## **Supplemental Information**

**Structural Basis of Teneurin-Latrophilin** 

**Interaction in Repulsive Guidance** 

of Migrating Neurons

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Data collection					
Protein	chicken Ten2 and	chicken Ten2 and			
	murine Lphn2 Lec	murine Lphn1 Lec-Olf			
PDB code	6SKE	6SKA			
Space group	P1	P4 <sub>3</sub> 22			
Cell dimensions					
a, b, c (Å)	90.87 109.92 152.21	96.26 96.26 809.22			
α, β, γ (°)	90.19 93.99 111.93	90.0 90.0 90.0			
Resolution (Å)	84-3.6 (3.71-3.6)	202-3.86 (4.05-3.86)			
R <sub>meas</sub>	0.334 (212)	0.556 (3.46)			
l/σ(l)	3.47 (0.56)	6.19 (1.41)			
Highest resolution shell	4.49 - 4.33 (R <sub>meas</sub> of	4.31-4.22 (R <sub>meas</sub> of shell			
with $I/\sigma(I) > 2$	shell = 0.532)	= 1.87)			
CC <sub>1/2</sub>	0.97 (0.25)	0.99 (0.45)			
Completeness (%)	96 (96)	91.2 (36.2)			
Redundancy	1.8 (1.8)	12.8 (13.7)			
Refinement					
Resolution (Å)	84-3.6 (3.64-3.6)	52-3.86 (3.98-3.86)			
$R_{work}/R_{free^*}$	0.27/0.27 (0.28/0.28)	0.26/0.26 (0.25/0.28)			
No of atoms					
protein	29198	17313			
other	580	305			
solvent	210	-			
mean B-values (Å <sup>2</sup> )	99.4	134			
Ramachandran plot					
favoured (%)	96.5	95.21			
outliers (%)	0.24	0.64			
Bond length deviations (Å)	0.007	0.007			
Bond angle deviations (°)	0.97	0.95			

**Table S1 related to Figure 1.** Crystallographic statistics for deposited structures 6SKE and 6SKA.

Ligand - Analyte	Apparent K <sub>D</sub>	RU analyte immobilised	Bmax	R <sup>2</sup>
mLphn3 <sup>ecto</sup> - mTen2 <sup>ecto</sup>	490 nM	730	874	0.995
mLphn2 <sup>ecto</sup> - mTen2 <sup>ecto</sup>	100 nM	700	750	0.971
mLphn1 <sup>ecto</sup> - mTen2 <sup>ecto</sup>	35 nM	670	1014	0.865
mLphn3 <sup>ecto</sup> - gTen2 <sup>ecto</sup>	680 nM	730	1173	0.991
mLphn2 <sup>ecto</sup> - gTen2 <sup>ecto</sup>	60 nM	700	985	0.931
mLphn1 <sup>ecto</sup> - gTen2 <sup>ecto</sup>	40 nM	670	1160	0.872
mLphn1 <sup>Lec-Olf</sup> - mTen2 <sup>ecto</sup>	290 nM	220	473	0.991
mLphn1 <sup>Lec-Olf</sup> - gTen2 <sup>ecto</sup>	590 nM	220	631	0.999

Table S2 related to Figure 2. Apparent binding affinities calculated from SPR measurements using Latrophilin and Teneurin constructs are shown. Here we use the following nomenclature for brevity: gTen2 = chicken Ten2, mTen2= murine Ten2, mLphn = murine Lphn. The construct boundaries and mutant types are indicated in superscript.  $K_D$  and  $R_{max}$  values were obtained by nonlinear curve fitting of a 1:1 Langmuir interaction model (bound =  $R_{max}/(K_D + C)$ , where C is analyte concentration calculated as a monomer. Note that Teneurin protein is dimeric and so these values are indicative only. All Ten<sup>ecto</sup> proteins were injected at the same molar concentrations. As done previously (Brasch et al., 2018; Dionne et al., 2018), the comparison of the resulting raw binding curves (Fig. 2G-H and S2C-E) is a more reliable way to assess relative binding.