

Statistical summary of synaptic parameters characterized in *Hoxb8* mutant mice

a

Cortex	Age (mths)	Parameters analyzed	Synapse type, Genotype and 'n'								Statistical test
			Asymmetric				Symmetric				
			WT	<i>Hoxb8</i> mutant	P	Figure	WT	<i>Hoxb8</i> mutant	P	Figure	
8 months			Mean ± SEM, n	Mean ± SEM, n			Mean ± SEM, n	Mean ± SEM, n			One-way ANOVA Tukey's posthoc test
	Presynaptic diameter	444 ± 8.4 (n= 250)	493 ± 7.1 (n= 250)	<0.0001*	Sup 3j	513 ± 6.9 (n= 250)	538 ± 7.2 (n= 250)	0.0102*	Sup 3j		
	Postsynaptic diameter	375 ± 5.7 (n= 250)	418 ± 5.4 (n= 250)	0.00171	Sup 3l	510 ± 6.7 (n= 250)	499 ± 5.6 (n= 250)	0.1842	Sup 3l		
	Synaptic length	373 ± 5.8 (n= 250)	403 ± 5.2 (n= 250)	0.00012	1e	441 ± 5.3 (n= 250)	475 ± 5.5 (n= 250)	<0.0001*	1e		
	Active zone length	248 ± 3.3 (n= 250)	321 ± 3.7 (n= 250)	<0.0001*	Sup 2i	345 ± 4.2 (n= 250)	406 ± 4.9 (n= 250)	<0.0001*	Sup 2i		
	Active zone thickness	21 ± 0.17 (n= 250)	21 ± 0.17 (n= 250)	0.54393	Sup 2k	20.5 ± 0.15 (n= 250)	20.9 ± 0.15 (n= 250)	0.07734	Sup 2k		
	PSD length	295 ± 3.6 (n= 250)	344 ± 3.5 (n= 250)	<0.0001*	1o	340 ± 3.8 (n= 250)	389 ± 5.6 (n= 250)	<0.0001*	1o		
PSD thickness	65 ± 0.64 (n= 250)	51 ± 0.63 (n= 250)	<0.0001*	1q	58 ± 0.53 (n= 250)	56 ± 0.82 (n= 250)	0.014*	1q			

b

Striatum	Age (mths)	Parameters analyzed	Synapse type, Genotype and 'n'								Statistical test
			Asymmetric				Symmetric				
			WT	<i>Hoxb8</i> mutant	P	Figure	WT	<i>Hoxb8</i> mutant	P	Figure	
8 months			Mean ± SEM, n	Mean ± SEM, n			Mean ± SEM, n	Mean ± SEM, n			One-way ANOVA Tukey's posthoc test
	Presynaptic diameter	548 ± 9.8 (n= 250)	369 ± 8.0 (n= 250)	<0.0001*	Sup 3k	547 ± 7.9 (n= 250)	454 ± 7.7 (n= 250)	<0.0001*	Sup 3k		
	Postsynaptic diameter	425 ± 8.6 (n= 250)	324 ± 6.9 (n= 250)	<0.0001*	Sup 3m	495 ± 6.4 (n= 250)	449 ± 6.9 (n= 250)	<0.0001*	Sup 3m		
	Synapse length	483 ± 6.2 (n= 250)	433 ± 5.4 (n= 250)	<0.0001*	1f	491 ± 7 (n= 250)	437 ± 5.8 (n= 250)	<0.0001*	1f		
	Active zone length	323 ± 6.6 (n= 250)	228 ± 4.7 (n= 250)	<0.0001*	Sup 2j	398 ± 5.6 (n= 250)	352 ± 5.3 (n= 250)	<0.0001*	Sup 2j		
	Active zone thickness	21 ± 0.17 (n= 250)	21 ± 0.22 (n= 250)	<0.0001*	Sup 2l	20 ± 0.15 (n= 250)	21 ± 0.16 (n= 250)	<0.0001*	Sup 2l		
	PSD length	282 ± 4.1 (n= 250)	254 ± 4.1 (n= 250)	<0.0001*	1p	371 ± 4.2 (n= 250)	346 ± 4.2 (n= 250)	<0.0001*	1p		
PSD thickness	60 ± 0.6 (n= 250)	54 ± 0.9 (n= 250)	<0.0001*	1r	55 ± 0.6 (n= 250)	60 ± 1 (n= 250)	<0.0001*	1r			

