## **Supplementary Tables**

In situ hybridization probes

Gene	Genbank Accession Number	Clone ID	Cat. No
Etv1	BC005645	PCR	
Etv4	BU053662	5062538	EMM1002-5544031
Etv5	BC034680	4036564	MMM1013-7511524
Fev	AI876263	UI-M-BH3- AVJ-B-02-0-UI	EMM1002-2537418
Elk1	BC054474	30045506	MMM1013-9201805

## **Antibodies**

Antigen	Species	Supplier/ Catalog No.	Dilution
DAT; Slc6a3	Rat, monoclonal	Millipore: MAB369	1:500
TH	Rabbit	Millipore: AB152	1:500
Vmat2; Slc18a2	Rabbit	Millipore: AB1598P	1:500
Nurr1; Nr4a2	Rabbit	Santa Cruz: SC-5568	1:500

**Supplementary Table 1. In situ hybridization probes and antibodies.** Clones containing probes were obtained from Open Biosystems and the corresponding catalog numbers are given. All antibodies gave staining patterns consistent with the mRNA expression of the corresponding gene determined by in situ hybridization in our experiments or in available expression databases. DA pathway antigens also produced signal in known targets of DA innervation, such as the striatum. Ectopic staining outside of these known areas was not observed.

## Allen atlas SN/VTA expression Other E15.5 CNS **Other Adult CNS** E11.5 E13.5 E15.5 expression expression NE Olf \_ -NE NE NE NE NE. NF.

Gene

**Ehf** 

Elf1

Elf2

Elf3	-	-	-	NE	NE
Elf4	-	-	-	NE	NE
Elf5	-	-	-	NE	Diffuse?
*Elk1	-	-	-	NE	Hip
Elk3	-	-	-	NE	CB, Hip, Olf?
Elk4	-	-	ND	NE	NE
*Etv1/ Er81	-	-	-	Ctx, Hab, IO	CB, Ctx, Hab, Hip
Erf	-	-	-	NE	NE
Erg/ V-ets	-	-	-	NE	NE
Ets1	-	-	-	NE	NE
*Ets2	-	-	-	NE	Ctx
Etv2/ Etsrp71	-	-	-	NE	NE
*Etv5/ Erm	-	-	-	NE	CB, Ctx, Hip, Olf, Str, SN, VTA
Etv6/ Tel	-	-	-	NE	NE
Fev/ Pet1	ND	ND	_	NE	5HT
Fli1	-	-	ND	NE	NE
Gabpa	-	-	-	NE	NE
Etv3/ Pe1	-	-	-	NE	NE
*Etv4/ Pea3	-	-	-	NE	NE
*Spedf/ Pse	-	-	-	NE	NE
Sfpi1/ Pu.1	-	-	-	NE	NE
Spib	-	-	-	NE	NE
Spic	_	_	_	NE	NE

**Supplementary Table 2. Ets gene family expression in developing and adult CNS.** 5HT, serotonergic neurons; CB, cerebellum; Ctx, cerebral cortex; Hab, habenula; Hip, hippocampal region; IO, inferior olive; Str, striatum; SN, substantial nigra; VTA, ventral tegmental area. ND, no data; NE, no expression. Mouse Ets family members correspond closely to those identified in

the human (Oikawa and Yamada, 2003; Hollenhorst et al., 2007), with the exception of Etv7, which does not have an identified mouse transcript in NCBI. \*See also Supplemental Figures S3-S6.

Hollenhorst PC, Shah AA, Hopkins C, Graves BJ (2007) Genome-wide analyses reveal properties of redundant and specific promoter occupancy within the ETS gene family. Genes Dev 21:1882-1894.

Oikawa T, Yamada T (2003) Molecular biology of the Ets family of transcription factors. Gene 303:11-34.

**Supplementary Figure 1. Etv5 expression in adult mouse brain.** The expression of Etv5 in adult mouse brain was visualized by ISH in coronal sections. (A) cerebral cortex, (B) ventral midbrain. SN, substantia nigra. Scale is 250µm.

