Supplementary Material

Supplementary Methods

A. Network concepts

Connectivity (k) is a measure of connectedness of a given gene, either in the context of its module (kIN) or the entire network (kTotal). Connectivity is defined as: $ki = \sum_{j=1}^{N} aij$, where i and j are genes, N is all the genes in the module or network, and a is the adjacency between genes i and j. In Supplementary Table 1 we present both kIN and kTotal for a given gene in Area X Juvenile (kIN.x and kTotal.x), VSP Juvenile (kIN.v and kTotal.v) and Area X adult networks (kIN.x.adult and kTotal.x.adult).

Gene significance (gs) is the Pearson correlation between a gene's expression profile and a given behavioral metric. We present gene significance and related q values for number of motifs sung (n.motifs), percentage of tutor learning (tutor.pct.learning), NS-UD entropy effect (ns.ud.entropy.effect; our VI paradigm metric), and motif identity (motif.identity) in both juvenile Area X (e.g., gs.n.motifs.x and q.gs.n.motifs.x), juvenile VSP (e.g., gs.n.motifs.v and q.gs.n.motifs.v) and adult Area X networks (e.g., gs.n.motifs.adult and q.gs.n.motifs.adult)

B. Hypergeometric Overlap Tests

Hypergeometric overlap tests in Supp. Fig. 1 were carried out using the Gene Overlap Package in R. Juvenile Area X modules were defined to include any gene with a module membership score significant for a module, as defined by the false discovery rate corrected p-value q. This allowed genes with multiple significant module membership scores to be included in all relevant modules. 'Convergent Striatum' and 'Convergent Area X' included all genes in Pfenning et al. 2014 with significant gene expression specializations (increased or decreased) relative to average brain expression that were directionally convergent between songbird and human striatum or songbird Area X and a human region in the putamen activated by speech, as detected by fMRI. 'Intelligence All' included all genes with SNPs in Savage et al. 2018 implicated by positional, eQTL, or chromatin interaction mapping associated with intelligence in a meta-analysis of 269,867 independent individuals. 'Intelligence Exons' included all intelligence-related genes with SNPs in exons (leading to protein coding changes) in Savage et al. 2018. Background was 13,781 genes, the largest number of songbird brain expressed genes detected by RNAseq in the Burkett et al. 2018 dataset. Statistically significant gene overlap is calculated using Fisher's Exact Test.

A False Discovery Rate correction with a cut off of 0.2 was performed, with the majority of the modules maintaining significance. As a control a random sample of brain-expressed chicken genes equivalent to the number of genes in each juvenile Area X module was run against these same datasets. No significant overlap was found, indicating that enrichment was not an artifact from using brain-expressed or avian genes.

C. Chi-Squared Test

The Chi-Squared Test was used to test for correlations between directional changes in juvenile Area X gene expression associated with entropy variability modulation (VI) described in Burkett et al. 2018 and directional changes in intelligence gene eQTLs described in Savage et al. 2018. The total number of eQTLs that went up or down was roughly equivalent (324 up and 360 down or 47.4% up and 52.6% down). In the songbirds there were greater bias within the dataset, with 1,520 genes correlated to greater variability after two hours (VI -, 65.3%) and 807 correlated to greater stereotypy after two hours (VI +, 34.7%). A total number of 69 eQTLs were significant for variability modulation. We predicted that if there was no relationship between the two data sets then the eQTLs should be randomly distributed, with half of the VI - genes going up and half going down in the human eQTLs (33% up and 33% down) and half the VI + genes going up and half going down (17% up and 17% down). The VI + genes were very close to the predicted random distribution (15.94% up and 18.84% down). However, there was a bias in the VI - genes (15.94% up and 49.28% down), which drove the significant Chi-Square score of 11.81 and two-tailed p-value of 0.008. As a control, we compared the distribution of a random sample of an equivalent number of Area X genes not correlated to vocal variability and found no bias in the distribution.

Table captions

Supplementary Table 1. Table displays behaviorally relevant genes that show transcriptional convergence between songbird Area X (Burkett et al, 2018) and human putamen (Pfenning et al, 2014).

