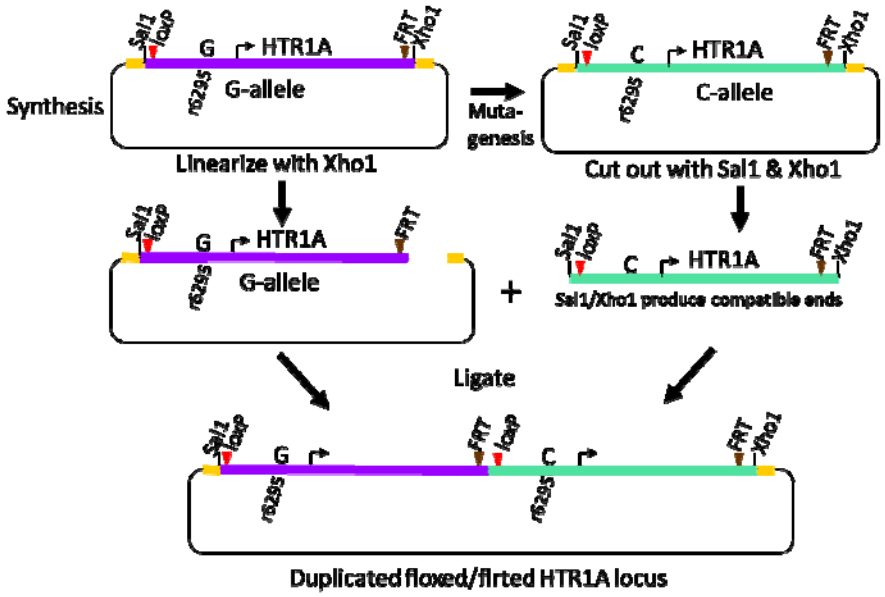
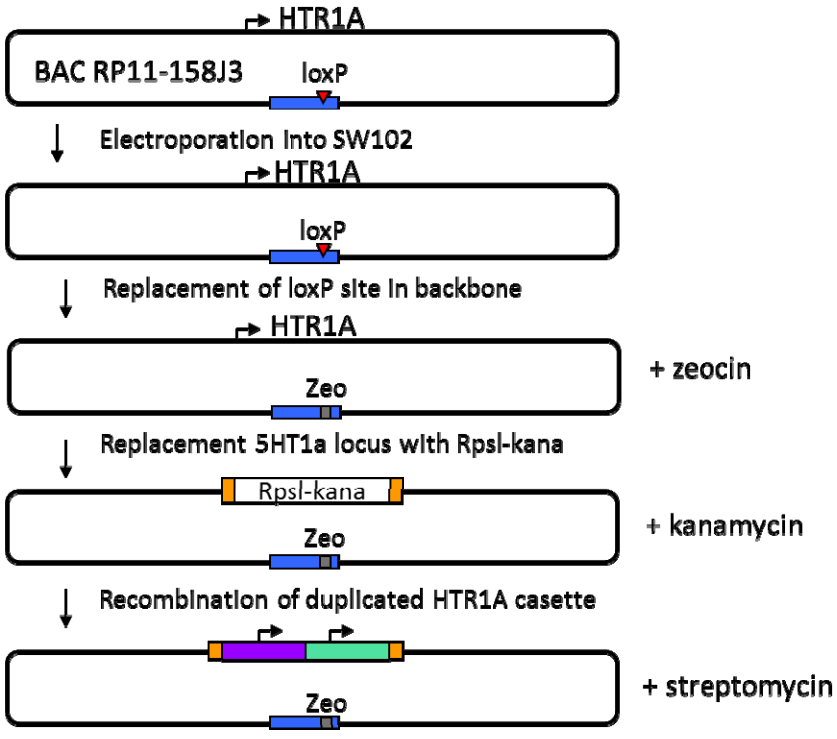
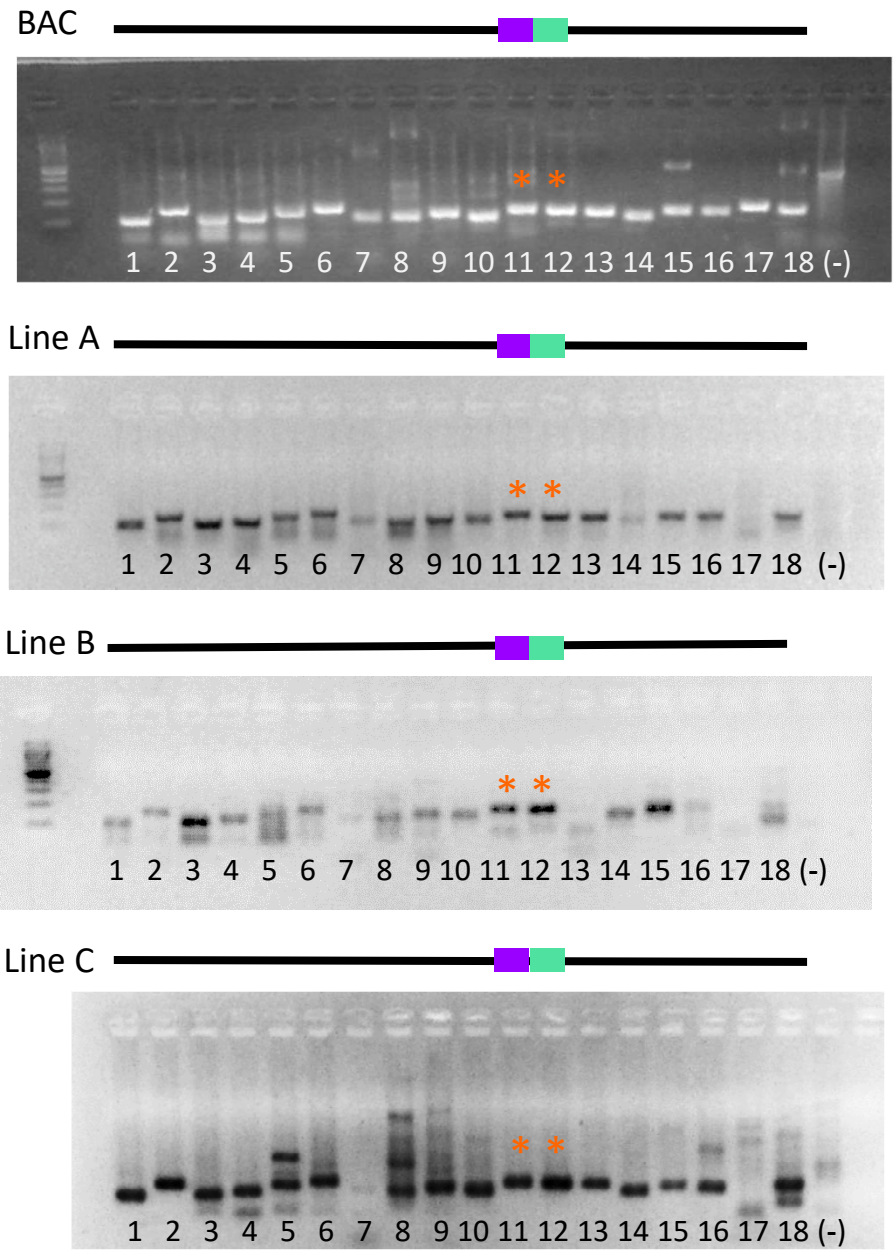
A. Generation of duplicated floxed/flrted HTR1A casette



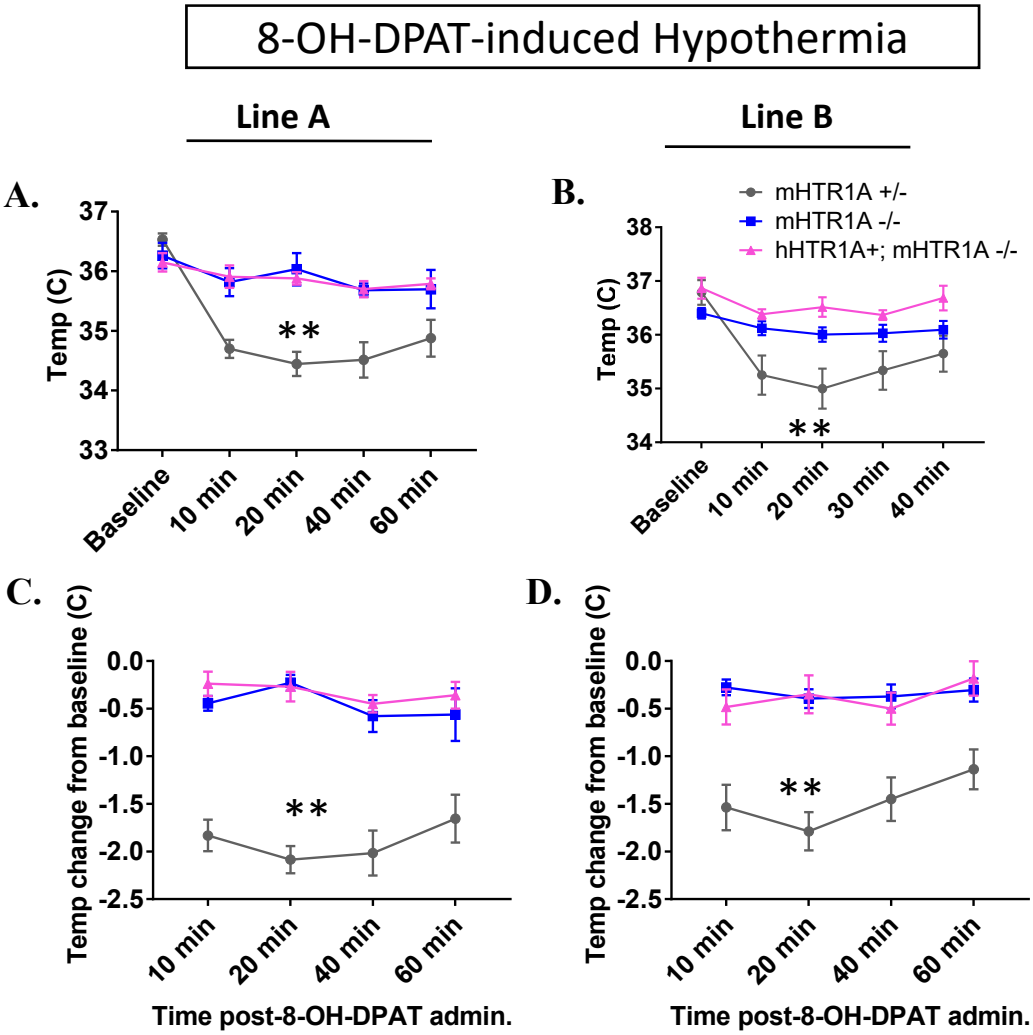
B. Generation of duplicated floxed/flrted HTR1A BAC



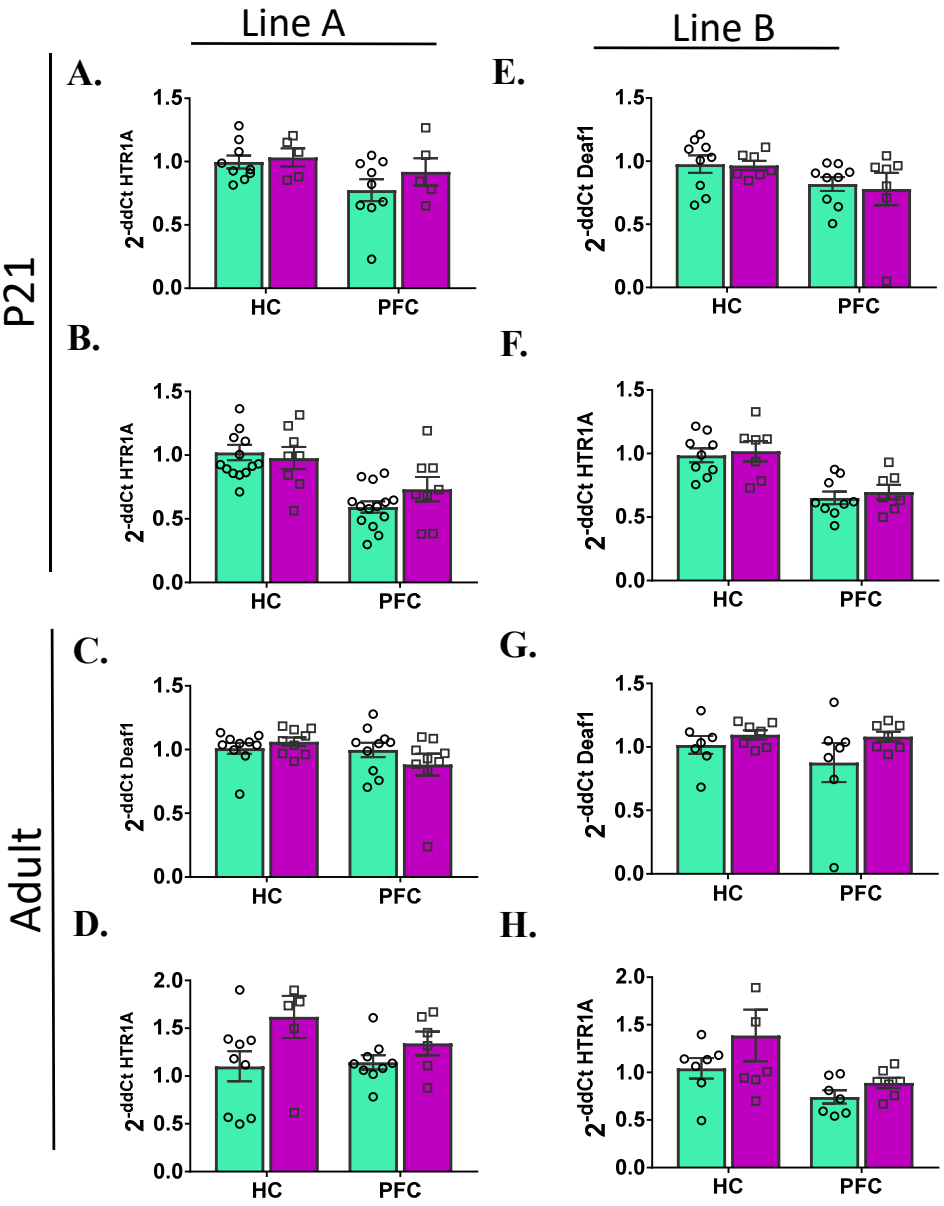
