Supplementary Information

Identification of distinct telencephalic progenitor pools for neuronal diversity in the amygdala

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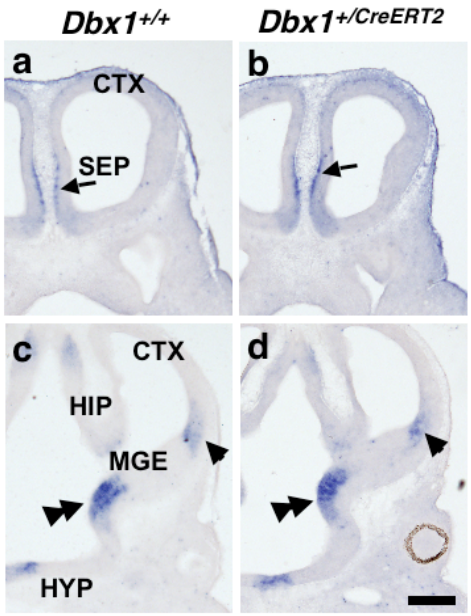
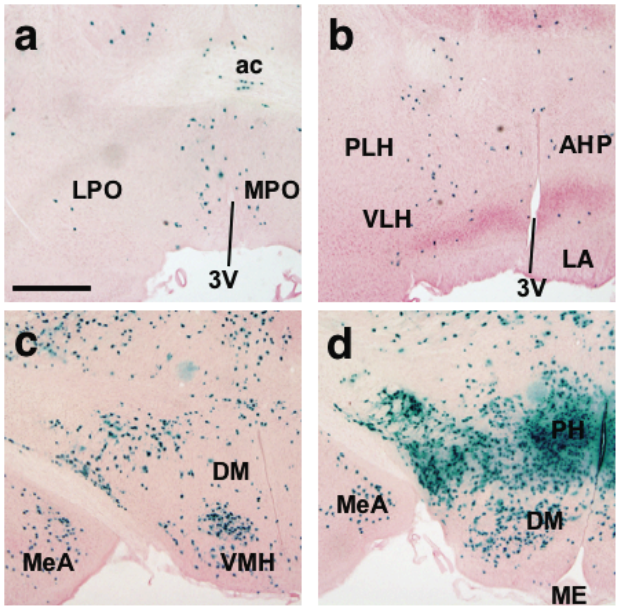
Supplementary Figure 1



Expression of *Dbx1* in knockin and control embryos

Expression of *Dbx1* in wild type (**a, c**) and *Dbx1*$^{+/CreERT2}$ knockin (**b, d**) embryos at 11.5 is shown at rostral and caudal levels in coronal sections. To examine endogenous *Dbx1* expression a *Dbx1* RNA probe was generated to the 5' coding region. In knockin embryos *Dbx1* expression was detected in the septum (arrow in **b**), ventral pallium (arrowhead in **d**), preoptic area (double arrowhead in **d**), in a pattern resembling that of control (**a, c**). Scale bar; 250 μm