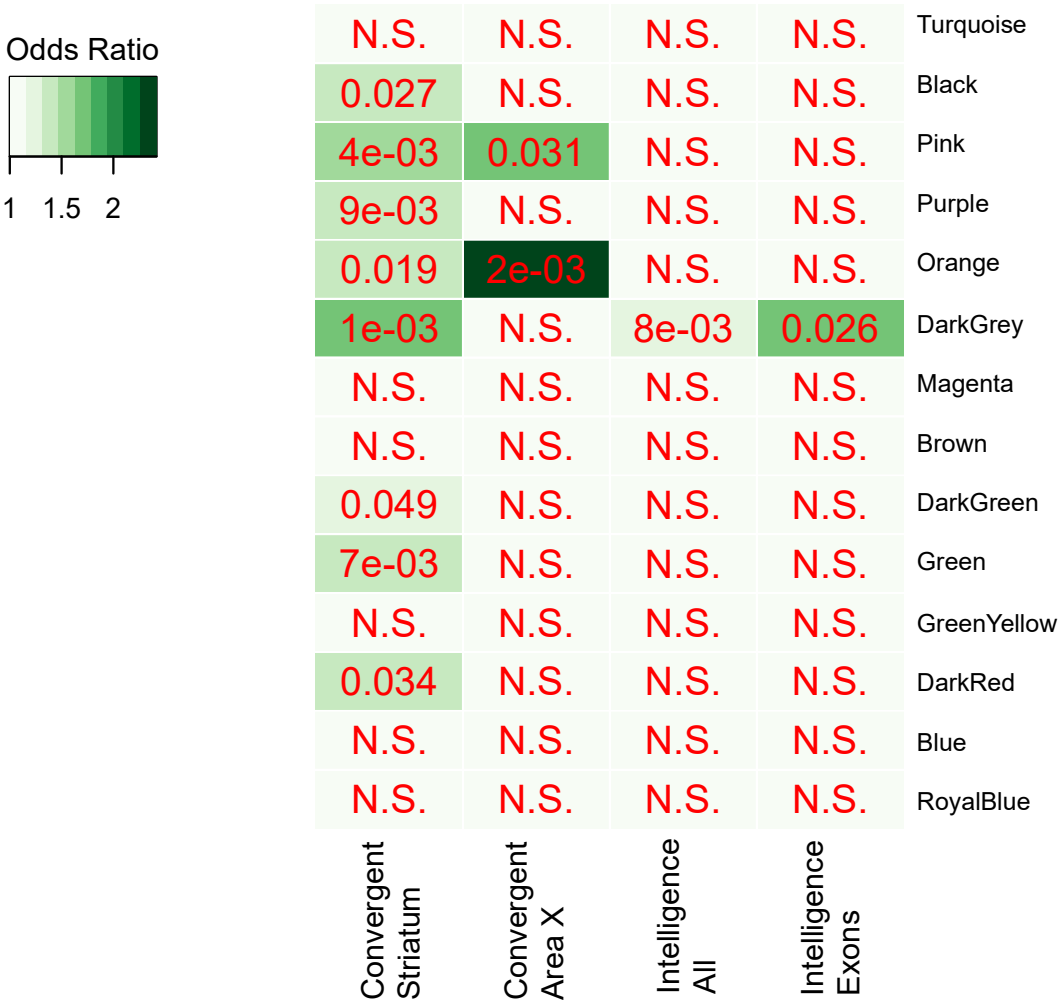
Supplementary Table 2. Table of non-coding human intelligence-related SNP genes detected in juvenile Area X modules. Variants associated with changes in gene expression are shown under 'eQTL Direction.' Primary juvenile Area X module assignment is listed under "module.x". Gene significance score for the variability induction paradigm is listed under "gs.ns.ud.entropy.effect" (column heading preserved from original publication in Burkett et al. 2018). FDR corrected p value q is listed under "q.gs.ns.ud.entropy.effect".

Supplementary Table 3. Table of protein-coding human intelligence-related SNPs detected in juvenile Area X modules. Primary juvenile Area X module assignment is listed under "module.x". Gene significance score for the variability induction paradigm is listed under "gs.ns.ud.entropy.effect" (column heading preserved from original publication in Burkett et al. 2018). FDR corrected p value q is listed under "q.gs.ns.ud.entropy.effect".