Supplementary Figure 2. Projections of ventral midbrain DA neurons in Etv5<sup>-/-</sup> mice.

Tyrosine hydroxylase (TH) immunostaining was used to visualize the projections of DA neurons in coronal sections of adult mice (diagram). (A,C) proximal part of medial forebrain bundle in control (WT, A) and Etv5<sup>-/-</sup> (KO, C) mice. (B,D) dorsal striatum of control (B) and Etv5<sup>-/-</sup> mice (D). Scale is 100µm.

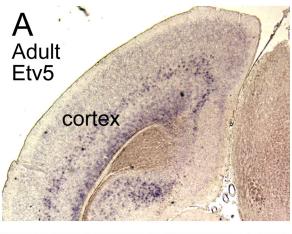
**Supplementary Figure 3.** Expression of Ets family members in GenePaint embryo maps at E14.5. Sagittal sections near the midline are shown. Expression patterns of TH and Nurr1 mRNA serve to localize precursors of the DA neurons of the SN/VTA. Expression of Ets family transcription factors, including Etv1, Etv4, Etv5, Spdef, Fev, Ets2 and Elk1 cannot be detected in the ventral midbrain in corresponding sections. Etv5 signal in the cerebral cortex and lung confirms effective hybridization with this probe. Ctx, cerebral cortex; DRG, dorsal root ganglia; Hb, habenula; Hyp, hypothalamus; OB, olfactory bulb; SC, superior colliculus; SG, sympathetic ganglia; SN/VTA, substantia nigra/ventral tegmental area.

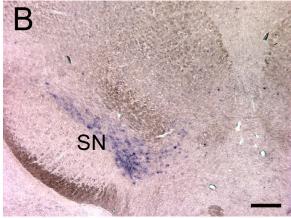
Supplementary Figure 4. Expression of Ets family transcripts in Allen Developing Mouse Brain Atlas at E11.5. Sagittal sections near the midline are shown. TH and Nurr1 expression identifies precursors of the DA neurons of the SN/VTA. None of these Ets transcription factors

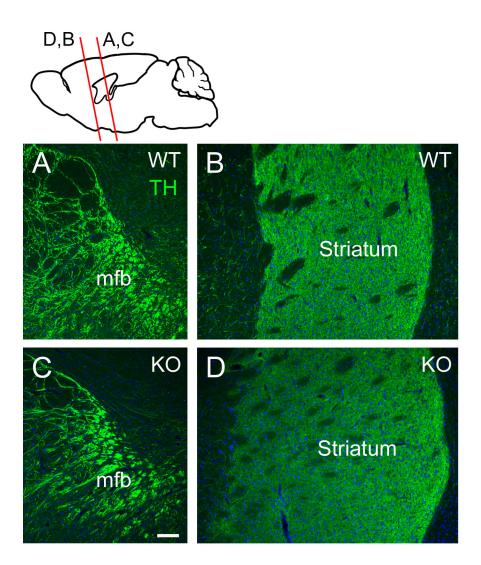
are detected in the ventral midbrain at this stage. SC, superior colliculus; SG, sympathetic ganglia; SN/VTA, substantia nigra/ventral tegmental area; Tel, telencephalon.

Supplementary Figure 5. Expression of Ets family transcripts in Allen Developing Mouse Brain Atlas at E13.5. Sagittal sections near the midline are shown. TH and Nurr1 expression identifies the DA neurons of the SN/VTA. None of these Ets transcription factors are detected in the ventral midbrain at this stage. Hb, habenula; OB, olfactory bulb; SC, superior colliculus; SN/VTA, substantia nigra/ventral tegmental area.

