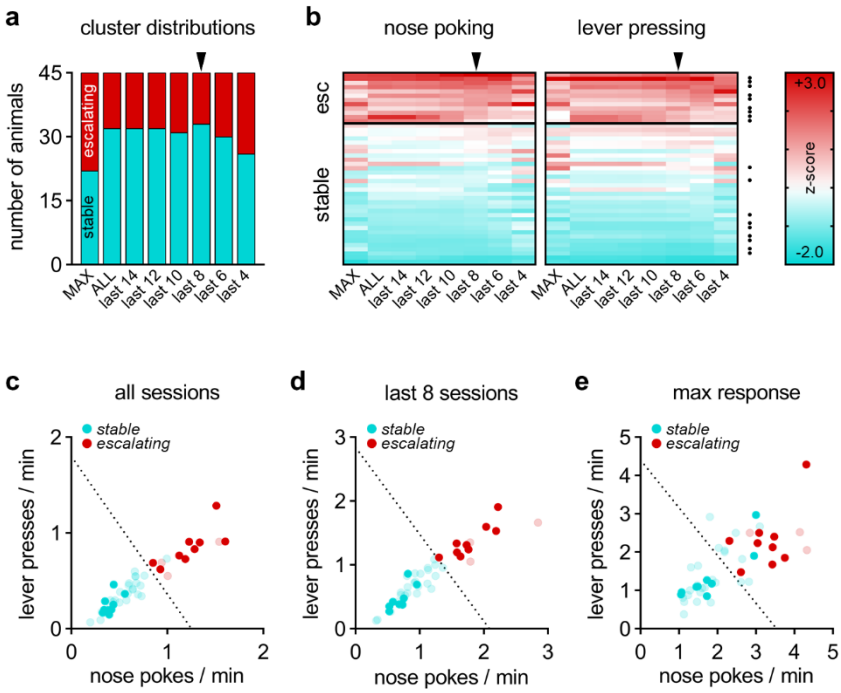
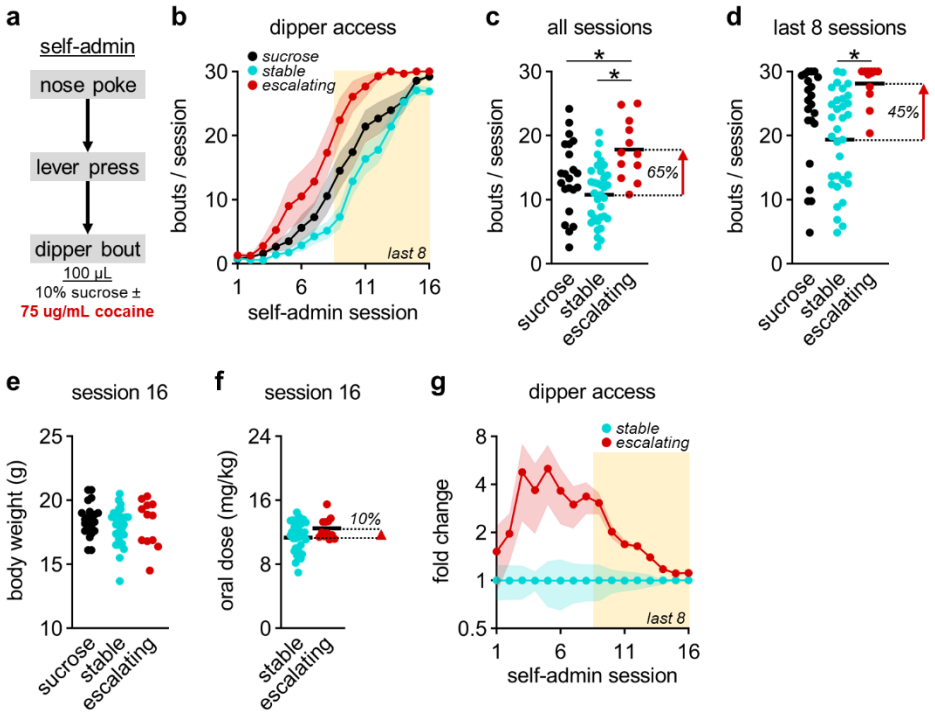
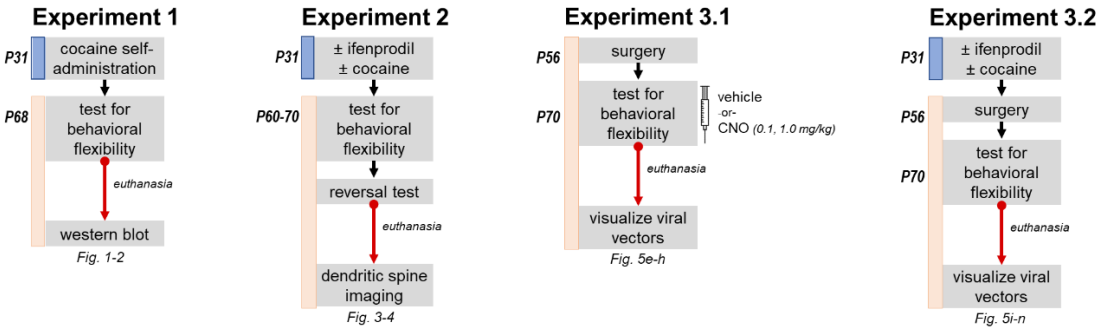


