Supplementary Figure Legends

Supplementary Video 1. Digital 3-D reconstruction of a TRN segment labeled with anti-MEC-5 antibodies.

This 3-D digital model was assembled from serial thin sections comprising a 1 μm segment of PLM. The rendering includes the cuticle (top, in solid cyan), the TRN plasma membrane (bottom, transparent royal blue), anti-MEC-5 labels (gold spheres), microtubules (aqua rods), and a segment of a large mitochondrion (magenta).

Supplementary Video 2. Virtual fly-through of 3-D model of a TRN segment labeled with anti-MEC-2 antibodies.

This 3-D, digital model was derived from serial sections (50 nm) of a 7.5 μm segment of a PLM neurite in a wild-type animal. The digital reconstruction includes the plasma membrane, the internal face of the cuticle, all microtubules, and all anti-MEC-2 labels. For clarity, only three structures are rendered in this video: the plasma membrane (gray), terminating microtubules (aqua cylinders; red caps indicate distal endpoints), and membrane-associated anti-MEC-2 labels (indicated by red cones).

Supplementary Figure 1. Microtubule-associated MEC-2 and MEC-4 labels.

A, Immunoelectron micrographs of sections labeled with anti-MEC-2 (top) and anti-MEC-4 (bottom). Insets are 2X digital magnifications showing gold beads directly associated with 15pf microtubules. *B,* Position of microtubule-associated anti-MEC-2 (top) and anti-MEC-4 (bottom) labels. Solid indicates the PLM plasma membrane; the

dashed line separates the neurite cross-section into equal-area central and peripheral (rim) zones. *C,* Fraction of labels in the center *vs.* rim region. Numbers in each category indicated in parentheses. Scale bar, 200 nm. Distribution was not different from that expected if half of the labels were in the center and half were in the rim (Chi square test).

Supplementary Figure 2. Synapses and synaptic vesicles in the ventral nerve cord.
Vesicles and presynaptic densities were detected in the ventral nerve cord of the same animals used for SS-IEM. Inset shows a digital magnification of a presynaptic density.

Supplementary Figure 3. Specificity of mouse polyclonal antibodies.
Wild-type (N2) and null-mutant animals were stained with antibodies directed against MEC-2 (a), MEC-4 (b) and MEC-5 (c). In each panel, the top image shows labeling in a wild-type PLM, the middle image shows labeling in a null mutant animal expressing GFP under the control of TRN-specific *mec-17* promoter and the bottom image shows GFP in the same animal. Strains and mutant alleles are described in the main text.