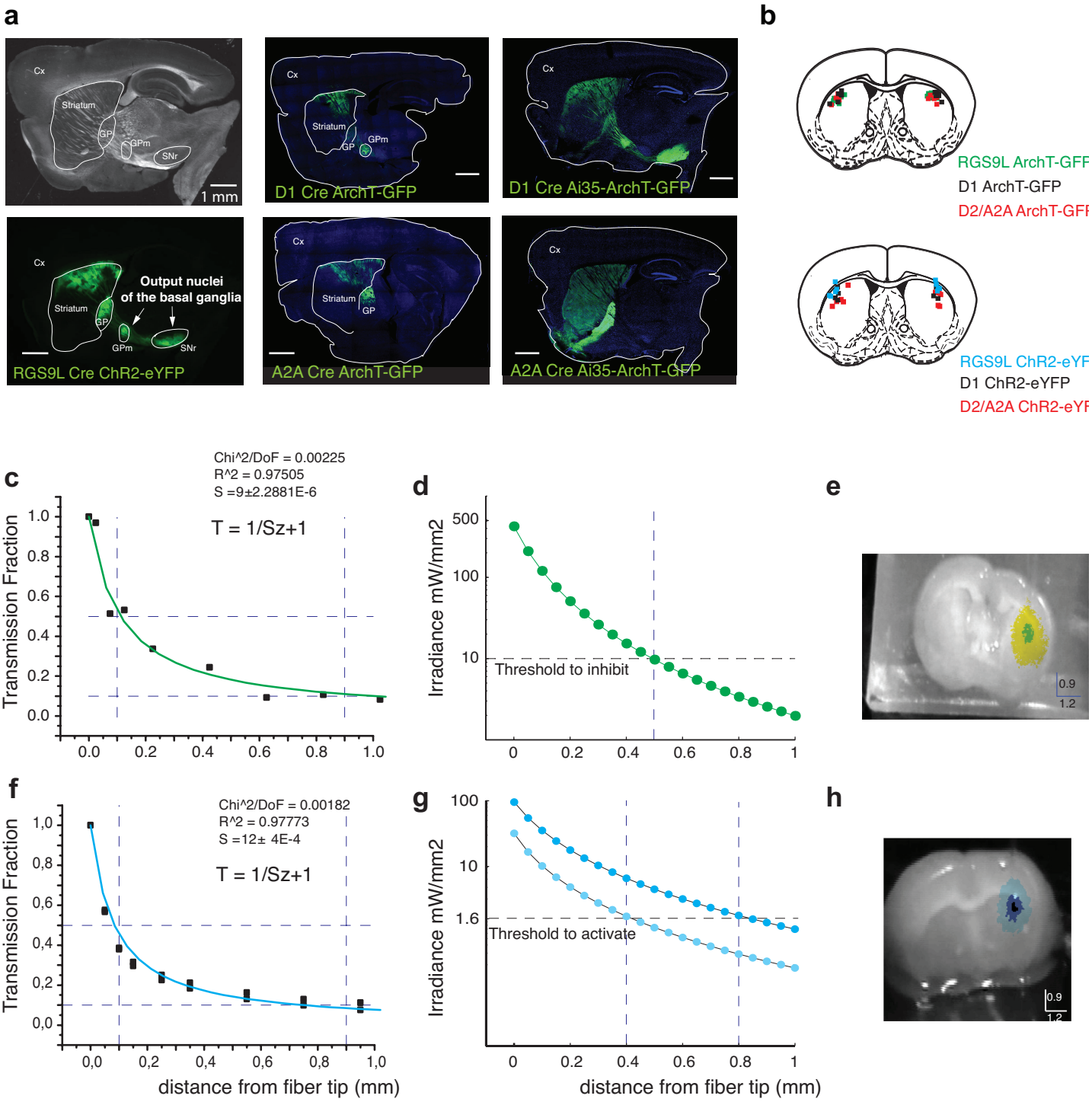
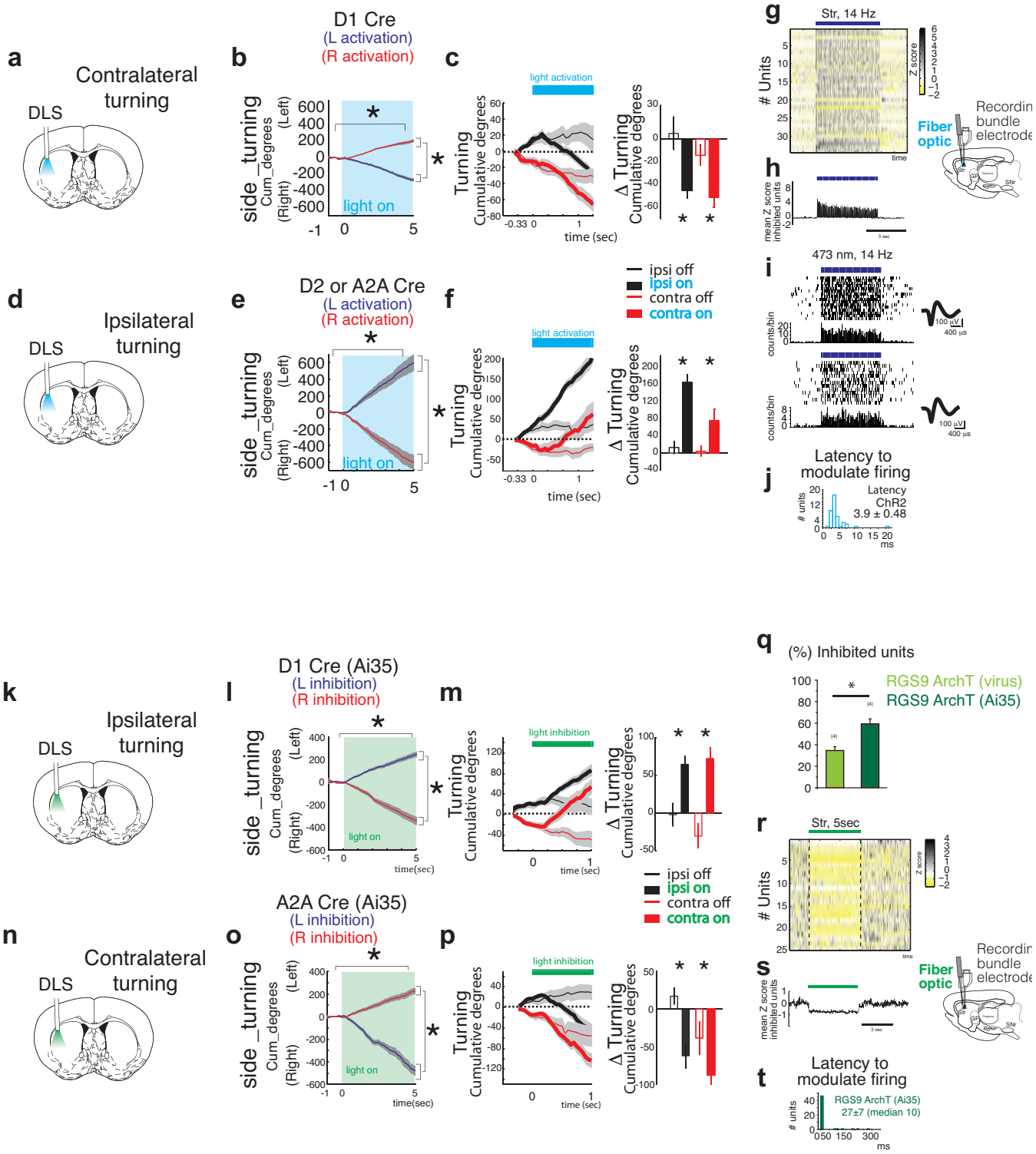
Supplementary Figure 1



Supplementary figure 1. Viral injection and fiber optic placement in the different mouse lines to optogenetically manipulate the activity of striatal projection pathways. (**a**) Photomicrographs from sagittal sections of mouse brain depicting the nuclei of the basal ganglia targeted in this study. The upper left picture shows a sagittal bright field example to illustrate the striatum and the projection target nuclei of the striatal cells (GP: globus pallidus, GPm: medial globus pallidus; SNr: subtantia nigra pars reticulata). The bottom left picture

shows a sagittal brain slice of a ChR2-EYFP injection into the striatum of a RGS9L-Cre mouse. Middle upper and bottom column pictures in a show ArchT-GFP injections into the striatum of D1 Cre (ERY217) and A2A Cre (KG139) mice. Note the presence of green fluorescence in the striatum (place of injection) and in the target nuclei of the striatal cells: simultaneously in the GP, GPm and SNr in the case the RGS9 Cre line showing that it targets both the striatonigral and the striatopallidal pathways, and selectively in GPm (and also SNr in different slices not shown) when using