Supporting Information

Krabbe et al. 10.1073/pnas.1500450112

SI Materials and Methods

Animals. Mice expressing the human D2R under control of the tetO promoter were generated on a C57BL/6-CBA(J) background and backcrossed for >10 generations to the C57BL/6(J) background. Crossing these animals with mice expressing the tetracycline transactivator (tTA) transgene under the calcium/calmodulin-dependent kinase IIα (CaMKIIα) promoter (1) ($129SveV_{Tac}$) background, backcrossed for >20 generations, resulted in double-transgenic mice expressing human D2Rs selectively in striatal medium spiny neurons. Littermates carrying a single transgene (tetO-hD2R or CaMKIIα-tTA) or no transgene were used as controls (2). Animals were housed in groups on a 12-h light/12-h dark cycle. Mice that received tracer injections were individually housed after surgery. Food and water were available ad libitum.

In Vivo Extracellular Single-Unit Recordings and Juxtacellular Labeling. Mice were anesthetized with isoflurane (Forene, Abbott GmbH) in O_2 at a rate of 0.35 L/min^{-1} [2.5% (vol/vol) for induction, 1.0–1.8% for maintenance] after i.p. injection of atropine (0.1 mg/kg of body weight) and s.c. injection of glucose in H_2O (1 g/kg of body weight). Animals were placed on a heating pad in a stereotactic frame (David Kopf Instruments). Rectal temperature (34–36 °C), heart rate (5–8 Hz), respiration (1–2 Hz), and electrocorticograms were constantly monitored as measure of anesthetic level. Stereotactic coordinates were derived from and provided relative to bregma. Epidural electrocorticograms were recorded using two 1-mm stainless-steel screws above the left cortex (rostral: +2.1 mm, lateral: 1.2 mm) and ipsilateral cerebellum (caudal: −5.5 to −6.5 mm, lateral: 1.2 mm). Bilateral craniotomies for single-unit recordings were performed at the following coordinates: VTA: caudal: −3.60 mm, lateral: 0.2–0.8 mm, recording depth: 3.0–4.2 mm; SN: caudal: −3.08 mm, lateral: 0.8–1.4 mm, recording depth: 3.2–4.4 mm. Rostrocaudal coordinates were adjusted to the skull size (VTA: −3.6 mm/4.2 * distance [bregma-lambda] + 0.2 mm, SN: −3.08 mm/4.2 * distance [bregma-lambda] + 0.33 mm). Lateral coordinates were aligned to the sagittal sinus to avoid rupture of the blood vessel.

Single-unit recordings were carried out using glass microelectrodes (15–25 MΩ, borosilicate capillaries; Harvard Apparatus) filled with 0.5 M NaCl, 10 mM Hepes, and 1.5% (wt/vol) neurobiotin (Vector Laboratories), which were lowered to the target area with a micromanipulator (SM-6; Luigs & Neumann). Extracellular signals were amplified 1,000-fold (ELC-03M; npi Electronics), notch- and bandpass-filtered (50 Hz–5 kHz, DFA-2FS; npi Electronics), and digitized with an EPC-10 A/D converter (Heka Electronics). Sampling rates were set to 12.5 kHz for spike trains and to 20 kHz for action potential waveform recording. Additionally, neuronal activity was displayed on an analog oscilloscope (Combiscope HM1008-2; HAMEG Instruments) and an audio monitor (AUDIS-01; npi Electronics). Putative DA neurons were identified by broad triphasic action potentials (≥1.1 ms) (3) and a spontaneous activity of 0.1–10 Hz. Discharges occurred as single-spike or burst activity with regular or irregular firing patterns. Following recording (≥10 min), neurons were labeled with neurobiotin using the juxtacellular technique (4). Positive current pulses (1–12 nA, 200 ms ON/OFF pulses) were applied through the microelectrode. Neuronal activity was constantly monitored, and the current amplitude was adjusted to obtain an increase of action potential firing selectively during ON pulses. Single-cell labeling was usually successful if entrainment of neuronal activity lasted for ≥25 s and spontaneous discharges continued after current application.