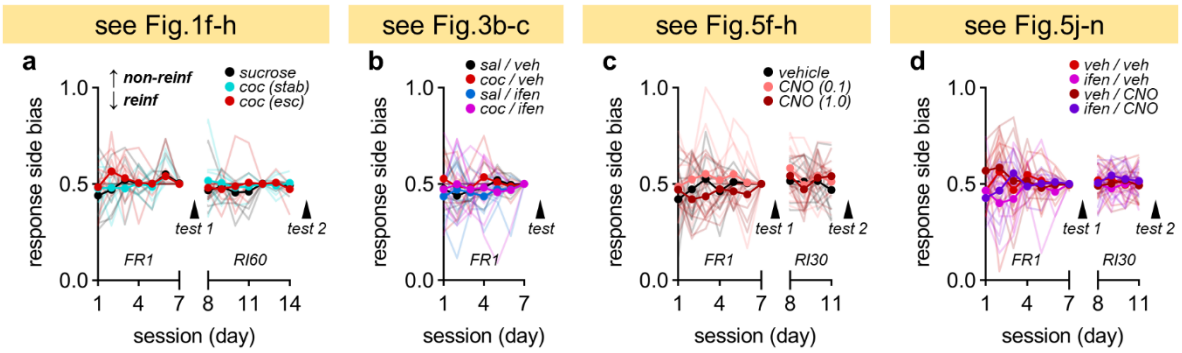
Supplementary Figure 1. Unbiased clustering of cocaine self-administration behavior is stable across response parameters. (a) Summary of *k*-means clustering distributions across response parameters. **(b)** Normalized response rates for individual nose-poking and lever-pressing responses across different clustering parameters. Animals clustered using last 8 sessions in main text (arrows). Animals included in follow-up experiments (dots; *see Fig.1e-h, Fig.2*). **(c-e)** *k*-means clustering results: average across all sessions (NP: $F_{1,43}$=97.2, p<0.001; LP: $F_{1,43}$=75.7, p<0.001), average across last 8 sessions (NP: $F_{1,43}$=87.4, p<0.001; LP: $F_{1,43}$=64.4, p<0.001), or maximum response session (NP: $F_{1,43}$=128, p<0.001; LP: $F_{1,43}$=88.3, p<0.001). Groups (stable *vs.* escalating) denote clustering results from main text (last 8). Animals included in follow-up experiments (solid points; *see Fig.1e-h, Fig.2*). Data presented as individual points.



