGC TAT TC-3'
Genotyping-neomycin-reverse	5'-AGG TGA GAT GAC AGG AGA TC-3'

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Supplementary Figure 1: Genotyping to demonstrate the absence of an intact nNOS gene in the genome of nNOS-KO mice and its presence in that of their WT littermates. Using primers for exon-2 of the nNOS gene, a cDNA fragment of the neomycin resistance gene (280 base pairs (Bp) long) was amplified, whereas the presence of the nNOS gene was demonstrated by the amplification of its exon-2 containing fragment (117 Bp long).



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Supplementary Figure 2: Shift of the nNOS isoform expression pattern in the rectus femoris muscle of PGC-1alpha transgenic (TG) mice in comparison to wild-type (WT) littermates (left side) and the Ponceau Red-stained blot matrices (right side). Note the higher expression of the alpha-isoform of nNOS (160 kDa) than the beta-isoform (140 kDa) in the PGC-1alpha TG mice than the WT mice. Shown are all immunoblots used for the densitometric analysis presented in Fig. 5C and 5D. Therefore, the densitometric values for nNOS-immunoreactive bands were normalized to the densitometric values of total protein loaded on the gels, as visualized by Ponceau Red-staining.



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Supplementary Figure 3: Immunoblot analysis for the demonstration of nNOS isoform expression in eight striated muscles from PGC-1alpha TG mice and their WT littermates. The immunoblots were used for the densitometric quantification of nNOS isoform expression shown in Fig. 5d and 5e. For both strains, detergent extracts of six mice (1-6) were subjected to the analysis. \* denote samples which were not included into the densitometry due to inaccurate gel loading and/or incorrect supply with ECL reagent during development of immunoblots.



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Supplementary Figure 4: Immunoblot analysis for the quantification of mitochondrial protein expression in six striated muscles from PGC-1alpha-TG mice and their WT littermates. For both strains, detergent extracts of six mice (1-6) were subjected to the analysis using the OXPHOS antibody cocktail accounting for ATP5A of complex (C) 5 (55 kDa), SDHB of C2 (30 kDa) and NDUFB8 of C1 (20 kDa) \* denote samples which contained higher (P<0.05) protein levels in the detergent extracts of the PGC-1alpha-TG mice than the WT mice. Please note that the immunoblots with the extracts of two additional striated muscles (RF and tongue) are presented in Fig. 6.