P3.04.

Retrograde Endocannabinoid Signaling Reduces Firing and GABA-ergic Synaptic Transmission to Gonadotropin-Releasing Hormone Neurons

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Cannabinoids suppress fertility via reducing hypothalamic GnRH output. GABAAR-mediated transmission is a major input to GnRH cells that can be excitatory. We hypothesized that cannabinoids act via inhibiting GABAergic input by a retrograde endocannabinoid signaling mechanism. Loose-patch studies of slices from adult male GnRH-GFP mice showed that bath application of WIN55,212 decreased GnRH neuron firing rate. This action was detectable in presence of kynurenic acid, but disappeared when bicuculline was also present, indicating GABAA-R involvement. In immunocytochemical experiments, CB1-IR axons formed contacts with GnRH neurons and a subset established symmetric synapses characteristic of GABA-ergic neurotransmission. Whole-cell patch-clamp studies revealed that WIN55,212 decreased the frequency of GABAA-R-mediated mPSCs, which was prevented with the AM251, indicating that activation of presynaptic CB1 inhibits GABA release. AM251 alone increased mPSC frequency, providing evidence that endocannabinoids tonically inhibit GABAA-R drive onto GnRH neurons. Increased mPSC frequency was absent when DAG-lipase was blocked intracellularly with THL, showing that tonic inhibition is caused by 2-AG production of GnRH neurons. When CdCl2 was applied to inhibit action potential-evoked calcium influx and endocannabinoid-mediated blockade of spontaneous vesicle fusion was blocked with AM251, GnRH neuron firing increased, revealing an endogenous endocannabinoid brake on GnRH neuron firing.