the D1 Cre line (striatonigral pathway) and in the GP when using the A2A cre line (striatopallidal pathway). The right column in a shows sagittal slices from striatonigral and striatopallidal basal ganglia pathways cross bread with the Ai35 line to express ArchT in all the cre expressing cells. Note the difference in the signals between the viral expression of ArchT (middle column in a) and the Ai35 ArchT driven expression (right column in a). Scale horizontal bars in (a) correspond to 1 millimeter. (**b**) Placements of the tip of the optic fibers from the experimental animals in this study for the ArchT-GFP manipulation (upper panel) and ChR2 manipulations (bottom row). (**c, f**) Light transmission through the striatal tissue as a function of distance from the tip of the fiber, for ArchT and ChR2, respectively. (**d, g**) Intensity of the light as a function of distance scaled to the power in the fiber tip, 30 mW in the case of green light (d) and 1 and 3 mW in the case of Chr2 (g). (**e, h**) pictures from brains coronally sectioned an implanted with a fiber optic (immediately after extraction), switching on the same laser power used for the experiments and depicting the spread of light inside the tissue (color), and the threshold range to modulate the activity of cells (darker center color). Scale bars in millimeters.

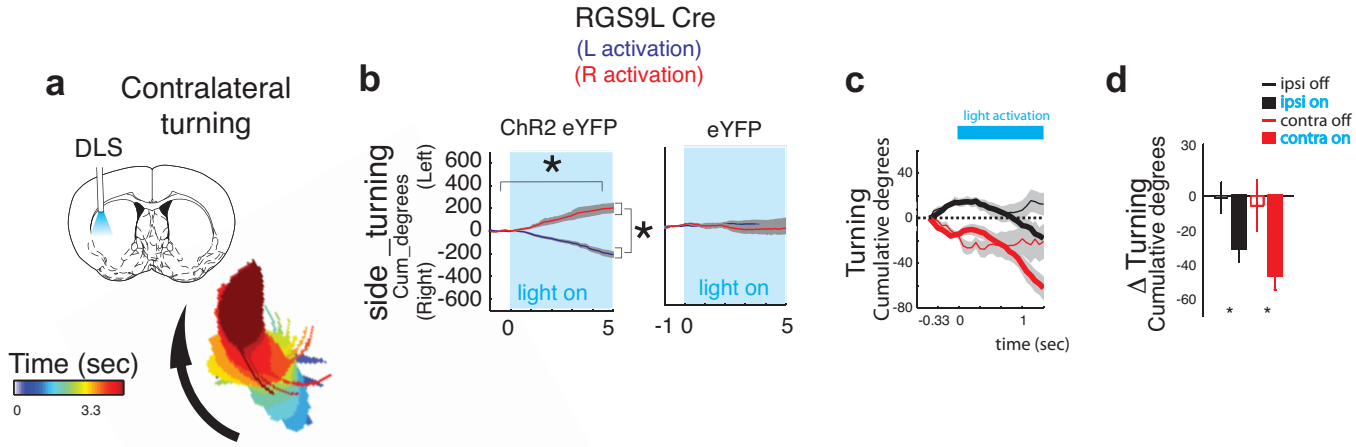
Supplementary Figure 2



Supplementary figure 2. Unbalanced activation or inhibition of the basal ganglia pathways shows opposite effects on contraversive movements. (a, d, k, n) Representation of coronal brain mice slices depicting the unilateral stimulation in the different Cre lines. **(b, e, l, o)** Temporal dynamics of the cumulative degrees of turning towards each side in response to 5 seconds of unilateral illumination to

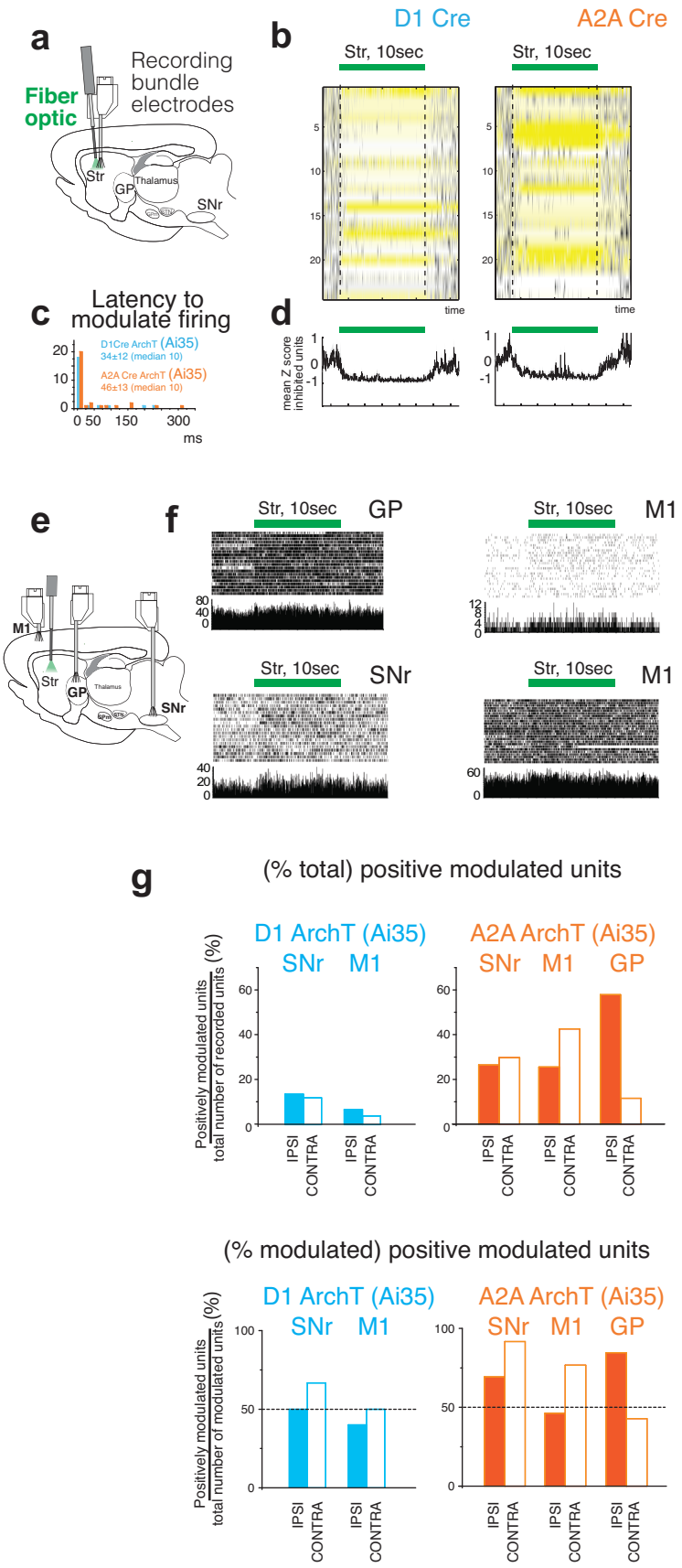
activate (b, e) or inhibit using the Ai35 mice line (l,o) the striatal cells, aligned to one second before the stimulation, dark blue: left side activation; red: right side activation. Data is presented as mean +/- standard error. The asterisk on top of the plots (b, e, l, o) represents $p < 0.05$, for a within-animal Wilcoxon test. **(c, f, m, p)** Left panels, mean +/- standard error of the individual trials from all animals categorized as ipsilateral turning (black color) and contralateral turning (red color) 200 ms before the onset of light. The thick lines correspond to trials receiving light illumination as depicted