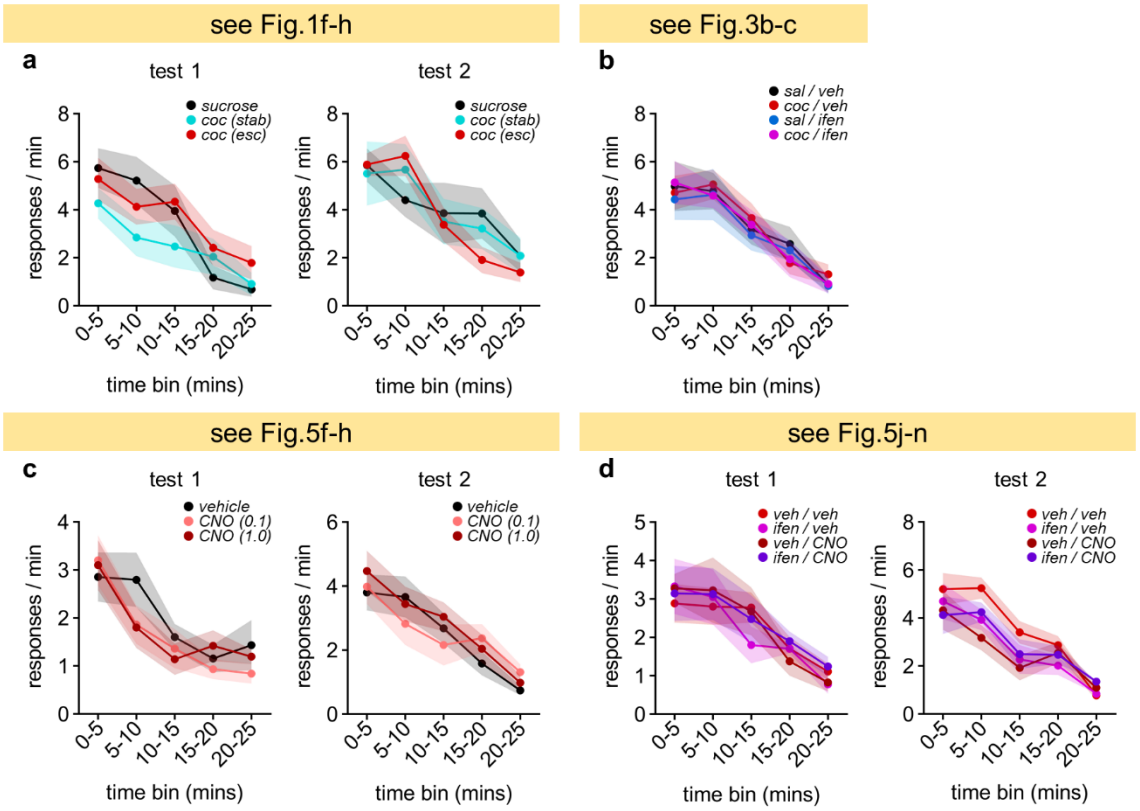
Supplementary Figure 2. Estimates of oral cocaine doses earned per self-administration session. (a) Response sequencing during self-administration sessions. **(b)** Number of dipper access bouts (10-second, 100 µL) per session (session: $F_{15,960}$=134, p<0.001, η^2=0.67; session × group: $F_{30,960}$=2.25, p<0.001, η^2=0.07). Sessions terminated following 30 access bouts were earned. **(c-d)** Dipper bouts earned per session across all ($F_{2,64}$=9.13, p<0.001, η^2=0.22) or last 8 sessions ($F_{2,64}$=7.04, p=0.002, η^2=0.18). Percent difference for escalating *vs.* stable responders noted. **(e)** Body weights during final self-administration session ($F_{2,64}$=1.00, p=0.372, η^2=0.03). **(f)** Maximum oral cocaine dose achievable during final self-administration session (t_{43}=1.94, p=0.059, *d*=0.65). Percent difference for escalating *vs.* stable responders noted. **(g)** Normalized dipper access bouts per session (session: $F_{15,645}$=4.31, p<0.001, η^2=0.09; session × group: $F_{15,645}$=4.31, p<0.001, η^2=0.09). Data presented as individual points or mean ± S.E.M. *p<0.05 (post hoc).



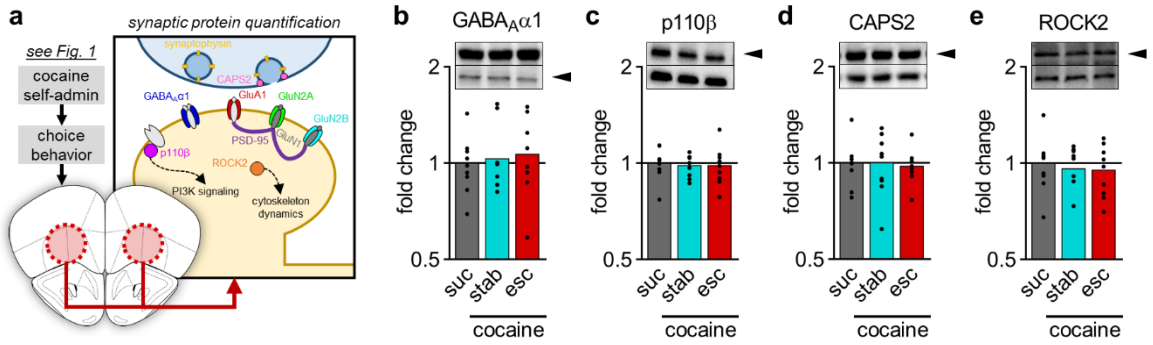
Supplementary Figure 3. Schematic of experiments in this report. Adolescent manipulations are indicated by blue tags, while the peach color indicates events occurring in adulthood. The age at the start of events is indicated at left. “P” refers to postnatal day. The corresponding figures in the main text are indicated at bottom. Experiment 1 utilized cocaine self-administration, while subsequent experiments utilized experimenter-administered cocaine. Experiment 3.1 established an appropriate dose of CNO for use in experiment 3.2.



