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## Supplemental information

## Synaptotagmin-1 is a Ca<sup>2+</sup> sensor

## for somatodendritic dopamine release

Joseph J. Lebowitz, Aditi Banerjee, Claire Qiao, James R. Bunzow, John T. Williams, and Pascal S. Kaeser



**Figure S1. Basal electrical properties are unchanged in Syt-1 cKO**<sup>DA</sup> **mice, related to Fig. 1. A, B.** Example traces (A) and quantification of spontaneous firing rate (B) recorded in cellattached mode prior to break-in, measured over a one-minute period, Syt-1 control 5 cells/3 mice, Syt-1 cKO<sup>DA</sup> 9 cells/3 mice.

**C**, **D**. Quantification of series resistance (C) and capacitance (D) measured following break-in, Syt-1 control 35 cells/8 mice, Syt-1 cKO<sup>DA</sup> 36 cells/8 mice.

Data are mean ± SEM; no significant differences were observed as determined by unpaired Student's t-tests (B, D) or a Mann Whitney rank sum test (C).



Figure S2. Assessment of D2-IPSC rise times in Syt-1 cKO<sup>DA</sup> mice, related to Fig. 1.

**A-C.** Absolute (A) and scaled (B) example traces and quantification of 10-90% rise time of singlestimulus D2-IPSCs of the recordings shown in Figs. 1D and 1G, Syt-1 control 25 cells/7 mice, Syt-1 cKO<sup>DA</sup> 8 cells/5 mice. Only D2-IPSCs larger than 10 pA were included. Because of the strong reduction in the D2-IPSC amplitude after Syt-1 ablation, there are fewer observations for Syt-1 cKO<sup>DA</sup>. The kinetics of the D2-IPSC are dominated by the time course of GPCR signaling and are unlikely to reflect release kinetics.

Data are mean ± SEM; no significant differences were observed as determined by unpaired Student's t-test (C).



Figure S3. D2 receptor overexpression does not alter evoked D2-IPSCs, related to Fig. 4.A. Strategy for AAV-mediated overexpression of D2 receptors (human, short version, hD2Rs) in

the midbrain.

**B**, **C**. Example traces (B) and quantification (C) of D2-IPSCs evoked by single stimuli after overexpression of D2 receptors, Syt-1 control 10 cells/3 mice, Syt-1 cKO<sup>DA</sup> 10 cells/4 mice.

**D**, **E**. Example traces (D) and quantification of paired pulse ratios (E) of D2-IPSCs evoked by two stimuli (1-s interval) after D2 receptor overexpression, Syt-1 control 14 cells/7 mice, Syt-1 cKO<sup>DA</sup> 9 cells/5 mice.

Data are mean ± SEM; \*\*\*p < 0.005, statistical significance determined by a Mann Whitney rank sum test (C) or an unpaired Student's t-test (E). Overall, D2-IPSCs are similar compared to experiments without D2 receptor overexpression (Fig. 1) in both genotypes.