Supplemental Figure and Table Legends

Supplemental File 1. Animals used in the current study. Number of animals and their genders from each BXD strain are listed for each experiment. Highlighted BXD strains represent lines that both 5-HT traits and midbrain transcriptome data were obtained.

Supplemental Table 1. Analysis of heritability of male blood, midbrain and diencephalon 5-HT traits. Numbers in parentheses designate the number of strains used in calculations. *P* values are calculated using one-way ANOVA F-test. Adjusted heritabilities (h^2) are calculated using Hegmann and Possidente's method.

Supplemental Figure 1. Gene expression networks associated with male midbrain 5-HT traits. Genes (top 25) with highest correlations with (A) 5-HT, (B) 5-HIAA, or (C) 5-HT Turnover (TO, 5-HIAA/5-HT) are graphed separately based on the direction of correlation (green: positive; red: negative). Gene node sizes and colors represent the significance of correlation. Gene pairs with correlation coefficients (Spearman rho) above 0.5 are connected by edges (green: positive; red: negative).

Supplemental Figure 2. Gene expression networks associated with determinants of midbrain 5-HT synthesis, metabolism and inactivation. Genes (top 25) with highest correlations with (A) *Gch1*, (B) *Maoa*, or (C) *Slc18a2* are graphed separately based on the direction of correlation (green: positive; red: negative). Gene node sizes and colors represent the significance of correlation. Gene pairs with correlation coefficients (Spearman rho) above 0.5 are connected by edges (green: positive; red: negative).

Supplemental Table 2. Analysis of gender effects in female blood, midbrain and diencephalon serotonergic traits. Numbers in parentheses designate the number of strains used in calculations. *P* values are calculated using one-way ANOVA F-test. Adjusted heritabilities (h^2) are calculated using Hegmann and Possidente's method.

Supplemental File 2. Archived BXD phenotypes that have significant correlations with measured serotonergic traits separated by genders ($P < 0.05$, Spearman's rank test).

Supplemental File 3. Archived BXD phenotypes that have significant correlations with midbrain 5-HT levels (gender pooled, $P < 0.05$, Spearman's rank test).

Supplemental File 4. Archived BXD phenotypes that have significant correlations with blood 5-HT levels (gender pooled, $P < 0.05$, Spearman's rank test).

Supplemental File 5. Genes located under the quantitative loci identified by the interval mapping of midbrain 5-HT levels. Confidence interval for each QTL peak was determined by 1.5 LOD dropoff method.

Supplemental File 6. Genes located under the quantitative loci identified by the interval mapping of blood 5-HT levels. Confidence interval for each QTL peak was determined by 1.5 LOD dropoff method.

Supplemental File 7. Lists of genes whose midbrain expression significantly correlated with midbrain 5-HT traits ($P < 0.05$, Spearman's rank test).

Supplemental File 8. Lists of overlapping genes whose expression correlated with midbrain 5-HT traits and 1) exhibit similar anatomical expression pattern defined by *Tph2* and *Slc6a4* using NeuroBlast or 2) demonstrated enrichment in E12.5 developing 5-HT neurons. Original gene lists are also included as separate sheets.

Supplemental File 9. Lists of genes whose midbrain expression significantly correlated with diencephalon 5-HT traits ($P < 0.05$, Spearman's rank test).

Supplemental File 10. List of genes identified by interval mapping of midbrain 5-HT levels filtered by their expression correlation with midbrain 5-HT traits. Each gene’s expression in the raphe nuclei was scored by visual inspection of *in situ* data set from Allen Brain Atlas (N/A: probe not available or signal not strong enough).