Supplementary Table 1. NovaScreen assay. (+)-Naloxone (0.1 uM and 10 uM) has no reliable activity at a broad range of neuronal targets, including neurotransmitters, steroids, ion channels, second messengers, growth factors, hormones, peptides, and enzymes. Values are expressed at the percent inhibition of specific binding; % control values below 50% are considered inactive by the contract laboratory (Caliper Life Sciences).

	(-)- Naloxone		(+)- Naloxone	
% Inhibition at:	1×10^{-7} M	1×10^{-5} M	1×10^{-7} M	1×10^{-5} M
Neurotransmitter Related				
Adenosine, Non-selective	-4.6%	-7.5%	4.2%	-0.8%
Adrenergic, Alpha 1, Non-selective	3.7%	9.8%	0.6%	2.6%
Adrenergic, Alpha 2, Non-selective	0.4%	5.6%	-6.8%	0.9%
Adrenergic, Beta1	7.4%	22.1%	7.6%	17.6%
Cannabinoid, CB1	2.2%	7.0%	3.5%	15.0%
Cannabinoid, CB2	12.3%	18.7%	9.2%	13.1%
Dopamine, D4.2	25.2%	28.3%	10.6%	-11.1%
GABA A, Agonist Site	0.8%	-0.6%	1.6%	2.2%
GABA A, BDZ, alpha 1 site	0.04%	11.4%	1.6%	0.7%
GABA-B	8.3%	-2.3%	15.4%	14.3%
Glutamate, AMPA Site (Ionotropic)	-0.3%	-1.0%	-0.4%	2.6%
Glutamate, Kainate Site (Ionotropic)	4.9%	-0.9%	-0.4%	2.4%
Glutamate, NMDA Agonist Site (Ionotropic)	-2.5%	4.2%	9.7%	8.4%
Glutamate, NMDA, Phencyclidine Site (Ionotropic)	8.5%	4.6%	4.1%	2.5%
Glutamate, mGluR1 (Metabotropic)	-4.0%	5.9%	-0.3%	0.8%

	(-)- Naloxone		(+)- Naloxone	
% Inhibition at:	1×10^{-7} M	1×10^{-5} M	1×10^{-7} M	1×10^{-5} M
Neurotransmitter Related				
Glutamate, mGluR5 (Metabotropic)	-12.2%	-2.0%	4.7%	5.1%
Glutamate, NMDA, Glycine (Stry-insens Site) Ionotropic)	-8.2%	-11.8%	5.7%	5.7%
Glycine, Strychnine-sensitive	9.0%	6.5%	25.1%	40.7%
Histamine, H1	2.7%	5.0%	5.8%	5.7%
Histamine, H2	10.8%	19.6%	-12.1%	9.7%
Histamine, H3	-3.2%	15.9%	-13.0%	10.9%
Muscarinic, M1 (	-5.5%	1.6%	-4.1%	8.3%
Muscarinic, M2 (	-0.3%	17.4%	6.4%	3.9%
Muscarinic, Non-selective, Central	0.8%	3.7%	1.1%	4.2%
Muscarinic, Non-selective, Peripheral	9.8%	-2.6%	-9.2%	9.9%
Nicotinic, Muscle (a-BnTx sensitive)	2.5%	10.7%	-6.8%	-5.6%
Nicotinic, Neuronal [a-BnTx insensitive]	-0.5%	9.7%	-14.0%	-3.0%
Opioid, Kappa 1	93.9%	97.4%	6.1%	26.3%
Opioid, Mu (h)	94.9%	100.0%	13.7%	13.0%

	(-)- Naloxone		(+)- Naloxone	
% Inhibition at:	1×10^{-7} M	1×10^{-5} M	1×10^{-7} M	1×10^{-5} M
Steroids				
Estrogen	11.5%	1.6%	-9.6%	-2.0%
Glucocorticoid	-1.0%	20.0%	-3.1%	0.8%
Testosterone (cytosolic)	-1.8%	-12.0%	12.7%	7.7%
Ion Channels				
Calcium Channel, Type L (Benzothiazepine Site)	10.3%	13.6%	17.0%	19.8%
Calcium Channel, Type L (Dihydropyridine Site)	13.6%	9.1%	-5.8%	14.4%
Calcium Channel, Type N	-4.4%	-2.0%	-2.9%	3.0%
Potassium Channel, ATP-Sensitive	10.3%	13.6%	17.0%	19.8%
Potassium Channel, Ca2+ Act., VI	13.6%	9.1%	-5.8%	14.4%
Sodium, Site 2	-4.4%	-2.0%	-2.9%	3.0%
Second Messengers				
Nitric Oxide, NOS (Neuronal-Binding)	9.9%	11.2%	-0.8%	-0.9%
Prostoglandins				
Leukotriene, LTB4 (BLT)	-8.8%	1.6%	-1.7%	1.9%
Leukotriene, LTD4 (CysLT1)	-17.0%	-13.1%	-3.9%	-11.4%
Thromboxane A2	8.5%	1.4%	-5.3%	1.9%