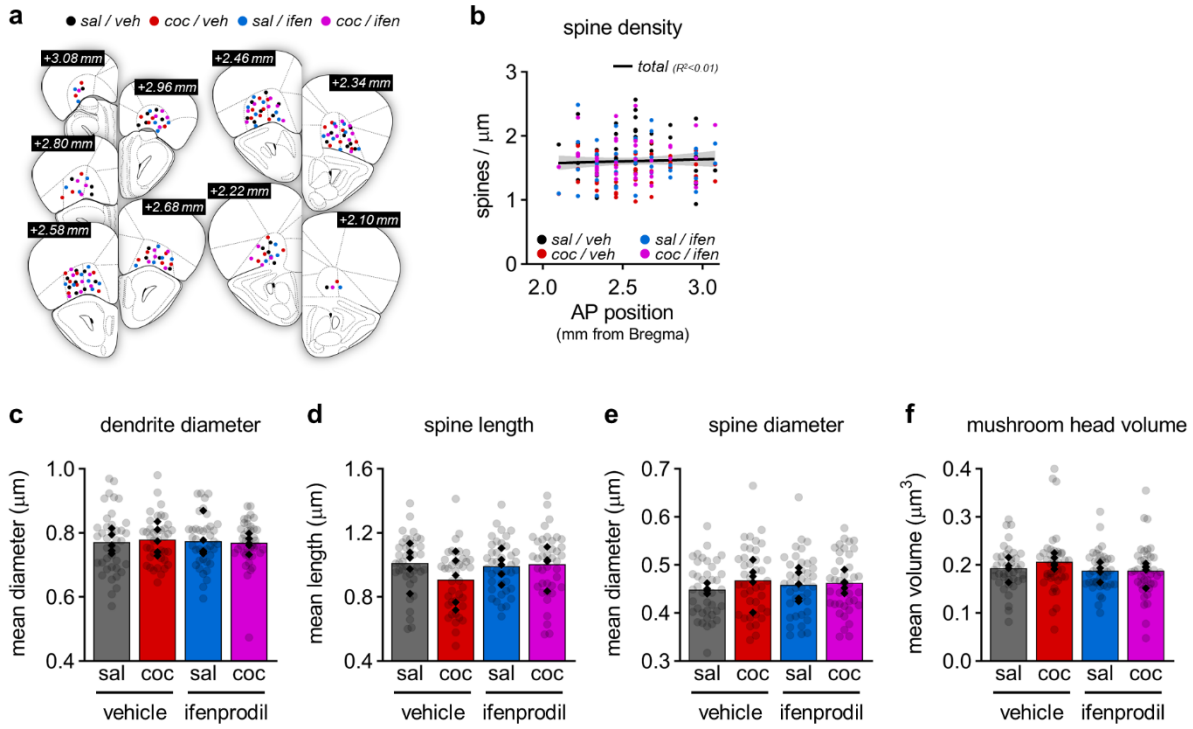
Supplementary Figure 4. Mice do not display preference for one nose-poke aperture during training. Response side bias (responses on aperture to be non-reinforced / total responses) during training sessions. **(a)** Following cocaine self-administration (session: $F_{13,312}=1.71$, $p=0.058$, $\eta^2=0.07$; session × group: $F_{13,312}<1$). **(b)** Cocaine/ifenprodil co-administration (session: $F_{6,198}<1$; session × cocaine: $F_{6,198}<1$; session × ifenprodil: $F_{6,198}=1.07$, $p=0.384$, $\eta^2=0.03$; session × cocaine × ifenprodil: $F_{6,198}<1$). **(c)** BLA→OFC inactivation: CNO dose (session: $F_{10,270}=1.48$, $p=0.145$, $\eta^2=0.05$; session × CNO: $F_{20,270}<1$), or **(d)** cocaine/ifenprodil co-administration (session: $F_{10,290}<1$; session × ifenprodil: $F_{10,290}=1.57$, $p=0.115$, $\eta^2=0.05$; session × CNO: $F_{10,290}<1$; session × ifenprodil × CNO: $F_{10,290}<1$). Data presented as individual points (semi-transparent) and group means (solid). Correspondence to main figures noted.



Supplementary Figure 5. Responding during non-reinforced sessions did not differ between groups prior to choice tests. All non-reinforced sessions were performed drug- and manipulation-free. **(a)** Cocaine self-administration: test 1 (time: $F_{4,96}$=21.8, p<0.001, η^2=0.48; time × group: $F_{8,96}$=1.44, p=0.191, η^2=0.11) or test 2 (time: $F_{4,96}$=26.1, p<0.001, η^2=0.52; time × group: $F_{8,96}$=1.62, p=0.128, η^2=0.12). **(b)** Cocaine/ifenprodil co-administration (time: $F_{4,132}$=43.3, p<0.001, η^2=0.57; time × cocaine: $F_{4,132}$<1; time × ifenprodil: $F_{4,132}$<1; time × cocaine × ifenprodil: $F_{4,132}$<1). **(c)** OFC→BLA inactivation (dose response): test 1 (time: $F_{4,108}$=16.5, p<0.001, η^2=0.38; time × group: $F_{8,108}$<1) or test 2 (time: $F_{4,108}$=17.9, p<0.001, η^2=0.40; time × group: $F_{8,108}$<1). **(d)** OFC→BLA inactivation (cocaine/ifenprodil co-administration): test 1 (time: $F_{4,116}$=13.2, p<0.001, η^2=0.31; time × ifenprodil: $F_{4,108}$<1; time × CNO: $F_{4,108}$<1; time × ifenprodil × CNO: $F_{4,108}$<1) or test 2 (time: $F_{4,116}$=31.2, p<0.001, η^2=0.52; time × ifenprodil: $F_{4,108}$<1; time × CNO: $F_{4,116}$=1.18, p=0.326, η^2=0.04; time × ifenprodil × CNO: $F_{4,108}$<1). Data presented as mean ± S.E.M. Correspondence to main figures noted.