Data were analyzed using Igor Pro-6.02 (WaveMetrics) with a NeuroMatic plug-in (www.neuromatic.thinkrandom.com), R statistical computing (www.r-project.org), and MATLAB 7 (MathWorks). Spontaneous firing rates and the CV were analyzed from ≥10-min spike train recordings. Interspike interval histograms were generated using 10-ms bins. To analyze bursting activity in detail, the classical 80/160-ms burst criterion (5) was applied. In brief, the beginning of a burst was defined when two spikes occurred within ≤80 ms, and the end was defined as soon as an interspike interval exceeded 160 ms. Bursts were further analyzed with respect to the mean number of spikes per burst, mean intraburst frequency, and maximum firing frequency. Action potential waveform parameters were derived from at least 20 individual spikes. The action potential width was measured from the beginning of the first peak to the trough.

Autocorrelation histograms (ACHs) were plotted in R (1-ms bins, smoothed with a Gaussian filter) to determine the dominant activity pattern by visual inspection: single-spike oscillatory, single-spike irregular, bursty oscillatory, or bursty irregular (6–8). Single-spike oscillatory-classified ACHs featured three or more equally distant peaks with decreasing amplitudes. Bursting neurons displayed a narrow initial peak, which was followed by a return to steady state (bursty irregular) or equidistant broader peaks with decreasing amplitudes (bursty oscillatory). All other cells were classified as single-spike irregular.

We also fitted a GLO stochastic model (7, 8) to the obtained spike trains to investigate irregularity and burstiness in more detail. The GLO model was developed for the description of characteristic in vivo firing patterns in dopaminergic spike trains and contains two stochastic steps. In the first step, a background oscillator with normally distributed intervals with oscillation period μ and variance σ_1^2 generates a background oscillation whose irregularity depends on σ_1/μ. In the second step, each background event is assumed to give rise to a Poissonian number of spikes with expectation λ ($\lambda > 1$ in the bursty case, $m = 1$) or to a single spike with probability λ ($\lambda < 1$ in the single-spike case, $m = 0$). The spikes are then placed around the backbone beat according to a normal distribution with an SD (or burst width) of σ_2. Therefore, the GLO parameters can be used to measure irregularity and burstiness: The irregularity parameter $\theta = \sqrt{(\sigma_1^2 + 2\,\sigma_2^2)}/\mu$ combines the irregularity of the background oscillation with the width of the bursts, σ_2, because bursts of high width may overlap, and thus increase irregularity. In addition, a value of λ much smaller than 1 for the single-spike case indicates a small probability of firing per background beat, and thus higher irregularity. The parameters can be estimated by fitting the empirical autocorrelation function to the empirical ACH of the obtained spike trains. The classification of firing patterns into the four classes of oscillatory bursty, oscillatory single spike, irregular bursty, and irregular single spike relies on the parameter m, which primarily uses the existence or absence of a central peak in the ACH to distinguish between bursty ($m = 1$) and single spike ($m = 0$). Irregularity is determined by a threshold on the parameter θ (compare ref. 8). We chose a threshold of $\theta = 0.35$ for cells firing in a single-spike fashion, whereas the threshold was set to $\theta = 0.4$ for cells firing bursts (7), because of the generally lower oscillatory nature of bursty spike trains. An additional parameter $\beta = \mu/\sigma_2$ (i.e., the reciprocal of the relative burst width) was used to classify extremely irregular spike trains.

Independent of the other parameters, spike trains with $\beta < 3$ were defined as irregular.

When fitting the GLO to the obtained spike trains, 12 spike trains needed to be excluded due to improper fits (five in D2R-OE, five in Dox-treated control, and two in Dox-treated D2R-OE), and the reported analyses refer to the remaining 119 spike trains. The classification of firing patterns displayed in the main text always relates to the GLO fitting; these results can be compared with the visual evaluation given in Tables S2, S3, and S5.