by the blue or green bar on top of each panel, while the thin lines corresponds to similar segments of time with no light illumination (obtained from periods of 6 seconds before light onset). Right panels, difference between cumulative degrees at the onset of light (0 seconds) and 1.33 seconds after light onset (20 frames later). The asterisk on the right plots (c, f, m, p) corresponds to $p < 0.05$, Mann Whitney U test. Data: D1 Cre-ChR2 ipsi-off: 11 trials from 8 animals (5±14), D1 Cre-ChR2 ipsi-on: 16 trials from 8 animals (-44±7), D1 Cre-ChR2 contra-off: 25 trials from 8 animals (14±8), D1 Cre-ChR2 contra-on 20 trials on from 8 animals (-51±8). D2/A2A Cre-ChR2 ipsi-off: 11 trials from 6 animals (12±15), D2/A2A Cre-ChR2 ipsi-on: 13 trials from 6 animals (166±17), D2/A2A Cre-ChR2 contra-off: 13 trials from 8 animals (4±12), D2/A2A Cre-ChR2 contra-on: 11 trials on from 8 animals (75±27). D1 Cre-ArchT(Ai35) ipsi-off: 11 trials from 2 animals (-5±14), D1 Cre-ArchT(Ai35) ipsi-on: 22 trials from 2 animals (64±11), D1 Cre-ArchT(Ai35) contra-off: 23 trials from 2 animals (-30±15), D1 Cre-ArchT(Ai35) contra-on 16 trials on from 2 animals (72±14). D2/A2A Cre-ArchT(Ai35) ipsi-off: 14 trials from 2 animals (17±11), D2/A2A Cre-ArchT(Ai35) ipsi-on: 