Supporting Information

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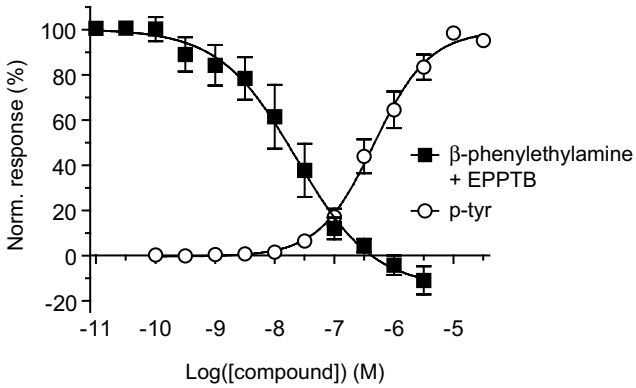

Fig. S1. Inhibition of the TAAR1-mediated increase in cAMP levels by EPPTB in transfected HEK293 cells. Open circles, cAMP accumulation in response to increasing concentrations of *p*-tyr (EC_{50} = 545 ± 179 nM). Data were normalized to the cAMP level obtained with 10 μM β-phenylethylamine. Filled squares, inhibition of β-phenylethylamine (1.5 μM)-mediated cAMP accumulation by increasing concentrations of EPPTB (IC_{50} = 27.5 ± 9.4 nM). Bars show mean values ± SEM of four experiments. Data analysis and curve fitting was performed using nonlinear regression (GraphPad Prism Version 5.01).

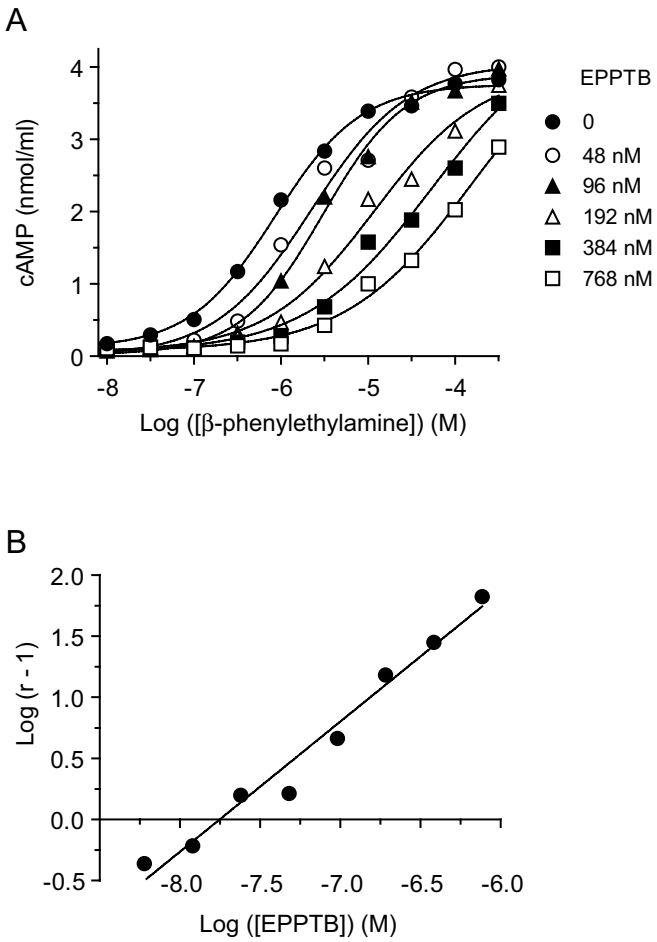

Fig. S2. Schild plot analysis of EPPTB in transfected HEK293. (A) Concentration-response curves for the β-phenylethylamine-induced accumulation of cAMP under control conditions and in the presence of increasing concentrations of EPPTB (48–768 nM, 2-fold increases). A representative experiment is shown. (B) Schild plot derived from the concentration-response curves of two independent experiments. The concentration ratio r is the EC_{50} for β-phenylethylamine in the presence of EPPTB divided by the EC_{50} for β-phenylethylamine under control conditions. The slope of the line fitted to the concentration ratios is 1.065, which suggests that EPPTB is a competitive antagonist.

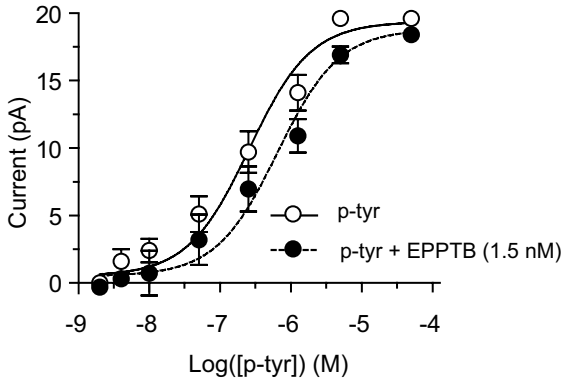

Fig. S3. The dose-response curve of the *p*-tyr-induced K^+ current amplitude at DA neurons is shifted to the right in the presence of 1.5 nM EPPTB, with no significant reduction in the maximally *p*-tyr-induced current amplitude (EC_{50} values: *p*-tyr, 305 ± 11 nM; *p*-tyr + EPPTB, 650 ± 11 nM). This is consistent with a competitive mode of action of EPPTB at the *p*-tyr binding-site.

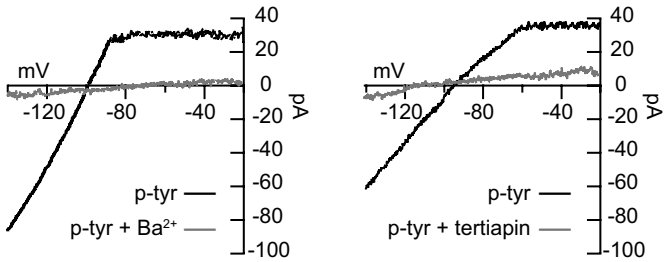

Fig. S4. The *p*-tyr induced K^+ current in DA neurons of the VTA is sensitive to Ba^{2+} and tertiapin. The *p*-tyr induced currents were obtained by calculating the difference between the I–V curves before and after addition of *p*-tyr (10 μM), both in the absence (black traces) and presence (gray traces) of the nonselective K^+ channel blocker Ba^{2+} (300 μM) and the Kir3 channel blocker tertiapin (10 μM), respectively. This suggests that the activation of Kir3 channels underlies the *p*-tyr induced K^+ current in DA neurons of the VTA.

Table S1. Percent inhibition of control specific binding (mean ± SEM) is shown for EPPTB (10 μM) on several receptors and transporters

Receptors/ Transporters	Assay	% Inhibition of Control Specific Binding	SEM % Control
Dopamine receptors	D1	25	6.3
	D2S	9	8.9
5-HT receptors	5-HT1A	4	1.6
	5-HT1B	53	4.2
	5-HT2A	21	3.3
	5-HT3	9	1.3
	5-HT5A	21	1.1
	5-HT6	7	0.9
	5-HT7	10	2.9
Adrenergic receptors	alpha 1 (non-selective)	8	3.6
	alpha 2 (non-selective)	23	4.0
	beta 1	4	1.5
	beta 2	6	3.1
Muscarinic receptors	M1	4	7.8
	M2	−1	6.5
	M3	−10	1.2
Neurotransmitter transporters	Norepinephrine transporter	44	2.5
	Dopamine transporter	43	1.2
	5-HT transporter	−7	4.6

The results were obtained from three independent experiments.