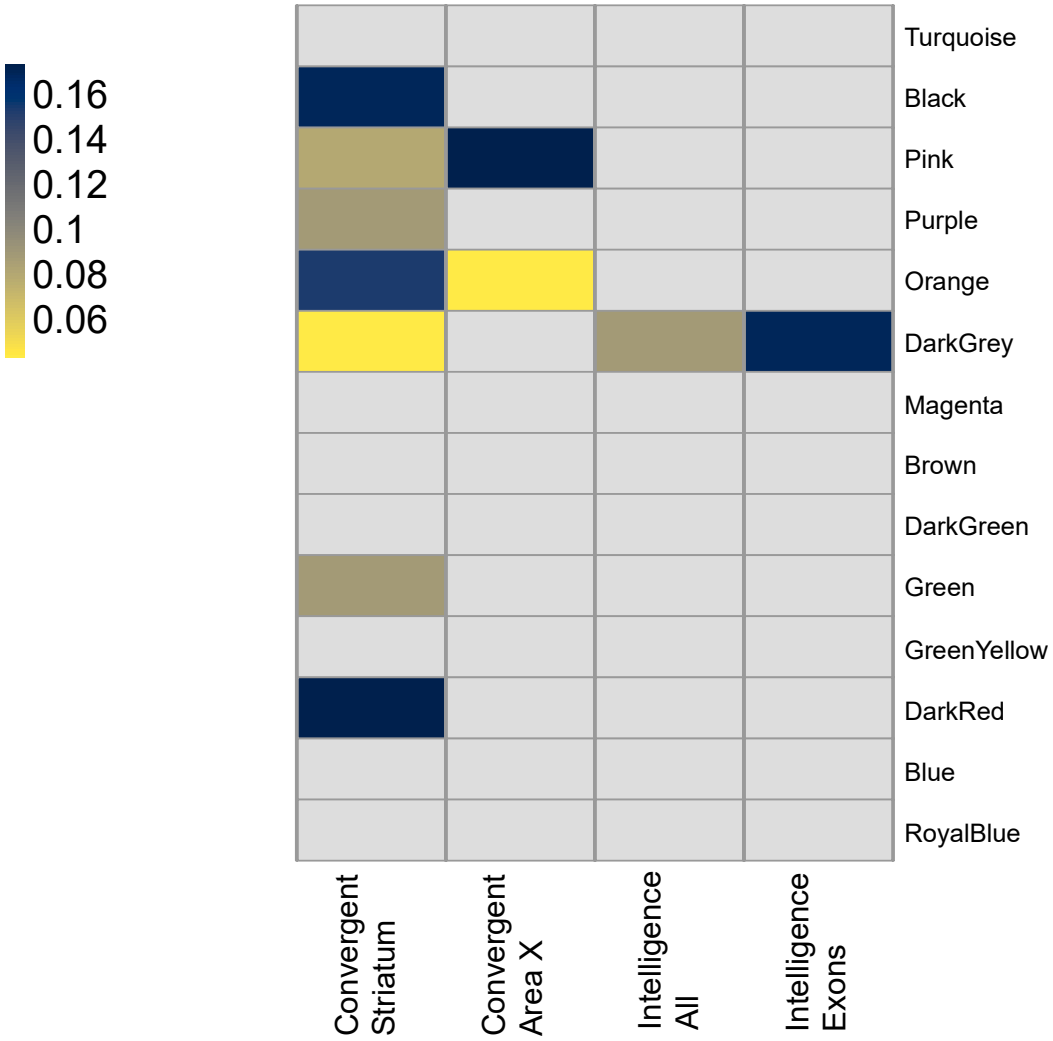
Supplementary Figures and Captions

Supplementary Figure 1. A) Heatmap showing hypergeometric overlap between gene sets described in the paper and juvenile Area X modules from Burkett et al. 2018 listed by color (R Gene Overlap package). 'Convergent Striatum' and 'Convergent Area X' include all genes in Pfenning et al. 2014 with significant gene expression specializations relative to average brain expression that were directionally convergent between songbird and human striatum or songbird Area X and a human region in the putamen activated by speech, as detected by fMRI. 'Intelligence All' includes all genes with SNPs in Savage et al. 2018 implicated by positional, eQTL, or chromatin interaction mapping associated with intelligence. 'Intelligence Exons' included all intelligence-related genes with SNPs in exons (leading to protein coding changes) in Savage et al. 2018. Statistical significance calculated using Fisher's Exact Test. B) Same data as in 2A but after a False Discovery Rate (FDR) correction with a cut off of 0.2. Significant modules (FDR-corrected p-value q<0.2) are indicated by color, with a key to the left.

A.



B.


0.16
0.14
0.12
0.1
0.08
0.06
Turquoise
Black
Pink
Purple
Orange
DarkGrey
Magenta
Brown
DarkGreen
Green
GreenYellow
DarkRed
Blue
RoyalBlue
Convergent Striatum
Convergent Area X
Intelligence All
Intelligence Exons