Immunohistochemistry and Confocal Analysis. After electrophysiological in vivo recordings, mice were deeply anesthetized with i.p. pentobarbital (0.7 g/kg of body weight, Narcoren; Merial GmbH) and transcardially perfused with an ice-cold solution containing 4% (wt/vol) paraformaldehyde and 15% (vol/vol) picric acid in phosphate buffer. After postfixation overnight at 4 °C, brains were sectioned in coronal slices (60 μm) using a vibratome (VT1000S; Leica). Free-floating sections were rinsed with PBS four times before incubation in blocking solution [10% (vol/vol) horse serum, 0.2% BSA, and 0.5% Triton X-100 in PBS] for 2 h at room temperature (20–22 °C). Subsequently, the blocking solution was replaced by the primary antibody diluted in carrier solution (1% horse serum, 0.2% BSA, and 0.5% Triton X-100 in PBS). Monoclonal mouse anti-TH antibody (1:750, catalog no. MAB318; Millipore) or polyclonal rabbit anti-TH antibody (1:1,000, catalog no. 657012; Millipore) was used for staining of DA midbrain neurons. After overnight incubation at room temperature, sections were washed with PBS four times before adding the secondary antibody and Alexa Fluor 488 streptavidin (1:1,000, catalog no. S11223; Molecular Probes) diluted in carrier solution. The following secondary antibodies were used at a ratio of 1:750: Alexa Fluor 568 (catalog no. A-11036) or Alexa Fluor 647 (catalog no. A-21245) and goat anti-rabbit IgG or Alexa Fluor 647 goat anti-mouse IgG (catalog no. A-21236; all from Molecular Probes). Incubation overnight at room temperature was followed by final washing with PBS (four times). Sections were mounted on slides and coverslipped. For confocal analysis, a laser-scanning confocal microscope (LSM 510 Meta; Zeiss) controlled with LSM software (version 3.2; Zeiss) was used. Overview pictures for localization of neurons within the midbrain subnuclei were acquired with a 10×/0.3 lens. A 63×/1.4 oil immersion lens was used for identification of the neurochemical phenotype. Identical confocal settings were chosen for all scanning sessions.

Retrograde Tracing. Stereotactic surgical procedures were performed as described for electrophysiological in vivo recordings. For retrograde labeling of mesolimbic DA VTA and nigrostriatal DA SN neurons, rhodamine-coupled latex microspheres (Retrobeads; Lumafluor) were used. Retrobeads were diluted 1:2 in sterile artificial cerebrospinal fluid and injected bilaterally into the NAc shell and core (45–60 nL; coordinates from bregma: rostral: +1.6 mm, lateral: 0.8 mm, ventral: 4.0 mm), or the dorsal striatum [caudate putamen (CPu); 2 × 60 nL; coordinates from bregma: rostral: +0.98 mm, lateral: 1.90 mm, ventral: 3.20 mm; rostral: −0.10 mm, lateral: 2.70 mm, ventral: 3.20 mm; all coordinates according to Paxinos and Franklin (9)] with a NanoFil syringe attached to a micropump (rate of 30 $nL \cdot min^{-1}$ for DA VTA tracing, 50 $nL \cdot min^{-1}$ for DA SN tracing; UMP3, World Precision Instruments). Animals were killed 14 d (NAc) or 7 d (CPu) after retrobead injection for UV-LMD and RT-qPCR.

For verification of tracer deposition, injection sites were fixated in 4% (wt/vol) paraformaldehyde in phosphate buffer overnight at 4 °C and cut in coronal slices (60 μm) using a vibratome. Fluorescent immunostaining for TH of free-floating sections was performed essentially as described above (primary antibody: rabbit anti-TH, 1:1,000, catalog no. 657012, Millipore; secondary antibody: Alexa Fluor 488 goat anti-rabbit IgG, 1:1,000, catalog no. A-11008, Molecular Probes).

qPCR Assay of Retrogradely Labeled Neurons. Details of all qPCR assays (TaqMan) and their respective standard curve parameters (derived from mouse midbrain tissue) used for RT-qPCR analysis in this study are provided in Table S5. The cDNA amount per cell in relation to the used standard was calculated according to Gründemann et al. (10):

$$cDNA\ amount\ per\ cell = \frac{S^{[(Ct - Y_{intercept})/slope]}}{No_{cells} \bullet cDNA\ fraction},$$

with S being the serial dilution factor of the standard curve (i.e., ref. 10), No_{cells} being the number of captured neurons per UV-LMD pool (i.e., 10), *cDNA fraction* being the fraction of the UV-LMD cDNA reaction sample that was used in an individual PCR assay (i.e., 5/17), and a $Y_{intercept}$ of 42 for all assays.