13 trials from 2 animals (-62±16), D2/A2A Cre-ArchT(Ai35) contra-off: 9 trials from 2 animals (-38±21), D2/A2A Cre-ArchT(Ai35) contra-on: 10 trials on from 2 animals (-87±12). **(q)** Comparison of proportion of cells been inhibited in animals using the viral ArchT expression vs animals cross bread to expressing ArchT using the Ai35 mice line [Mean % of inhibited units $_{\text{Ai35-ArchT-GFP}}$ = 59±4 % versus the mean % of inhibited units$_{\text{viralArchT-GFP}}$ = 34±7; $p < 0.05$; Mann Whitney test]. **(g, r)** Perievent histograms of the Z scored timestamps of extracellularly recorded action potentials of striatal neurons, relative to onset of light in freely moving mice expressing either ChR2-eYFP or ArchT-GFP respectively, while turning contralateraly or ipsilateral in an open field. **(h, s)** Mean Z-score for the units that were significantly modulated by the blue or green light illumination in g and r, respectively. **(i).** Perievent histograms examples from individual units depicted in g, bottom to top trials **(j, t).** Latency to modulate spikes in ChR2-eYFP or ArchT-GFP expressing striatal cells. D1 Cre: line targeting the striatonigral pathway. D2/A2ACre: line targeting the striatopallidal pathway.

