Oxidative stress induces overgrowth of the *Drosophila* neuromuscular junction

Valerie J. Milton¹, Helen E. Jarrett², Kate Gowers¹, Laura Briggs¹, Iain M. Robinson², and Sean T. Sweeney¹.*

¹Department of Biology and Hull York Medical School, University of York, York YO10 5DD UK ²Peninsula College of Medicine and Dentistry, University of Plymouth

Synaptic terminals are known to expand and contract throughout an animal’s life. The physiological constraints and demands that regulate appropriate synaptic growth and connectivity are currently poorly understood. In previous work, we identified a *Drosophila* model of Lysosomal Storage Disease (LSD), *spinster* (*spin*), with larval neuromuscular synapse overgrowth. Here we identify a reactive oxygen species (ROS) burden in *spin* that may be attributable to previously identified lipofuscin deposition, a cellular hallmark of LSD. Reducing ROS in *spin* mutants rescues synaptic overgrowth and electrophysiological deficits. Synapse overgrowth was also observed in mutants defective for protection from ROS, and animals subjected to excessive ROS. ROS are known to stimulate ASK, JNK and Fos signalling. Furthermore, JNK and Fos in turn are known potent activators of synapse growth and function. Inhibiting JNK and Fos activity in *spin* also rescues synapse overgrowth and electrophysiological deficits. Similarly inhibiting ASK, JNK, Fos and Jun activity in animals subjected to oxidative stress rescues the overgrowth phenotype. These data suggest that ROS, via activation the JNK signalling pathway, are a major regulator of synapse overgrowth. Oxidative stress has been shown to transcriptionally activate autophagy, and transcriptional activation of autophagy genes have been shown to stimulate synapse growth. In support of this, here we report impaired autophagy gene function reverses overgrowth in *spin* and oxidative stress models. Our data describe a novel link between oxidative stress and synapse overgrowth via the JNK signalling pathway.