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Table S1. Summary of results of in vivo single-unit recordings and complete statistical results

	VTA			SN			
Variable	Control	D2R-OE	*P* value	control	D2R-OE	*P* value	Test
Firing parameters							
Mean frequency, Hz	3.7 ± 0.4	2.4 ± 0.3	0.010*	4.6 ± 0.5	3.9 ± 0.4	0.291	*t*
Action potential duration, ms	1.43 ± 0.05	1.34 ± 0.04	0.112	1.48 ± 0.09	1.40 ± 0.07	0.498	*t*
Burst rate, min^{-1}	4.4 (0.8/16.4)	0.4 (0/4.1)	0.005**	2.5 (0.8/14.2)	3.6 (0.7/13.8)	0.868	MWU
% SFB	5.3 (1.3/29.6)	0.5 (0/6.5)	0.012*	2.7 (0.8/22.3)	5.2 (0.8/21.8)	0.819	MWU
CV, %	58.1 (37.5/99.9)	53.0 (32.3/83.9)	0.551	46.0 (25.6/53.3)	59.6 (34.3/83.9)	0.077	MWU
	n = 28	*n* = 22		*n* = 16	*n* = 14		
Burst characteristics							
Spikes per burst	2.6 (2.3/3.1)	2.4 (2.2/2.8)	0.478	2.9 (2.1/3.5)	2.6 (2.1/3.3)	0.661	MWU
Short bursts (2 spikes), %	59.3 ± 5.6	64.3 ± 7.9	0.603	57.3 ± 7.2	62.2 ± 7.3	0.645	*t*
Long bursts (≥5 spikes), %	4.0 (0/9.3)	0 (0/1.8)	0.078	1.8 (0/21.9)	2.2 (0/15.6)	0.773	MWU
Mean intraburst frequency, Hz	15.2 (13.4/17.0)	16.8 (13.3/20.7)	0.389	13.6 (11.6/15.9)	13.5 (12.9/15.1)	0.819	MWU
Maximum frequency, Hz	38.3 (21.8/57.3)	30.1 (22.1/97.0)	0.902	27.9 (21.0/35.5)	30.5 (18.7/41.6)	0.633	MWU
	n = 25	*n* = 13		*n* = 16	*n* = 14		
Pattern analysis							
Visual classification, % of cells							
Single-spike oscillatory	28.57	31.82	0.403	62.50	21.43	0.091	χ^2
Single-spike irregular	42.86	59.09		18.75	57.14		
Bursty oscillatory	10.71	4.55		12.50	7.14		
Bursty irregular	17.86	4.55		6.25	14.29		
	n = 28	*n* = 22		*n* = 16	*n* = 14		
GLO classification, % of cells							
Single-spike oscillatory	35.71	42.11	0.687	62.50	58.33	0.212	χ^2
Single-spike irregular	28.57	36.84		6.25	16.67		
Bursty oscillatory	17.86	15.79		31.25	8.33		
Bursty irregular	17.86	5.26		0	16.67		
	n = 28	*n* = 19		*n* = 16	*n* = 12		

This table is related to Fig. 1. To compare genotypes, data that were normally distributed were statistically tested using a Student's *t* test (*t*); in such case, we present the mean ± SEM for each genotype. In the case of nonnormally distributed data (criteria are discussed in *Materials and Methods*), the MWU test was applied, and we provide median values (25–75% quantiles). The distribution of in vivo firing patterns was analyzed using Pearsons's χ^2 test. For all datasets, *n* depicts the number of cells used in each test and *N* is the number of animals from which the neurons were derived (DA VTA neurons: control: $n = 14$; D2R-OE: $n = 8$; DA SN neurons: control: $n = 8$, D2R-OE: $n = 7$). *$P < 0.05$; **$P < 0.01$.