Supplemental Figure 1. Generation of duplicated floxed/flrted HTR1A BAC. A) Cloning strategy to generate targeting plasmid with duplicated HTR1A gene containing rs6295G and C-allele. Yellow regions = homology arms for targeting recombination in BAC. B) Modification of BAC to delete loxP site from backbone and replace HTR1A gene with duplicated locus from A. Purple = G-allele. Green = C-allele. Yellow = homology arms for targeting. Not to scale.



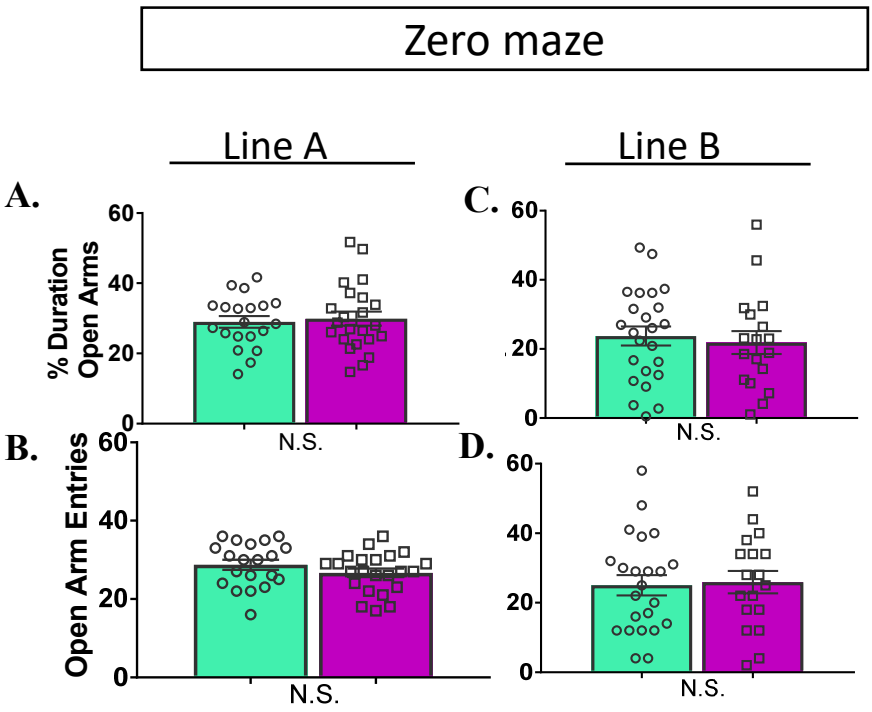
Supplemental Figure 2. PCR tiling to assess integrated BAC integrity. Each line was screened via PCR reactions designed to amplify small fragments every ~10kb across the BAC. Orange asterisks indicate amplicons that fell within the duplicated region containing rs6295 and hHTR1A, which is shown in purple (rs6295G) and green (rs6295C) in the diagram above. Red dashes indicate missing amplicons, suggesting a truncation of the 3' end of the BAC in Line B between primer pair 16 and 17. Primer locations are provided in supplementary table 2.



