

LEGENDS TO SUPPLEMENTAL FIGURES

Supplemental Figure 1. Cultured cerebellar neurons express Glut4 mRNA. Total RNA was isolated from the adult mouse cerebellum (lane 1), 7-day-old mouse cerebellum (lane 2) and primary cerebellar neurons cultured in vitro for 6 days (lane 3), and the expression of Glut4 mRNA was analyzed by RT-PCR. The panel represents 1.5% agarose gel stained with EtBr.

Supplemental Figure 2. Sequence analysis of Glut4 mRNA. Total RNA was isolated from the adult mouse cerebellum, reverse transcribed by RT-PCR and the DNA fragments were separated in the agarose gel as shown in Fig. S1. DNA was extracted from the gel using the QIAquick Gel Extraction Kit (Qiagen, Valencia, CA) and sequenced at Boston University Medical Campus Molecular Genetics Core Facility (Boston, MA).

Supplemental Figure 3. Localization of Glut4 and synaptophysin in cerebellar neurons. Primary cultures of cerebellar neurons were serum-starved for 2 hrs. Cells were stained with the polyclonal antibody MC2A against Glut4 and a monoclonal antibody against synaptophysin followed by Alexa488-conjugated donkey anti-rabbit and Cy3-conjugated donkey anti-mouse antibodies. Bottom panels show control stainings with non-specific rabbit and mouse IgG.

Supplemental Figure 4. Glut4 content in the striatum. Samples of striatum (S) and cerebellum (C) of 25-day-old CD-1 mice were homogenized in a ball-bearing homogenizer, post-nuclear supernatant was prepared and 30 μ g aliquots were analyzed

by Western blotting with MC2A antibody. Dotted lines indicate that intervening lanes have been spliced out.

Supplemental Figure 5. Isolation of the plasma membrane fraction from the mouse cerebellum. Plasma membrane fraction was isolated from cerebelli of exercised and control mice as described in Materials and Methods and analyzed by Western blotting along with S2 in duplicate (20 μ g per lane). Cadherin and Glut3 represent plasma membrane markers, synaptophysin is a ubiquitous protein.