Table S2. Details of GLO analysis: Results of GLO analysis for DA VTA and DA SN neurons from control and D2R-OE mice under control conditions

	DA VTA neurons						DA SN neurons					
	Single spike			Bursty			Single spike			Bursty		
Parameter	Control	D2R-OE	*P* value	Control	D2R-OE	*P* value	Control	D2R-OE	*P* value	Control	D2R-OE	*P* value
θ	0.32 (0.25–0.93)	0.35 (0.24–0.56)	0.93	0.41 (0.30–0.46)	0.28 (0.24–0.34)	0.3	0.25 (0.21–0.27)	0.31 (0.28–0.34)	0.02*	0.31 (0.28–0.35)	0.46 (0.34–0.58)	—
σ_1/μ	0.24 (0.14–0.30)	0.25 (0.19–0.40)	0.24	0.17 (0.16–0.31)	0.21 (0.17–0.29)	0.84	0.21 (0.17–0.24)	0.31 (0.27–0.31)	0.03*	0.22 (0.21–0.30)	0.28 (0.23–0.49)	—
σ_2/μ	0.13 (0.02–0.52)	0.07 (0.003–0.30)	0.82	0.25 (0.12–0.27)	0.10 (0.06–0.13)	0.19	0.07 (0.06–0.12)	0.09 (0.05–0.15)	0.77	0.13 (0.13–0.15)	0.09 (0.08–0.17)	—
λ	0.83 (0.67–0.95)	0.90 (0.82–0.95)	0.38	3.4 (1.6–4.8)	3.1 (2.9–3.7)	0.95	0.91 (0.86–0.98)	0.87 (0.85–0.91)	0.94	1.6 (1.1–2.3)	3.2 (2.3–3.3)	—
	$n = 18$	$n = 15$		$n = 10$	$n = 4$		$n = 11$	$n = 9$		$n = 5$	$n = 3$	

Parameters are as follows: θ (irregularity parameter) = $\sqrt{(\sigma_1^2 + 2\,\sigma_2^2)}/\mu$, corresponds to inverse height of first ACH-side peak relative to baseline level and incorporates the following two irregularity parameters: σ_1/μ (irregularity of background oscillation) = SD of background increments, relative to the oscillation period; σ_2/μ (relative burst width/coupling to background oscillation) = SD of spikes placed around a background beat, relative to the oscillation period. λ = expected number of spikes per background beat: (*i*) in the bursty case: expected number of spikes per burst, with number of spikes following a Poisson distribution, and (*ii*) in the single-spike case: probability to observe a spike at a background beat. This table is related to Figs. 2 and 4. Because most distributions were asymmetrical, we applied the MWU test to compare genotypes and provide the median (25–75% quantiles) for each genotype for each parameter. Because the investigated parameters tend to differ between bursty and single-spike cells, we performed separate statistical comparisons for these groups, applying MWU tests. Statistical tests were only performed if the sample size of both samples summed to at least 10. Otherwise, we omitted statistical significance tests. In the interpretation of statistical *P* values, we first focused on the irregularity parameter θ, applying a Bonferroni correction for the number of performed tests. As a result, only $P = 0.0003$ was considered significant. The remaining parameters were subjected to exploratory analyses. Statistical significance levels are as follows: $^*P < 0.05$.

Table S3. Details of GLO analysis: Results of GLO analysis for DA VTA and DA SN neurons from control and D2R-OE mice after switching off transgenic overexpression with Dox treatment

	DA VTA neurons + Dox						DA SN neurons + Dox					
	Single spike			Bursty			Single spike			Bursty		
Parameter	Control	D2R-OE	*P* value	Control	D2R-OE	*P* value	Control	D2R-OE	*P* value	Control	D2R-OE	*P* value
θ	0.44 (0.40–0.68)	0.21 (0.17–0.31)	0.0003***	0.42 (0.37–0.49)	0.4	—	0.30 (0.26–0.33)	0.25 (0.22–0.27)	—	0.62 (0.52–0.90)	0.39 (0.32–0.40)	0.01*
σ_1/μ	0.26 (0.12–0.33)	0.18 (0.12–0.22)	0.51	0.28 (0.37–0.49)	0.28	—	0.27 (0.23–0.29)	0.16 (0.14–0.18)	—	0.43 (0.39–0.48)	0.31 (0.28–0.34)	0.07
σ_2/μ	0.23 (0.16–0.48)	0.07 (0.02–0.11)	0.01*	0.22 (0.17–0.27)	0.19	—	0.06 (0.001–0.12)	0.13 (0.08–0.17)	—	0.32 (0.19–0.55)	0.15 (0.09–0.16)	0.17
λ	0.69 (0.59–0.85)	0.92 (0.90–0.98)	0.034*	2.3 (2.2–2.9)	3.0	—	0.85 (0.80–0.89)	0.94 (0.88–0.99)	—	2.3 (1.8–3.1)	2.9 (2.5–4.1)	0.61
	$n = 9$	$n = 12$		$n = 3$	$n = 2$		$n = 4$	$n = 4$		$n = 4$	$n = 6$	