Supplementary Figure 6. Quantification of additional synaptic and plasticity-related protein levels in the OFC following cocaine self-administration. (a) Synaptic localization of additional quantified proteins in the OFC: **(b)** $GABA_A\alpha1$, **(c)** p110β, **(d)** CAPS2, and **(e)** ROCK2 (all $F_{2,24}<1$) Values normalized to loading controls (HSP-70) and expressed as fold change from sucrose controls. Representative blots show target protein (black arrow) and HSP-70 loading controls (no arrow). Data presented as individual points.



Supplementary Figure 7. Additional OFC dendritic spine parameters. (a) Location of sampled dendrites by anterior-posterior (AP) distance from bregma. **(b)** Dendritic spine density across A-P extent of the OFC ($F_{1,158}$<1). 95% confidence interval (shading). **(c)** Dendrite diameter (cocaine: $F_{1,16}$<1; ifenprodil: $F_{1,16}$<1; cocaine × ifenprodil: $F_{1,16}$<1). **(d)** Dendritic spine length (cocaine: $F_{1,16}$<1; ifenprodil: $F_{1,16}$<1; cocaine × ifenprodil: $F_{1,16}$=1.17, p=0.30). **(e)** Dendritic spine diameter (cocaine: $F_{1,16}$<1; ifenprodil: $F_{1,16}$<1; cocaine × ifenprodil: $F_{1,16}$<1). **(f)** Head volume of mushroom-type spines (cocaine: $F_{1,16}$<1; ifenprodil: $F_{1,16}$<1; cocaine × ifenprodil: $F_{1,16}$=2.61, p=0.13). Data presented as individual points (solid=per animal; transparent=per dendrite) or mean ± S.E.M.

Supplementary Table 1

Antibody	Manufacturer	Product	Dilution	Identifier
Rabbit anti-PSD-95	Cell Signaling Technology	#3450	1:1000	RRID AB_2292883
Rabbit anti-Synaptophysin	Abcam	#ab32127	1:10000	RRID AB_2286949
Mouse anti-GluN1	Millipore	#05-432	1:500	RRID AB_390129
Rabbit anti-GluN2A	Invitrogen	#PA5-35377	1:500	RRID AB_2552687
Mouse anti-GluN2B	Novus	#NB100-74475	1:500	RRID AB_1049238
Rabbit anti-GluA1	Abcam	#ab31232	1:1000	RRID AB_2113447
Rabbit anti-$GABA_A\alpha1$	Millipore	#06-868	1:1000	RRID AB_310272
Rabbit anti-p110β	Millipore	#09-482	1:500	RRID AB_1977425
Rabbit anti-CAPS2	Abcam	#ab69894	1:1000	RRID AB_2068181
Rabbit anti-ROCK2	Abcam	#ab71598	1:500	RRID AB_1566688
Mouse anti-HSP-70	Santa Cruz Biotechnology	#sc-7298	1:1000	RRID AB_627761
Rabbit anti-HSP-70	Cell Signaling Technology	#4872	1:1000	RRID AB_2279841
Goat anti-mouse, peroxidase-conjugated	Jackson ImmunoResearch	#115-035-003	1:1000	RRID AB_10015289
Goat anti-rabbit, peroxidase conjugated	Vector Laboratories	#PI-1000	1:10000	RRID AB_2336198

Supplementary Table 2

Figure	Measure	Groups	Analysis	Comparison	Statistic	p-value	Effect size
1b	response rate (NP *vs.* LP)	sucrose *(n=22)* cocaine (stab) *(n=33)* cocaine (esc) *(n=12)*	*k*-means clustering	nose poke	$F_{1,43}$=97.2	p<0.001	n/a
				lever press	$F_{1,43}$=75.7	p<0.001	
1b inset	seek:take		ANOVA	group	$F_{2,64}$=1.63	p=0.204	η^2=0.05
1c left	response rate		RM ANOVA	session	$F_{15,960}$=50.2	p<0.001	η^2=0.44
				session × group	$F_{30,960}$=3.63	p<0.001	η^2=0.10
1c right			ANOVA	group	$F_{2,64}$=51.4	p<0.001	η^2=0.62
			post-hoc: Dunnett	sucrose *vs.* stable	Δ=0.09±0.08	p=0.492	n/a
				sucrose *vs.* escalating	Δ =0.94±0.11	p<0.001	
1d left			RM ANOVA	session	$F_{15,960}$=68.1	p<0.001	η^2=0.52
				session × group	$F_{30,960}$=3.64	p<0.001	η^2=0.10
1d right			ANOVA	group	$F_{2,64}$=32.4	p<0.01	η^2=0.50
			post-hoc: Dunnett	sucrose *vs.* stable	Δ=0.16±0.08	p=0.072	n/a
				sucrose *vs.* escalating	Δ =0.60±0.10	p<0.001	
1f		sucrose *(n=9)* cocaine (stab) *(n=9)* cocaine (esc) *(n=9)*	RM ANOVA	session	$F_{13,312}$=7.39	p<0.001	η^2=0.24
				session × group	$F_{26,312}$=1.10	p=0.338	η^2=0.08
1g			RM ANOVA	reinforcement	$F_{1,24}$=18.54	p<0.001	η^2=0.44
				reinforcement × group	$F_{1,24}$=3.47	p=0.048	η^2=0.22
			post-hoc: paired *t*-test	reinf *vs.* non-reinf (suc)	t_8=3.11	p=0.015	*d*=1.04
				reinf *vs.* non-reinf (stab)	t_8=5.83	p<0.001	*d*=1.94
				reinf *vs.* non-reinf (esc)	t_8=0.29	p=0.777	*d*=0.10
1h			RM ANOVA	reinforcement	$F_{1,24}$=3.56	p=0.078	η^2=0.13
				reinforcement × group	$F_{1,24}$=0.09	p=0.913	η^2<0.01