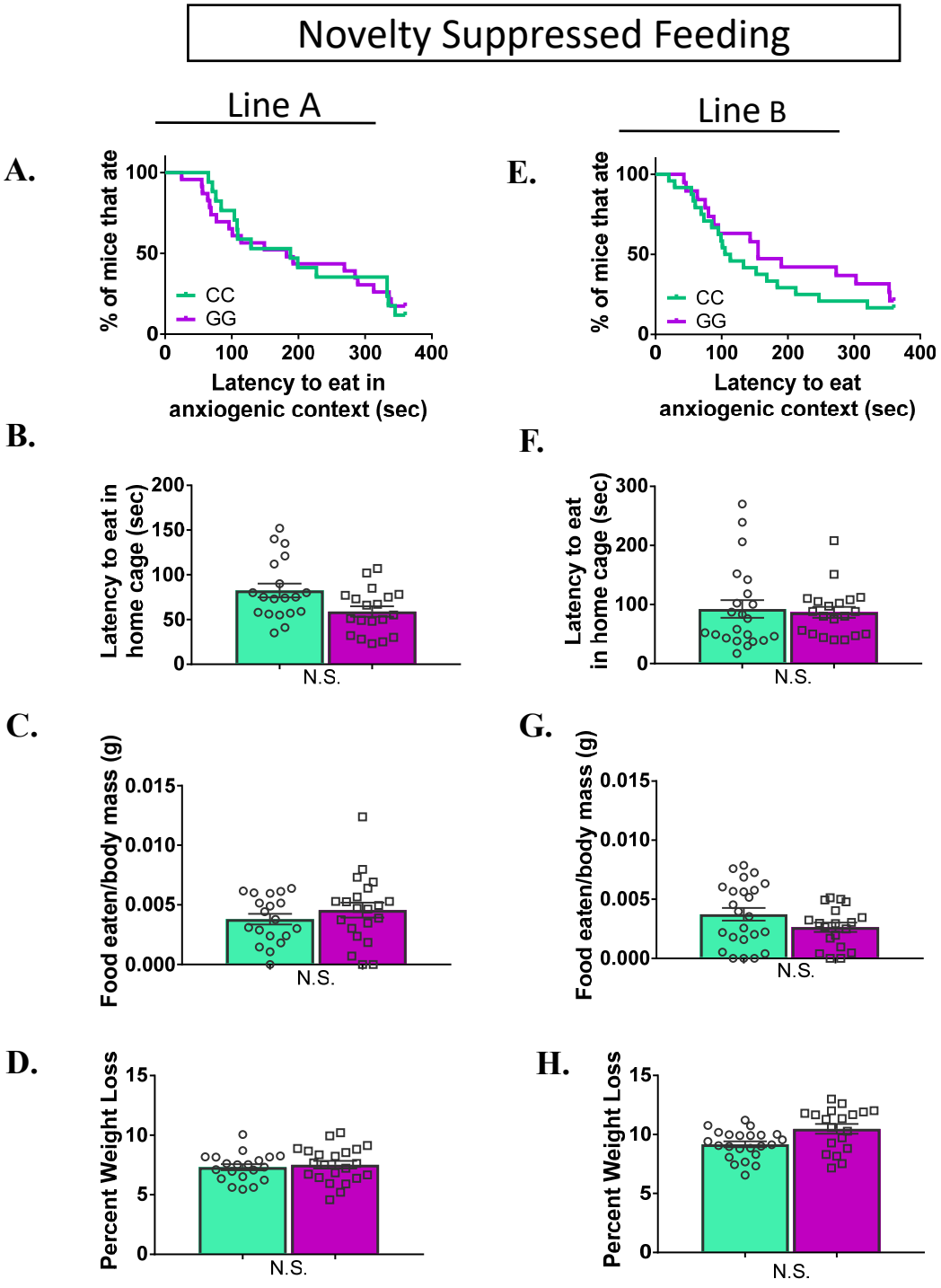
Supplemental Figure 3. Lack of 8-OH-DPAT induced hypothermia in hHTR1A mice. mHTR1A +/- mice exhibited normal hypothermic responses following administration of 8-OH-DPAT, which were not observed in mHTR1A - /- or hHTR1A; mHTR1A -/- mice, indicating a lack of functional 5-HT1A autoreceptor expression. In the Line, **(A)** Body temperature before and after administration of 8-OH DPAT, (**B**) change in temperature relative to baseline ($p < 0.0001$). Similar results were found in line B. **(C)** Body temperature before and after administration of 8-OH DPAT, (**D**) change in temperature relative to baseline ($p < 0.0001$). Values represent mean ±SEM.