Parameters are as follows: θ (irregularity parameter) = $\sqrt{(\sigma_1^2 + 2\,\sigma_2^2)}/\mu$, corresponds to inverse height of first ACH-side peak relative to baseline level and incorporates the following two irregularity parameters: σ_1/μ (irregularity of background oscillation) = SD of background increments, relative to the oscillation period; σ_2/μ (relative burst width/coupling to background oscillation) = SD of spikes placed around a background beat, relative to the oscillation period. λ = expected number of spikes per background beat: (*i*) in the bursty case: expected number of spikes per burst, with number of spikes following a Poisson distribution, and (*ii*) in the single-spike case: probability to observe a spike at a background beat. This table is related to Figs. 2 and 4. Because most distributions were asymmetrical, we applied the MWU test to compare genotypes and provide the median (25–75% quantiles) for each genotype for each parameter. Because the investigated parameters tend to differ between bursty and single-spike cells, we performed separate statistical comparisons for these groups, applying MWU tests. Statistical tests were only performed if the sample size of both samples summed to at least 10. Otherwise, we omitted statistical significance tests. In the interpretation of statistical *P* values, we first focused on the irregularity parameter θ, applying a Bonferroni correction for the number of performed tests. As a result, only $P = 0.0003$ was considered significant. The remaining parameters were subjected to exploratory analyses. Statistical significance levels are as follows: $*P < 0.05$; $***P < 0.001$.

Table S4. Summary of results of in vivo single-unit recordings and complete statistical results after Dox treatment

	VTA + Dox			SN + Dox			
Variable	Control	D2R-OE	*P* value	Control	D2R-OE	*P* value	Test
Firing parameters							
Mean frequency, Hz	4.0 ± 0.6	3.7 ± 0.5	0.713	4.8 ± 0.6	4.4 ± 0.5	0.569	*t*
Action potential duration, ms	1.26 ± 0.05	1.24 ± 0.05	0.738	1.47 ± 0.08	1.37 ± 0.04	0.272	*t*
Burst rate, min^{-1}	4.9 (0.5/53.3)	0.4 (0.03/4.4)	0.039*	9.7 (2.9/26.1)	16.8 (0.6/24)	0.925	MWU
% SFB	5.2 (0.7/53.2)	0.6 (0.01/4.3)	0.031*	9.4 (3.6/41.9)	17.3 (0.5/40)	0.971	MWU
CV, %	67.6 (46.5/75.8)	38.8 (31.2/69.9)	0.041*	76.2 (33.2/96.9)	62.4 (43.5/103.2)	0.563	MWU
	n = 15	*n* = 16		*n* = 10	*n* = 10		
Burst characteristics							
Spikes per burst	2.4 (2.2/0.3)	2.6 (2.4/3.0)	0.738	3.3 (2.6/4.1)	2.9 (2.4/3.9)	0.597	MWU
Short bursts (2 spikes), %	61.3 ± 7.7	58.0 ± 8.0	0.774	48.2 ± 7.0	43.5 ± 8.9	0.685	*t*
Long bursts (≥5 spikes), %	2.6 (0/15.2)	1.6 (0/9.6)	0.766	17.5 (2.2/26.2)	2.3 (0/26.7)	0.517	MWU
Mean intraburst frequency, Hz	14.3 (13.8/16.7)	13.2 (12.6/16.0)	0.105	13.7 (11.5/14.6)	12.6 (12.2/15.0)	0.796	MWU
Maximum frequency, Hz	45.6 (22.9/68.6)	32.7 (21.5/40.2)	0.190	34.8 (20.6/51.9)	39.8 (26.6/60.7)	0.796	MWU
	n = 14	*n* = 12		*n* = 10	*n* = 10		
Pattern analysis							
Visual classification, % of cells							
Single-spike oscillatory	13.33	50	0.046*	30	40	0.088	χ^2
Single-spike irregular	53.33	37.50		30	10		
Bursty oscillatory	0	6.25		0	40		
Bursty irregular	33.33	6.25		40	10		
	n = 15	*n* = 16		*n* = 10	*n* = 10		
GLO classification, % of cells							
Single-spike oscillatory	16.67	71.43	0.024*,†	50	40	0.044*,‡	χ^2
Single-spike irregular	58.33	14.29		0	0		
Bursty oscillatory	8.33	7.14		0	50		
Bursty irregular	16.67	7.14		50	10		
	n = 12	*n* = 14		*n* = 10	*n* = 10		