	(-)- Naloxone		(+)- Naloxone	
% Inhibition at:	1×10^{-7} M	1×10^{-5} M	1×10^{-7} M	1×10^{-5} M
Growth Factors/Hormones				
Corticotropin Releasing Factor, Non-selective	-5.4%	-3.6%	3.1%	1.6%
Oxytocin	6.6%	1.9%	-3.9%	4.5%
Platelet Activating Factor, PAF	-19.5%	-7.3%	11.4%	4.2%
Thyrotropin Releasing Hormone, TRH	12.2%	-0.5%	-3.4%	6.4%
Brain/Gut Peptides				
Angiotensin II, AT1 (h)	-1.2%	-1.6%	5.9%	-6.1%
Angiotensin II, AT2	7.3%	3.4%	0.9%	10.6%
Bradykinin, BK2	-4.8%	-9.0%	-3.9%	-9.4%
Cholecystokinin, CCK1 (CCKA)	-4.5%	3.0%	-13.3%	-8.1%
Cholecystokinin, CCK2 (CCKB)	0.9%	3.0%	5.0%	6.4%
Endothelin, ET-A (h)	-12.8%	4.9%	0.0%	-0.3%
Endothelin, ET-B (h)	1.0%	-2.7%	-12.0%	-2.9%
Galanin, Non-Selective	-3.0%	-16.9%	-6.8%	-14.1%
Neurokinin, NK1	0.0%	-0.6%	-1.4%	-1.4%
Neuroknin, NK2 (NKA) (h)	-9.9%	8.5%	-3.4%	-10.5%
Neurokinin, NK3 (NKB)	6.9%	9.7%	16.3%	16.4%
Vasoactive Intestinal Peptide, Non-selective	13.2%	14.7%	0.6%	2.2%
Vasopressin 1	7.1%	7.9%	-2.2%	-11.9%

% Inhibition at:	(-)- Naloxone 1×10^{-7} M	(-)- Naloxone 1×10^{-5} M	(+)- Naloxone 1×10^{-7} M	(+)- Naloxone 1×10^{-5} M
Enzymes				
Decarboxylase, Glutamic Acid	1.5%	-6.0%	1.0%	3.6%
Esterase, Acetylcholine (h)	3.8%	4.8%	4.5%	2.4%
Oxidase, MAO-A, Peripheral	4.7%	7.2%	1.2%	6.4%
Oxidase, MAO-B, Peripheral	0.0%	9.0%	-4.2%	-1.6%
Transferase, Choline Acetyl	6.8%	26.8%	1.1%	4.0%

<u>Supplementary Table 2.</u> Dopamine transporter and sigma1 receptor assays support that (+)-naloxone does not reliably bind to those sites. Both (+)- and (-)-naloxone failed to displace [^{3}H]WIN 35,428 and [^{3}H](+)-pentazocine from the dopamine transporter in rat striatum and sigma receptors from guinea pig brain, respectively. Historical values for cocaine and haloperidol are also provided for DAT and sigma receptor binding as positive controls. As indicated by the table values, (+)-naloxone failed to reliably binding at any of these sites. *Historical values from previously conducted studies in this laboratory using identical conditions. ^Values from previously conducted studies in this laboratory using identical conditions (Garces-Ramiriez, et al., 2011)

Compound	DAT Ki Value (nM)	$Sigma_1$ Receptor Ki Value (nM)	$Sigma_2$ Receptor Ki Value (nM)
(+)-Naloxone	>10,000	>10,000	>10,000
(-)-Naloxone	>10,000	>10,000	>10,000
Cocaine	76.6 (72.6-80.5)^	5,190 (3,800-7060)	19,300 (16,000-23,300)
Haloperidol	NT	2.91 (2.69-3.14)*	19.6 (15.6-24.6)*

<u>Supplementary Table 3.</u> Biogenic amine transporter assays support that (+)-naloxone does not reliably affect their binding or function. (+)-Naloxone and cocaine (positive control) were tested (2-3 tests/dose) by a contract research laboratory (Research Service, R&D22, Dept. of Veterans Affairs Medical Center, Portland, OR) for their effects on radioligand ([^{125}I]RTI-55) binding to, and transporter specific neurotransmitter uptake by, human dopamine (hDAT), serotonin (hSERT), and norepinephrine (hNET) transporters stably over-expressed in human embryonic kidney (HEK) cells. The Ki value for [^{125}I]RTI-55 displacement and, when significant displacement was found, the Hill coefficient were calculated. When [^{125}I]RTI-55 displacement was measurable (i.e., <10 uM), the IC_{50} for radiolabeled neurotransmitter uptake was also calculated. As indicated by the table values, (+)-naloxone failed to reliably affect the binding or function of any of the biogenic amine transporters.

HEK-hDAT cells	**33,113**	**Cocaine**
[^{125}I]RTI-55 Binding Ki (nM)	>10 μM	411 ± 61
Hill coefficient		-1.2 ± 0.1
[^{3}H]Dopamine Uptake IC_{50} (nM)		
HEK-hSERT cells	**33,113**	**Cocaine**
[^{125}I]RTI-55 Binding Ki (nM)	>8,300	385 ± 66
Hill coefficient		-1.12 ± 0.1
[^{3}H]Serotonin Uptake IC_{50} (nM)	>10 μM	319 ± 36
HEK-hNET cells	**33,113**	**Cocaine**
[^{125}I]RTI-55 Binding Ki (nM)	>7,100	632 ± 51
Hill coefficient		-1.0 ± 0.1
[^{3}H]NE Uptake IC_{50} (nM)	>10 μM	445 ± 43