**Supplementary Figure 6.** Expression of Ets family transcripts in Allen Developing Mouse Brain Atlas at E15.5. Sagittal sections near the midline are shown. TH and Nurr1 expression identifies the DA neurons of the SN and VTA. None of these Ets transcription factors are detected in the ventral midbrain at this stage. Hb, habenula; Hyp, hypothalamus; OB, olfactory bulb; SC, superior colliculus; SN/VTA, substantia nigra/ventral tegmental area.





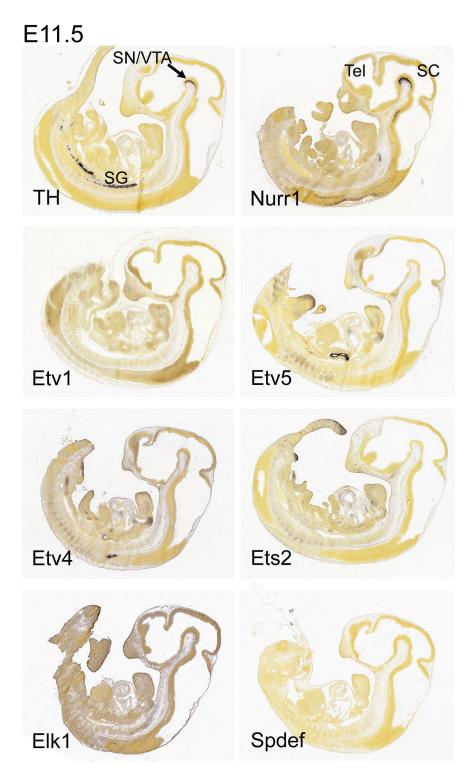


E14.5 GenePaint Hb. SN/VTA Нур DRG sq' Nurr1 TH Etv1 Ctx lung Spdef Etv4 Etv5

Ets2

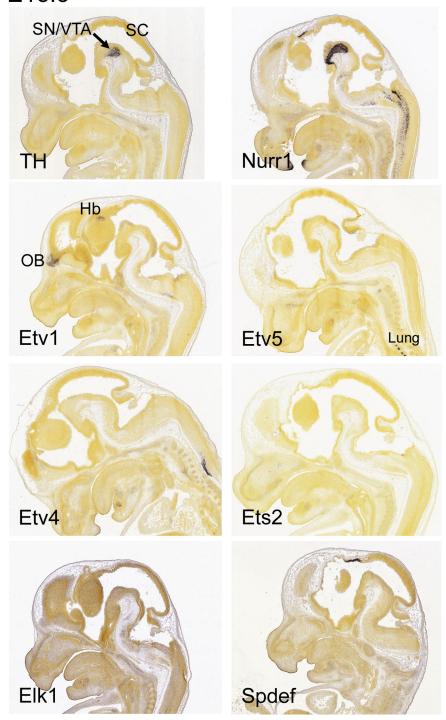
Fev

Elk1



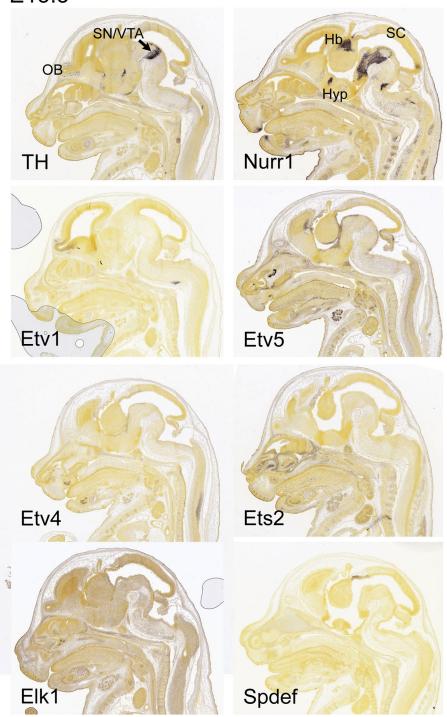
Wang and Turner Supplementary Figure 4

E13.5



Wang and Turner Supplementary Figure 5

## E15.5



Wang and Turner Supplementary Figure 6