Supplementary Figure 3



Supplementary figure 3. Yet, concurrent over-activation of both striatal pathways favors contraversive movements. (a) Representation of a coronal brain mice slice depicting the unilateral stimulation in b-d**. (b)** Temporal dynamics of the cumulative degrees of turning towards each side in response to 5 seconds of unilateral illumination to activate the striatal cells, aligned to one second before the stimulation, dark blue: left side activation; red: right side activation. Data is presented as mean +/- standard error. Data: RGS9L-ChR2$_{Lside\text{-}activation}$: -187+/-28 cumulative degrees to the left and RGS9L-ChR2$_{Rside\text{-}activation}$: 192+/-45 cumulative degrees to the right; Wilcoxon test, $p < 0.05$, against baseline and left vs. right; n=6; no effect in EYFP controls. **(c)** mean +/- standard error of the individual trials from all animals categorized as ipsilateral turning (black color) and contralateral turning (red color) 200 ms before the onset of light. The thick lines correspond to trials receiving light illumination as depicted by the blue or green bar on top of each panel, while the thin lines corresponds to similar segments of time with no light illumination (obtained from periods of 6 seconds before light onset). **(d)** Difference between cumulative degrees at the onset of light (0 seconds) and 1.33 seconds after light onset (20 frames later). The asterisk on the bottom of the plot corresponds to $p < 0.05$, Mann Whitney U test. RGS9L Cre: line targeting simultaneously the striatonigral and the striatopallidal basal ganglia pathways.

Supplementary Figure 4



Supplementary figure 4. Recordings on the target nuclei of the basal ganglia pathway while unilaterally inhibiting the basal ganglia pathways. (a) Representation of a sagittal brain mice slice depicting the unilateral stimulation and recordings inside the striatum for the data presented in b-d **(b)** Perievent histograms of the Z scored timestamps of extracellular recorded action potentials of striatal neurons, relative to onset of green light in freely moving mice expressing ArchT-GFP using the Ai35 mice line, while turning in an open field. **(c).** Latency to modulate spikes from the data presented in b. **(d)** Mean Z-score for the units that were significantly modulated by the green light illumination. **(e)** Representation of a sagittal brain mice slice depicting the unilateral striatal stimulation and recordings on GP (globus pallidus) or SNr (sustantia nigra pars reticulata) or M1 (motor cortex) for the data presented in f-g. **(f)** Perievent histograms examples from individual units recorded in the target nuclei of the basal ganglia while inhibiting the striatum for the analysis presented in g. **(g)** Upper panels, percentage of positive modulated units from the total number of units recorded in the target nuclei of unilateral striatonigral inhibition (blue) or striatopallidal inhibition (orange). Lower panels correspond to the percentage of positively modulated units in relation to the total number of modulated cells (positive and negative). Data: D1-Ai35-ArchT IPSI Striatal inhibition SNr recordings 15 units, 2 sessions, 1 animal; D1-Ai35-ArchT CONTRA Striatal inhibition SNr recordings 17 units, 2 sessions,1 animal; D1-Ai35-ArchT IPSI Striatal inhibition M1 recordings 60 units, 3 sessions,1 animal; D1-Ai35-ArchT CONTRA Striatal inhibition M1 recordings 52 units, 3 sessions,1 animal. A2A-Ai35-ArchT IPSI Striatal inhibition SNr recordings 34 units, 4 sessions, 2 animal; A2A-Ai35-ArchT CONTRA Striatal inhibition SNr recordings 37 units, 4 sessions, 2 animal; A2A-Ai35-ArchT IPSI Striatal inhibition M1 recordings 47 units, 3 sessions, 2 animal; A2A-Ai35-ArchT CONTRA Striatal inhibition M1 recordings 54 units, 3 sessions, 1 animal; A2A-Ai35-ArchT IPSI Striatal inhibition GP recordings 18 units, 2 sessions, 1 animal; A2A-Ai35-ArchT CONTRA Striatal inhibition GP recordings 26 units, 2 sessions, 1 animal. Note that we used movable arrays that were advanced 200 microns from one session to the next.