c

Age (mths)	Parameters analyzed	Cortex		Striatum	
		Asymmetric	Symmetric	Asymmetric	Symmetric
		<i>Hoxb8</i> Vs WT	<i>Hoxb8</i> Vs WT	<i>Hoxb8</i> Vs WT	<i>Hoxb8</i> Vs WT
8 months	Presynaptic diameter	↗	↗	↘	↘
	Postsynaptic diameter	↗	↔	↘	↘
	Synapse length	↗	↗	↘	↘
	Active zone length	↗	↗	↘	↘
	Active zone thickness	↔	↔	↘	↗
	PSD length	↗	↗	↘	↘
	PSD thickness	↘	↘	↗	↗

c1

Age (mths)	Parameters analyzed	P values for Paired two-tailed t test			
		Cortex		Striatum	
		Asymmetric	Symmetric	Asymmetric	Symmetric
8 months		<i>Hoxb8</i> Vs WT	<i>Hoxb8</i> Vs WT	<i>Hoxb8</i> Vs WT	<i>Hoxb8</i> Vs WT
	Presynaptic diameter	<0.0001*	<0.0001*	<0.0001*	<0.0001*
	Postsynaptic diameter	<0.0001*	<0.0001*	0.1573	<0.0001*
	Synapse length	<0.0001*	<0.0001*	<0.0001*	<0.0001*
	Active zone length	<0.0001*	<0.0001*	<0.0001*	<0.0001*
	Active zone thickness	<0.0001*	0.0717*	<0.0001*	<0.0001*
	PSD length	<0.0001*	<0.0001*	<0.0001*	0.0002*
PSD thickness	<0.0001*	0.0078*	<0.0001*	<0.0001*	

d

Age (mths)	Parameters analyzed	Cortex		Cortex		Cortex		Cortex	
		<i>Hoxb8</i> WT	<i>Hoxb8</i> KO	<i>Hoxb8</i> WT	<i>Hoxb8</i> KO	<i>Hoxb8</i> WT	<i>Hoxb8</i> KO	<i>Hoxb8</i> WT	<i>Hoxb8</i> KO
		Mouse # 1	Mouse # 1	Mouse # 2	Mouse # 2	Mouse # 3	Mouse # 3	Mouse # 3	Mouse # 3
8 months	Total neurons (#)	11	12	13	15	11	14		
	Slices analyzed (#)	5	5	5	5	5	5		
	Total dendrites (#)	21	25	26	26	21	26		
	Mean ± SEM (Spines/10 μm)	5.74 ± 0.47	7.38 ± 0.52	7.97 ± 0.3	10.8 ± 1.0	7.54 ± 0.78	9.57 ± 0.49		
	P value (T-test)		0.0251		0.012		0.03		
	F probability (T-test)		0.4138		< 0.0001		0.09		
	P value (ANOVA)		0.0273		0.01		0.027		
F value (ANOVA)		5.207		7.141		5.196			

e

Age (mths)	Parameters analyzed	Striatum		Striatum		Striatum		Striatum	
		<i>Hoxb8</i> WT	<i>Hoxb8</i> KO	<i>Hoxb8</i> WT	<i>Hoxb8</i> KO	<i>Hoxb8</i> WT	<i>Hoxb8</i> KO	<i>Hoxb8</i> WT	<i>Hoxb8</i> KO
		Mouse # 1	Mouse # 1	Mouse # 2	Mouse # 2	Mouse # 3	Mouse # 3	Mouse # 3	Mouse # 3
8 months	Total neurons (#)	5	5	5	6	3	7		
	Slices analyzed (#)	3	4	3	4	3	4		
	Total dendrites (#)	15	15	16	16	13	12		
	Mean ± SEM (Spines/10 μm)	8.17 ± 0.52	6.09 ± 0.36	7.99 ± 0.68	7.29 ± 0.48	8.82 ± 0.62	6.92 ± 0.54		
	P value (T-test)		0.003		0.4062		0.032		
	F probability (T-test)		0.2076		0.1903		0.4931		
	P value (ANOVA)		0.0029		0.4054		0.0359		
F value (ANOVA)		10.65		0.711		4.9948			

f

Age (mths)	Parameters analyzed (Total)	Dendritic spine analysis	
		Cortex	Cortex
		<i>Hoxb8</i> WT	<i>Hoxb8</i> KO
8 months	Mice	3	3
	Slices	15	15
	Neurons	21	22
	Dendrites	68	77

Age (mths)	Parameters analyzed (Total)	Striatum	
		<i>Hoxb8</i> WT	<i>Hoxb8</i> KO
		8 months	Mice
Slices	10		12
Neurons	13		18
Dendrites	44		43