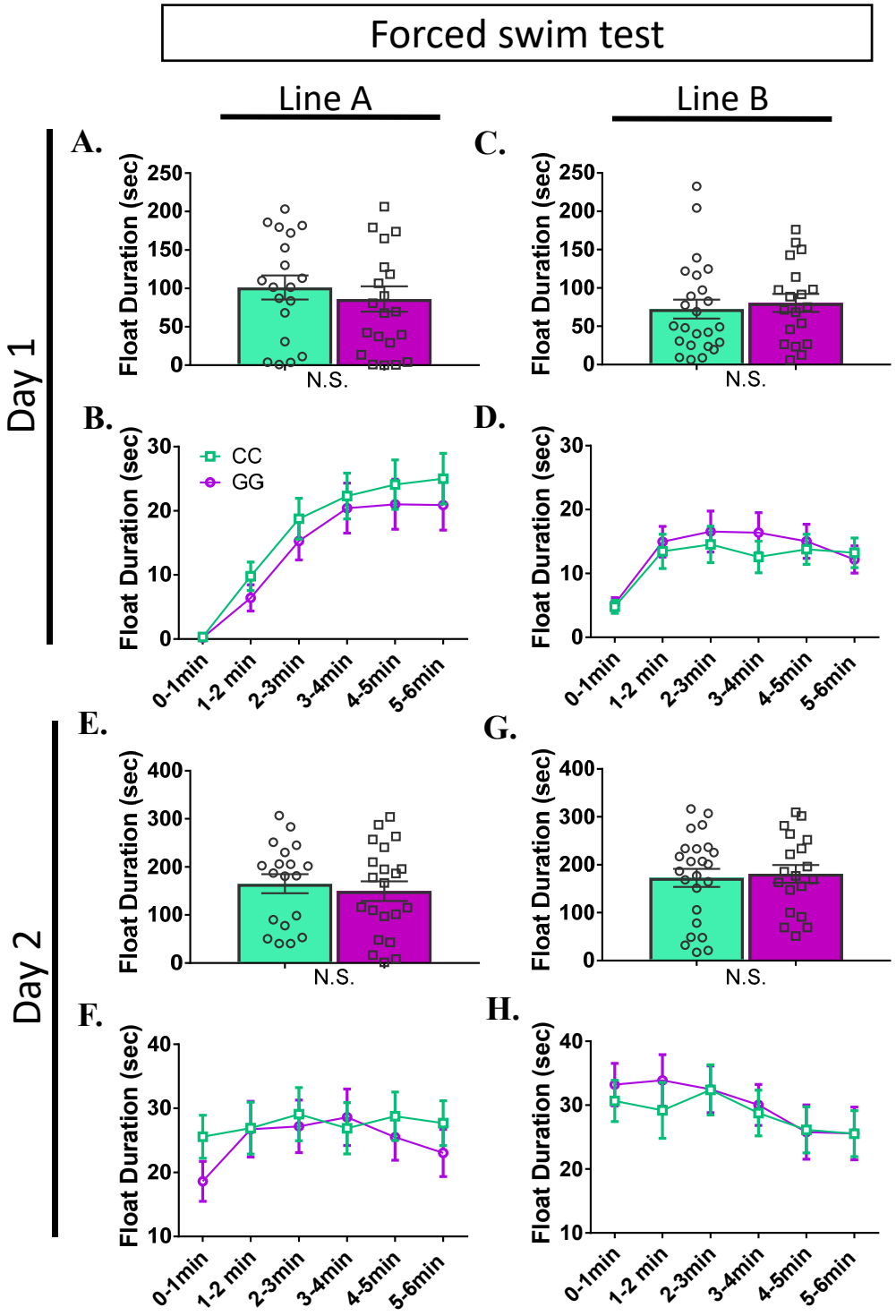
Supplementary Figure 4. No genotype-dependent differences in mDeaf1 or hHTR1A mRNA expression. qRT-PCR analysis of mouse Deaf1 and human HTR1A expression in microdissected prefrontal cortex and hippocampus did not reveal differences in mDeaf1 or hHtr1A mRNA expression in either line examined. Group sizes and statistical analyses available in supplemental table 4. Bar graphs are mean +/- SEM.



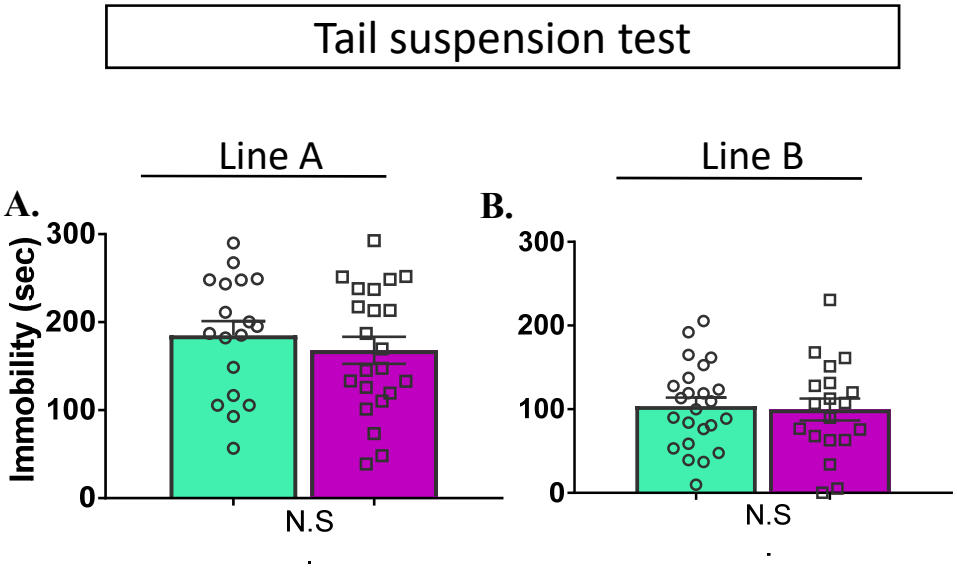
Supplemental Figure 5. No differences between genotypes in zero maze performance. In line A, there was no difference in **(A)** percent time spent in the open arms or (**B**) open arm entries (CC=20 GG=23; p=0.215) between genotypes. In line B, there were no differences in **(C)** percent time in open arms or **(D)** open arm entries. Values represent mean ±SEM.



Supplemental Figure 6. No differences between genotypes in novelty suppressed feeding. In line A, there was no difference between genotypes in **(A)** latency to eat in the anxiogenic context, (**B**) latency to each in the homecage, **(C)** grams of food eaten by body mass or **(D)** the percent weight loss In line B, there was no difference in **(E)** latency to eat in the anxiogenic context, **(F)** latency to eat in the home cage, **(G)** the percent food eaten by body mass or (**H**) weight change. Values represent mean ±SEM.