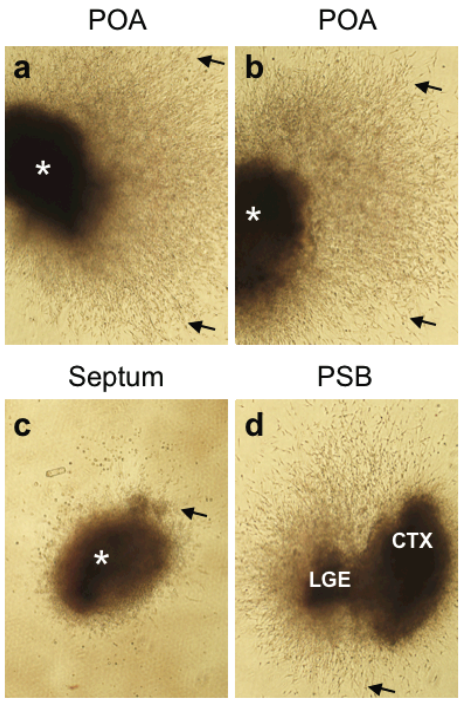
Supplementary Figure 2



Contribution of *Dbx1*-derived cells to the post-natal diencephalon

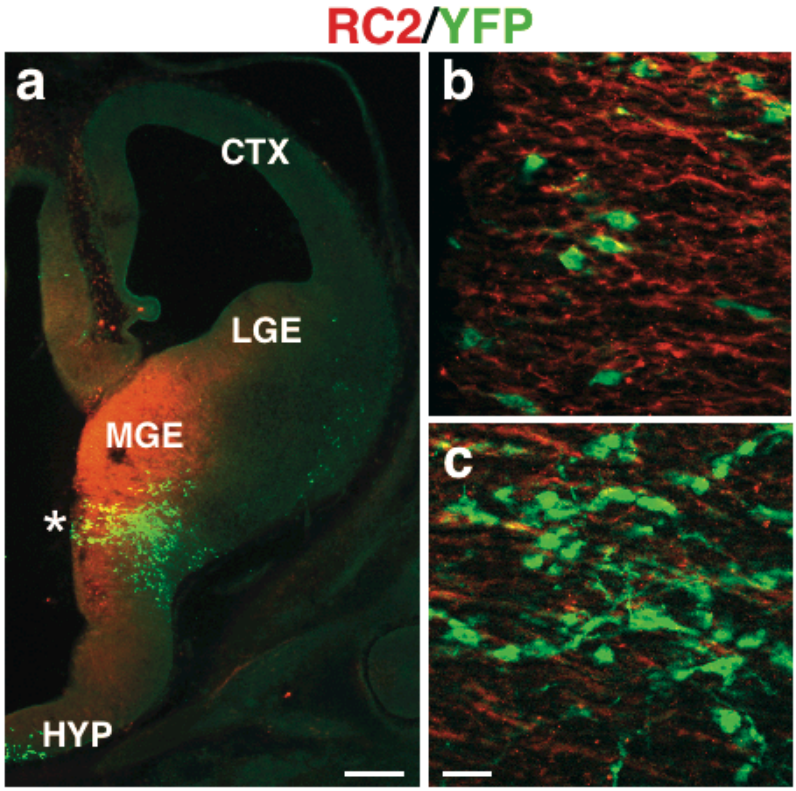
β-galactosidase staining of coronal sections from *Dbx1*$^{+/CreERT2}$*;R26RLacZ* brains at P21, which was treated with tamoxifen at E10.5, are shown at rostral and caudal levels throughout the extent of the hypothalamus. Many of *Dbx1*-derived cells contributed to the adult diencephalon including preoptic area (**a**) and multiple other hypothalamic nuclei (**b-d**). *Abbr*: 3v; 3rd ventricle, ac; anterior commissure, AHP; anterior hypothalamic area, posterior part, DM; dorsomedial hypothalamic nucleus, LA; lateroanterior hypothalamic nucleus, LPO; lateral preoptic nucleus, ME; median eminence, MeA; Medial amygdala nucleus, MPO; medial preoptic nucleus, PH; posterior hypothalamic nucleus, PLH; peduncular part of lateral hypothalamus, VMH; ventromedial hypothalamus nucleus Scale bar; 500 μm

Supplementary Fig 3



Extensive migration from the POA *in vitro*. Explants from the POA (**a**,**b**), septum (**c**) and PSB (**d**) were dissected from E12.5 embryos and cultured in matrigel for three DIV. Numerous migrating cells are observed from the POA explants (arrows, **a**, **b**; in each case asterisks mark the center of the explant). This pattern of migration is observed in most (17/21) POA explants. All septal explants (n=8) displayed little to no migration, with only a few cells occasionally migrating just a short distance from the explant (**c**, arrow). Typical migration away from the cortico-striatal angle in explants from the PSB is also shown (**d**, arrow). This pattern of migration is observed in the majority (14/16) of PSB explants.

Supplementary Figure 4



Radial glia mark the migratory route of *Dbx1*+ POA derived cells

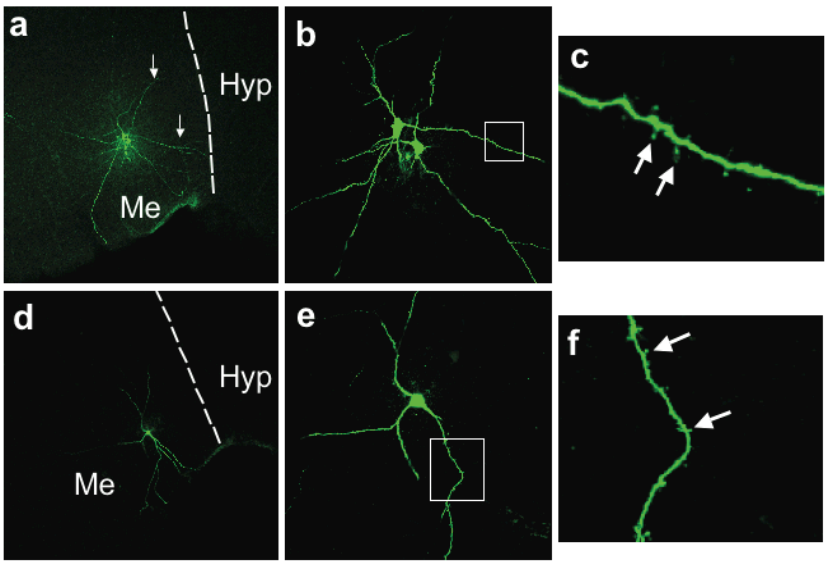
Immunohistochemistry for RC2 (red) and YFP (green) in coronal sections from E12.5 *Dbx1*$^{+/CreERT2}$;R26RYFP embryos treated with tamoxifen at E10.5 are shown (**a-c**). *Dbx1*-derived cells migrate through a region of RC2 expression (**a,** asterisk marks the POA). Higher power magnification the PAS is shown in **b** and **c** and reveals YFP+ cells (arrows) closely associated with RC2+ radial glia. Scale bar in (a); 250μm Scale bar in (c); 20 μm for (b,c)