This table is related to Fig. 4. To compare genotypes, data that were normally distributed were statistically tested using a Student's *t* test (*t*); in such case, we present the mean ± SEM for each genotype. In the case of nonnormally distributed data (criteria are discussed in *Materials and Methods*), the MWU test was applied and we provide median values (25–75% quantiles). The distribution of in vivo firing patterns was analyzed using Pearsons's χ^2 test. In the case of a significant χ^2 test result, we performed Fisher's exact test and report only the statistically significant results of those tests (symbols listed below). For all datasets, *n* depicts the number of cells used in each test and *N* is the number of animals from which the neurons were derived (DA VTA neurons: control: *n* = 6; D2R-OE: *n* = 6; DA SN neurons: control: *n* = 5, D2R-OE: *n* = 4).

*$P < 0.05$.

†Fisher's exact test: *P* (single-spike oscillatory) = 0.008, *P* (single-spike irregular) = 0.038.

‡Fisher's exact test: *P* (bursty oscillatory) = 0.036.

Table S5. Details of qPCR assays, amplicons, and assay-specific standard curve parameters

Assay ID	Target gene reporter-context sequence-quencher (primer sequences)	GenBank accession no. (NCBI)	Amplicon length, bp	Exon spanning	Threshold	Standard curve data			
						Y-intercept	Slope	R^2	n
Mm01336438_m1	Mouse mNR1 FAM–TCTACTCTGACAAGAGCATCCACCT–NFQ	NM_001177656.1	63	2–3	0.7	42.01 ± 0.09	−3.42 ± 0.07	0.99 ± 0.00	5
Mm00433801_m1	Mouse mNR2A FAM–ACTCTGCACCAATTTATGGTCAATG–NFQ	NM_008170.2	58	2–3	0.55	44.29 ± 0.19	−3.33 ± 0.04	0.99 ± 0.00	6
Mm01265041_m1	Mouse mNR2B FAM–CTTCTACAAACCAAGAGAGTCGACG–NFQ	NM_008171.3	62	3–4	0.55	43.20 ± 0.08	−3.20 ± 0.04	0.99 ± 0.00	5
Mm00439181_g1	Mouse mNR2C FAM–ACCACACCTTCAGCAGCGGGGATAT–NFQ	NM_010350.2	74	7–8	0.75	46.35 ± 0.22	−3.31 ± 0.12	0.99 ± 0.01	5
Mm01194083_g1	Mouse mNR2D FAM–CGTACCCACAGCCCTCCGCCTGACG–NFQ	NM_008172.2	74	4–5	0.5	42.80 ± 0.42	−3.35 ± 0.08	0.99 ± 0.01	5

Details of all qPCR assays (TaqMan) and their respective standard curve parameters (derived from mouse midbrain tissue) used for RT-qPCR analysis in this study. Assay accession no. is as given by Applied Biosystems. Threshold indicates the value at which real-time PCR cycle thresholds were analyzed and *n* represents the number of independent experiments for standard curve data generation. FAM, Applied Biosystems dye; NCBI, National Center for Biotechnology Information; NFQ, nonfluorescent quencher.