Supplementary Table 3

Figure	Measure	Groups	Analysis	Comparison	Statistic	p-value	Effect size
2b	fold change	sucrose *(n=9)* cocaine (stab) *(n=9)* cocaine (esc) *(n=9)*	ANOVA	group	$F_{2,24}$=8.31	p=0.002	η^2=0.41
			post-hoc: Tukey HSD	sucrose *vs.* stable	Δ=0.01±0.04	p=0.969	n/a
				sucrose *vs.* escalating	Δ=0.13±0.04	p=0.034	
				stable *vs.* escalating	Δ=0.13±0.04	p=0.004	
2c			ANOVA	group	$F_{2,24}$=2.32	p=0.120	η^2=0.16
2d			ANOVA	group	$F_{2,24}$=0.13	p=0.876	η^2=0.01
2e			ANOVA	group	$F_{2,24}$=0.59	p=0.564	η^2=0.05
2f			ANOVA	group	$F_{2,24}$=4.81	p=0.017	η^2=0.29
			post-hoc: Tukey HSD	sucrose *vs.* stable	Δ=0.14±0.06	p=0.045	n/a
				sucrose *vs.* escalating	Δ=0.01±0.06	p=0.962	
				stable *vs.* escalating	Δ=0.16±0.06	p=0.025	
2g			ANOVA	group	$F_{2,24}$=0.52	p=0.603	η^2=0.04
2h	choice *vs.* protein level	cocaine (stab) *(n=9)* cocaine (esc) *(n=9)*	simple linear regression	cocaine (total)	$F_{1,16}$=11.7	p=0.004	R^2=0.42
				cocaine (stable)	$F_{1,7}$=6.76	p=0.035	R^2=0.49
				cocaine (escalating)	$F_{1,7}$=0.83	p=0.392	R^2=0.11
2i			simple linear regression	cocaine (total)	$F_{1,16}$=12.0	p=0.003	R^2=0.43
				cocaine (stable)	$F_{1,7}$=4.42	p=0.074	R^2=0.39
				cocaine (escalating)	$F_{1,7}$=2.01	p=0.199	R^2=0.22

