TITLE: Establishing a Synaptoneurosome Preparation to Investigate Memory Formation of a Meal

AUTHORS: Alexa Faulkner, Amy Ross

FACULTY SPONSER: Dr. Marise Parent, Neuroscience Institute

The hippocampus is critical for learning and memory. Memory plays an important role in feeding behavior. Our lab previously found that inhibiting hippocampal neurons, which are important for memory, accelerates meal onset. However, it has yet to be established that the hippocampus forms a memory of a meal. Long-term potentiation (LTP) is a strengthening of synaptic connections that is believed to be the molecular basis for hippocampal memory. We hypothesize that eating a meal will cause LTP in the hippocampus. To test this hypothesis, we need to isolate synaptoneurosomes, which are synapses that have been detached from a neuron. Successful isolation of a synaptic fraction will allow measuring biochemical markers of LTP in the hippocampus. Therefore, the aim of this experiment is to establish a synaptoneurosome preparation. We have compared three synaptoneurosome protocols for their efficacy in isolating synaptoneurosomes in rat hippocampi. PSD-95 and Acetyl H3 were measured as positive controls in homogenate tissue and isolated synaptoneurosomes. PSD-95 is a protein found in the post-synaptic membrane, so it should be more highly concentrated in the synaptoneurosome preparation than in the homogenate. Acetyl H3 is a nuclear marker, so it should only appear in homogenate tissue. Our results showed that Acetyl H3 was only visible in the homogenate, indicating that we successfully excluded nuclear material. However, PSD-95 was comparable between synaptoneurosome and homogenate. This suggests that we did not successfully recover all the synaptoneurosome material from the homogenate. We intend to attempt a new protocol utilizing fresh tissue instead of frozen, try different extraction reagents, and measure a post-synaptic protein other than PSD-95 as a positive control. Once synaptoneurosomes are successfully isolated, we intend to later implement the protocol into

experiments to test whether a meal causes LTP. One potential approach is to measure in GLuR1, a subunit of AMPA receptors that is upregulated after LTP. Determining the most successful synaptoneurosome preparation also has applications beyond learning models. A synaptoneurosome preparation could be very advantageous in experiments involving cellular metabolism, pharmacological treatments, and neurological disease models.

KEYWORDS: Hippocampus; Memory; Learning; Synaptoneurosome; Obesity; LTP; AMPA; AMPAR; Feeding; Eating; NMDA