Supplementary Table 1

Membrane resistance (MΩ)	491. 34±26.7 (n=35)
Membrane potential (mV)	-58. 21±1.1 (n=35)
Peak Amplitude (mV)	41.9 7±1.67 (n=35)
Maximum Frequency (HZ)	27.4±1.45 (n=35)
Accommodation Ratio	0.568±0.029 (n=35)
Sag (mV)	4.1±0.55 (n=27)
Half-width (ms)	1.59±0.039 (n=35)

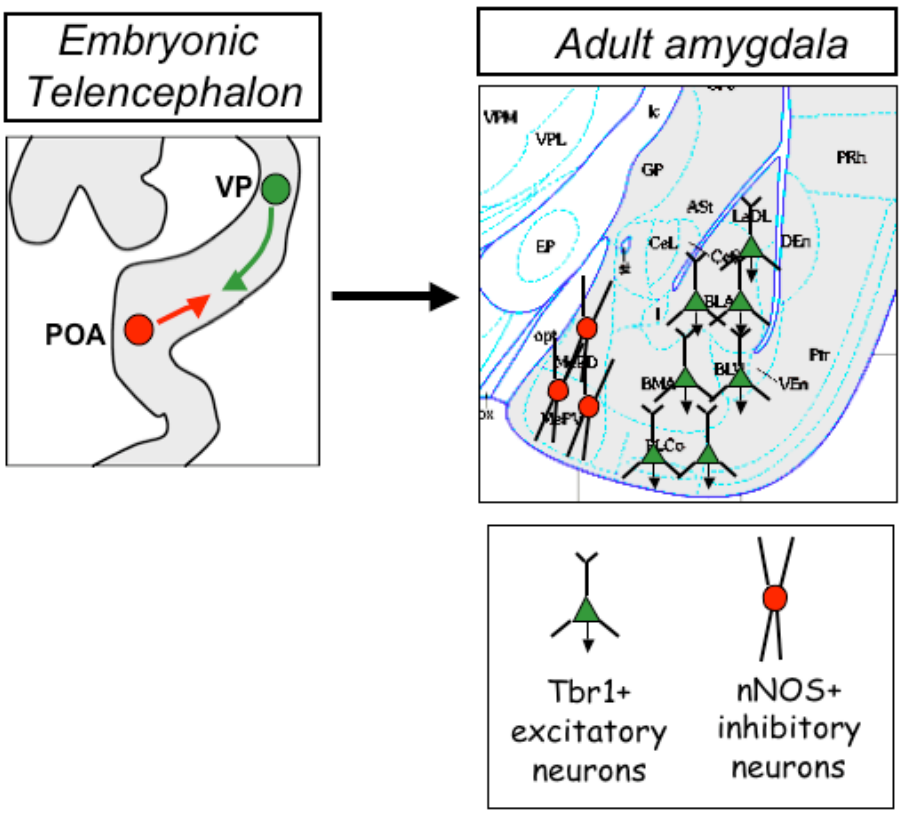
Intrinsic electrophysiological properties of medial amygdala *Dbx1*-derived neurons

Supplementary Figure 5



Morphologies of recombined medial amygdala neurons. Post physiological recordings, YFP+ medial amygdala nuclei cells were filled with biocytin. Typical morphologies of YFP+ neurons are shown in (**a-f**). Lower power magnification reveals cells with ovoid cell bodies and bipolar-oriented processes that extend over long distances directed in both dorsal and ventral directions that also appear to extend toward the hypothalamus (arrows, **a, d**). Higher magnification reveals that these long processes appear to be sparsely to moderately spiny (**b, e**). Blow up of boxed regions in **b, c** show the presence of individual spines on two separate processes (arrows, **c, f**)

Supplementary Figure 6



Schematic of *Dbx1*-derived POA and PSB contribution to the amygdala. Panel on the left depicts the ventral aspect of an approximately E11.5 embryonic telencephalon. Two separate *Dbx1+* progenitor pools are shown in the VP (green oval) and the POA (red oval), with arrows indicating their general routes of migration to the region of the developing amygdala along the lateral cortical stream (LCS) and POA-amygdala migrating stream (PAS), respectively. The panel on the right depicts the fate of these progenitor pools in the post natal amygdala, with the VP *Dbx1*-derived pool giving rise to a population of Tbr1+ excitatory neurons of the basolateral complex and cortical amygdala nuclei and the POA *Dbx1*-derived progenitor pool giving rise to a population of inhibitory nNOS+

neurons in the medial nucleus of the amygdala.