Supplemental Figure 7. No difference in depressive phenotypes in forced swim test. Forced swim test (FST) was run across two days. **(A, E, C, G)** There was no difference in Line A total float duration on either day. **(B, D)** There was a significant effect of time on float duration on day 1 ($p < 0.0001$). On day one and day two of FST, in the 8000 lineValues represent mean ±SEM.



Supplemental Figure 8. No genotype differences in immobility in tail suspension test. Values represent mean ±SEM.

Table 1. Transgene transmission from founders				
Line	Founder Sex	BAC+ offspring	Total offspring	Percent transmission
Line A	M	9	45	20%
Line B	F	22	37	59.40%
Line C	M	7	47	14.90%

Table 2. Oligos for checking BAC integrity						
Amplicon	Forward_primer	Reverse_primer	Forward_tm	Reverse_tm	Product_size	Location
1	TGATCGGGCAATCATTTCTG	AAACCATCCAGCGATGTCAA	61.926	61.449	104	2795
2	TGCCTTTGAAGTTGGCTCAG	AAATGAGGCAGAGCGGAAAC	61.481	61.646	150	15019
3	CTCCAATGCTGTCTGGCAAC	GCATATTCACCACCCAGACG	61.81	61.329	107	25108
4	GGGGATAAGTTCAGGGGTGA	GGCCGCCTATAACCAATAGC	61.076	60.788	115	33993
5	TGAGAAGCCCCATTTCTTCC	GGTTCTGGCTTGGGATCACT	61.46	61.429	138	47325
6	TGTGTTATTGGGAGGCAGGA	ACGCCTCAGCAATGTCTCTG	61.436	61.566	150	57091
7	TCAAGATGCCTGAGCTTTCG	TTGGGGGATGCGATTAAACT	61.593	61.542	107	61965
8	CAGAGAGGCCAGTGTGTGCT	TCGTATCCTGGGGCTTCAAT	61.649	61.724	109	79283
9	ATTGAACTGCAAGGCAGCAG	AGGCCTCGCAGAGCTGTAGT	61.513	61.645	121	84637
10	CCTGTGGTCCCCAGCTACTT	TCACCCAGGCTAGAGTGCAG	61.461	61.552	108	94964
11	CTCCTCGGAGATACCCTTCG	AGAAAAAGCAGCGCGAAGAT	61.103	61.502	138	103443
12	GGTAACCTGCGACCTGTTCA	GGCGTCCTCTTGTTCACGTA	61.096	61.236	132	110058
13	TGGCATCTCGTGAACCTCA	TTCTTTCCCACAGGGTAGGC	61.431	61.37	123	125123
14	TGCCAGTTGCAGTTTTTGGT	CAGCTGGTCTGACCCCCTAT	61.636	61.436	104	130630
15	ACATGGTGGCCACAAGAGAA	TGAATCATGGTGGCCAGTCT	61.527	61.501	125	146638
16	GAGGATATGCCCCTCCCATA	TTATTCTGCCACCCTCACCA	61.006	61.436	123	150687
17	TTGGATGCCTCACAAGCAA	CCATCAGGCGGATTACACTG	61.379	61.455	148	160459
18	CCATTGTGGGCAGAGAACAC	GAGGTTGCAGTGAGCCAAGA	61.527	61.546	133	171189

Table 3. qPCR probes and efficiencies		
Target Gene	**Primer Sequences**	**Probe**
mGAPDH	Thermofisher taqman Mm99999915_g1, cat # 4331182	
mDeaf1	5'-CAACATCAGTGGCAAACGC-3'	5'-56FAM/TGTTCAGGC/ZEN/ TGTCACCGATCTGC/3IABkFQ/-3'
	5'-CTCCACAATGCTCCCATCTG-3'	
hHTR1A	5'-TGTCGCTTCTCACAAACTCTC-3'	5'-56FAM/CCCTCCCCA/ZEN/ CTTCCTGCTCC/3IABkFQ/-3'
	5'-CTTCTCCTCTTTCTCTCTGCTC-3'	

Target Gene	**R^2**	**Efficiency %**
mGAPDH	0.996	90.034
mDeaf1	0.993	93.533
hHTR1A	0.989	93.616