Supplementary Table 4

Figure	Measure	Groups	Analysis	Comparison	Statistic	p-value	Effect size
3b	response rate	saline/vehicle *(n=9)* cocaine/vehicle *(n=9)* saline/ifenprodil *(n=10)* cocaine/ifenprodil *(n=9)*	RM ANOVA	session	$F_{6,198}$=74.1	p<0.001	η^2=0.69
				session × cocaine	$F_{6,198}$=0.73	p=0.626	η^2=0.02
				session × ifenprodil	$F_{6,198}$=1.69	p=0.124	η^2=0.05
				session × coc × ifen	$F_{6,198}$=1.68	p=0.127	η^2=0.05
3c			RM ANOVA	reinforcement	$F_{1,33}$=44.7	p<0.001	η^2=0.58
				reinforcement × cocaine	$F_{1,33}$=11.7	p=0.002	η^2=0.26
				reinforcement × ifenprodil	$F_{1,33}$=3.56	p=0.068	η^2=0.10
				reinforcement × coc × ifen	$F_{1,33}$=5.85	p=0.021	η^2=0.15
			post-hoc: paired *t*-test	reinf *vs.* non-reinf (sal/veh)	t_8=4.68	p=0.002	*d*=1.56
				reinf *vs.* non-reinf (coc/veh)	t_8=1.33	p=0.219	*d*=0.44
				reinf *vs.* non-reinf (sal/ifen)	t_9=4.16	p=0.002	*d*=1.32
				reinf *vs.* non-reinf (coc/ifen)	t_8=4.56	p=0.007	*d*=1.19
3d	reversal rate		RM ANOVA	session	$F_{3,99}$=76.4	p<0.001	η^2=0.70
				session × cocaine	$F_{3,99}$=2.55	p=0.060	η^2=0.07
				session × ifenprodil	$F_{3,99}$=1.52	p=0.214	η^2=0.04
				session × coc × ifen	$F_{3,99}$=3.71	p=0.014	η^2=0.10
			post-hoc: ANOVA	coc × ifen (session 1)	$F_{1,33}$=6.14	p=0.019	η^2=0.16
				coc × ifen (session 2)	$F_{1,33}$=15.1	p<0.001	η^2=0.31
				coc × ifen (session 3)	$F_{1,33}$=2.76	p=0.106	η^2=0.08
				coc × ifen (session 4)	$F_{1,33}$=1.13	p=0.295	η^2=0.03
3e	response rate		RM ANOVA	session	$F_{3,99}$=10.3	p<0.001	η^2=0.24
				session × cocaine	$F_{3,99}$=1.03	p=0.381	η^2=0.03
				session × ifenprodil	$F_{3,99}$=0.55	p=0.651	η^2=0.02
				session × coc × ifen	$F_{3,99}$=0.06	p=0.983	η^2<0.01
3f			RM ANOVA	session	$F_{3,99}$=40.1	p<0.001	η^2=0.55
				session × cocaine	$F_{3,99}$=1.39	p=0.251	η^2=0.04
				session × ifenprodil	$F_{3,99}$=0.69	p=0.560	η^2=0.02
				session × coc × ifen	$F_{3,99}$=2.78	p=0.045	η^2=0.08
			post-hoc: ANOVA	coc × ifen (session 1)	$F_{1,33}$=6.14	p=0.019	η^2=0.16
				coc × ifen (session 2)	$F_{1,33}$=15.1	p<0.001	η^2=0.31
				coc × ifen (session 3)	$F_{1,33}$=2.76	p=0.106	η^2=0.08
				coc × ifen (session 4)	$F_{1,33}$=1.13	p=0.295	η^2=0.03

Supplementary Table 5

Figure	Measure	Groups	Analysis	Comparison	Statistic	p-value	Effect size
4c	spine density	saline/vehicle *(n=40 dendrites, 5 mice)* cocaine/vehicle *(n=40 dendrites, 5 mice)* saline/ifenprodil *(n=40 dendrites, 5 mice)* cocaine/ifenprodil *(n=40 dendrites, 5 mice)*	2-factor LMM	cocaine	$F_{1,16}$=2.80	p=0.114	n/a
				ifenprodil	$F_{1,16}$=0.35	p=0.560	
				cocaine × ifenprodil	$F_{1,16}$=7.35	p=0.015	
			post-hoc: 1-factor LMM	sal/veh *vs.* coc/veh	$F_{1,8}$=9.86	p=0.014	
				sal/veh *vs.* sal/ifen	$F_{1,8}$=2.22	p=0.175	
				sal/ifen *vs.* coc/ifen	$F_{1,8}$=0.53	p=0.489	
				coc/veh *vs.* coc/ifen	$F_{1,8}$=5.52	p=0.047	
4d	choice *vs.* spine density		simple linear regression	all groups	$F_{1,18}$=4.68	p=0.044	R^2=0.21
4e	spine density		2-factor LMM	cocaine	$F_{1,16}$=5.10	p=0.038	n/a
				ifenprodil	$F_{1,16}$=0.42	p=0.527	
				cocaine × ifenprodil	$F_{1,16}$=6.96	p=0.009	
			post-hoc: 1-factor LMM	sal/veh *vs.* coc/veh	$F_{1,8}$=18.0	p=0.003	
				sal/veh *vs.* sal/ifen	$F_{1,8}$=3.92	p=0.083	
				sal/ifen *vs.* coc/ifen	$F_{1,8}$=0.22	p=0.653	
				coc/veh *vs.* coc/ifen	$F_{1,8}$=5.11	p=0.045	
4f			2-factor LMM	cocaine	$F_{1,16}$=0.13	p=0.721	
				ifenprodil	$F_{1,16}$=0.03	p=0.873	
				cocaine × ifenprodil	$F_{1,16}$=1.12	p=0.306	
4g			2-factor LMM	cocaine	$F_{1,16}$=0.03	p=0.864	
				ifenprodil	$F_{1,16}$=0.83	p=0.376	
				cocaine × ifenprodil	$F_{1,16}$=0.01	p=0.949	
4h	spine-type ratio		2-factor LMM	cocaine	$F_{1,16}$=1.25	p=0.280	
				ifenprodil	$F_{1,16}$=1.08	p=0.315	
				cocaine × ifenprodil	$F_{1,16}$=5.20	p=0.033	
			post-hoc: 1-factor LMM	sal/veh *vs.* coc/veh	$F_{1,8}$=5.10	p=0.037	
				sal/veh *vs.* sal/ifen	$F_{1,8}$=0.13	p=0.727	
				sal/ifen *vs.* coc/ifen	$F_{1,8}$=0.10	p=0.764	
				coc/veh *vs.* coc/ifen	$F_{1,8}$=4.94	p=0.042	

Supplementary Table 6

Figure	Measure	Groups	Analysis	Comparison	Statistic	p-value	Effect size
5f	response rate	vehicle *(n=10)* CNO 0.1 mg/kg *(n=10)* CNO 1.0 mg/kg *(n=10)*	RM ANOVA	session	$F_{10,270}$=140	p<0.001	η^2=0.84
				session × CNO	$F_{20,270}$=0.65	p=0.871	η^2=0.05
5g			RM ANOVA	reinforcement	$F_{1,27}$=32.4	p<0.001	η^2=0.55
				reinforcement × CNO	$F_{2,27}$=4.92	p=0.015	η^2=0.27
			post-hoc: paired t-test	reinf *vs.* non-reinf (veh)	t_9=4.92	p<0.001	*d*=1.56
				reinf *vs.* non-reinf (CNO 0.1)	t_9=3.09	p=0.013	*d*=0.98
				reinf *vs.* non-reinf (CNO 1.0)	t_9=1.40	p=0.194	*d*=0.44
5h			RM ANOVA	reinforcement	$F_{1,27}$=15.8	p<0.001	η^2=0.37
				reinforcement × CNO	$F_{2,27}$=2.39	p=0.111	η^2=0.15
			*post-hoc: paired t-test	reinf *vs.* non-reinf (veh)	t_9=3.19	p=0.011	*d*=1.01
				reinf *vs.* non-reinf (CNO 0.1)	t_9=1.69	p=0.126	*d*=0.53
				reinf *vs.* non-reinf (CNO 1.0)	t_9=1.73	p=0.118	*d*=0.55
5j		*cocaine* + vehicle/vehicle *(n=9)* vehicle/CNO *(n=8)* ifenprodil/vehicle *(n=8)* ifenprodil/CNO *(n=8)*	RM ANOVA	session	$F_{10,290}$=189	p<0.001	η^2=0.87
				session × ifenprodil	$F_{10,290}$=1.10	p=0.365	η^2=0.04
				session × CNO	$F_{10,290}$=0.80	p=0.626	η^2=0.03
				session × ifen × CNO	$F_{10,290}$=0.13	p=0.999	η^2<0.01
5k			RM ANOVA	reinforcement	$F_{1,29}$=3.75	p=0.063	η^2=011
				reinforcement × ifenprodil	$F_{1,29}$=5.39	p=0.027	η^2=0.16
				reinforcement × CNO	$F_{1,29}$=5.87	p=0.022	η^2=0.17
				reinforcement × ifen × CNO	$F_{1,29}$=4.38	p=0.045	η^2=0.13
			post-hoc: paired t-test	reinf *vs.* non-reinf (veh/veh)	t_8=0.03	p=0.974	*d*=0.01
				reinf *vs.* non-reinf (veh/CNO)	t_7=3.18	p=0.016	*d*=1.12
				reinf *vs.* non-reinf (ifen/veh)	t_7=0.37	p=0.726	*d*=0.13
				reinf *vs.* non-reinf (ifen/CNO)	t_7=0.20	p=0.845	*d*=0.07
5l	preference ratio		ANOVA	ifenprodil	$F_{1,29}$=8.08	p=0.008	η^2=0.22
				CNO	$F_{1,29}$=4.92	p=0.035	η^2=0.15
				ifenprodil × CNO	$F_{1,29}$=4.79	p=0.037	η^2=0.14
			post-hoc: unpaired *t*-test	veh/veh *vs.* ifen/veh	t_{15}=2.13	p=0.050	*d*=1.03
				veh/veh *vs.* veh/CNO	t_{15}=0.06	p=0.950	*d*=0.03
				veh/CNO *vs.* ifen/CNO	t_{14}=0.39	p=0.702	*d*=0.20
				ifen/veh *vs.* ifen/CNO	t_{14}=2.20	p=0.045	*d*=1.10
5m	response rate		RM ANOVA	reinforcement	$F_{1,29}$=3.13	p=0.088	η^2=0.10
				reinforcement × ifenprodil	$F_{1,29}$=0.10	p=0.761	η^2<0.01
				reinforcement × CNO	$F_{1,29}$=0.33	p=0.138	η^2=0.07
				reinforcement × ifen × CNO	$F_{1,29}$=0.14	p=0.716	η^2=0.01
5n	preference ratio		ANOVA	ifenprodil	$F_{1,29}$=0.10	p=0.759	η^2<0.01
				CNO	$F_{1,29}$=4.37	p=0.046	η^2=0.13
				ifenprodil × CNO	$F_{1,29}$=0.22	p=0.641	η^2=0.01

*planned comparison