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Article

From GWAS to Mechanism: Synaptic Pruning Emerges as a Key Polygenic Driver of Cognitive Ability

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Abstract

Human intelligence is strongly heritable, but the genes that fine-tune the brain's wiring are still being mapped. We re-examined the large IQ genome-wide association study by Savage et al. (2018; N = 269,867) with three complementary tools—partitioned SNP-heritability, MAGMA gene tests, and a transcriptome-wide association study (TWAS)—to ask whether synaptic pruning makes an independent contribution beyond classic glutamatergic signalling. Seven pre-registered gene sets were contrasted: two glutamate lists, two pruning lists, two negative-control lists (monoamine and housekeeping genes), and a "pruning-only" set that removed every glutamatergic gene. Heritability was significantly enriched in both glutamate and pruning sets, but the pruning-only panel still carried a clear signal (1.10-fold, $P \approx 5 \times 10^{-15}$), showing that pruning effects are not simply spill-over from glutamate pathways. MAGMA supported this pattern, highlighting SEMA3F, RHOA, MAP1LC3B and TCF4 after Bonferroni correction. TWAS added tissue context: pruning genes showed the strongest over-representation (core set 1.38-fold, $P \approx 2 \times 10^{-5}$), with RHOA down-regulated in caudate and SEMA3F up-regulated in anterior cingulate. Together, the results inspire a "Calibrated Pruning Framework." We propose that common variants adjust the timing of a multistep pruning cascade—TCF4 keeps critical periods open a little longer, SEMA3F–RHOA steers branch retraction, MAP1LC3B clears debris, and HLA tags mark synapses for removal—thereby fine-tuning network efficiency and, ultimately, cognitive ability. Limits of the work include the European bias of the base GWAS and reliance on adult-brain expression panels; future longitudinal imaging and multi-ancestry studies will be needed to test the model's predictions.

Keywords: intelligence; IQ; GWAS; TWAS; MAGMA; partitioned heritability; pruning; plasticity

Introduction

Intelligence captures common variation in reasoning, memory, and problem solving. Decades of twin and family research show that these differences are strongly genetic, with heritability climbing from roughly 40 % in childhood to upward of 80 % in later life [1]. Until recently, however, the field lacked the statistical power to locate the many DNA variants behind this observation. Large genome-wide association studies (GWAS) have changed that landscape. A landmark analysis of educational attainment in more than one million people mapped over 1,200 significant single-nucleotide polymorphisms (SNPs) and produced polygenic scores explaining 11–13 % of variance in years of schooling [2]. When attention shifted to direct measures of intelligence, a meta-analysis of 269,867 participants uncovered 190 new loci, many active in brain tissues that guide neural development and synaptic signalling [3]. The emerging picture is one of extreme polygenicity: thousands of small-effect variants combine to influence cognitive performance.

Patterns in these data repeatedly point to the synapse. Gene sets tied to axon guidance, vesicle release, and postsynaptic scaffolding are over-represented, supporting models in which efficient transmission among neurons underlies higher cognitive scores [4]. Glutamatergic pathways are especially prominent, implicating receptor subunits such as GRIN2A and GRIA4 that are known to

affect learning and memory [5]. Yet synaptic strength is only one side of circuit refinement. During development, surplus connections are trimmed away through microglia-driven pruning. Molecular players in this process—most famously the complement cascade—have been linked to psychiatric conditions marked by altered cognition [6]. Whether variation in pruning genes also shapes normal differences in intelligence, distinct from glutamatergic effects, has not been tested systematically.

The present work tackles that question. Using summary statistics from the [3] GWAS of intelligence, we evaluate seven curated gene sets: canonical glutamatergic pathways, established pruning pathways, expanded versions of each, and a "pruning-only" set that removes all genes shared with glutamatergic lists. We apply partitioned heritability, MAGMA gene-based testing, and transcriptome-wide association analysis (TWAS) to gauge their contributions. The findings form the basis of a "Calibrated Pruning Framework," which proposes that genetic tuning of pruning cascades fine-tunes cognitive networks by eliminating connections at developmentally appropriate times.

Methods

MAGMA Gene- and Gene-Set Analysis

All gene-based statistics were generated with MAGMA v1.10 [7]. Single-nucleotide polymorphisms (SNPs) were assigned to genes according to the NCBI Build 37.3 coordinates, extending 35 kb upstream and 10 kb downstream so that common promoter and 3'-UTR elements were captured. Public summary statistics from the intelligence meta-GWAS [3; total $n = 269,867$] served as input; after the standard MAGMA quality-control steps the effective sample size was 225,955. Linkage disequilibrium (LD) structure was estimated with the European subset of the 1000 Genomes Phase 3 reference panel.

Seven biologically motivated gene sets were contrasted against the genomic background in competitive mode. For each set MAGMA converts gene p-values to Z-scores and then applies a one-sided t-test that compares the mean Z for the set with the genome-wide mean. Multiple testing across the seven sets was controlled both by Bonferroni correction ($\alpha = 0.0071$) and by the Benjamini-Hochberg false-discovery rate (FDR). Gene-level significance followed the conventional Bonferroni threshold for 18,370 tests ($p < 2.72 \times 10^{-6}$).

To tease apart pruning-related effects from those linked to glutamatergic neurotransmission, we removed 37 overlapping genes from the broad pruning list, producing a pruning-specific collection (Set G). This left the glutamatergic list (Set B) unchanged while ensuring that pruning results could not be driven by shared loci.

Annotation Enrichment and Partitioned-Heritability Analysis

Summary statistics from the large intelligence meta-GWAS [3] were formatted with the LD-score regression "munge" utility. Variants with $\text{INFO} < 0.90$ or minor-allele frequency (MAF) < 0.01 were removed, leaving 3,603,022 high-quality SNPs.

We looked at seven gene sets. Four were biological hypotheses: Set A (23 genes) was a small candidate list of glutamatergic receptor and plasticity genes; Set B (130 genes) was a larger panel of glutamatergic pathway genes; Set C (38 genes) was a core synaptic pruning list; and Set D (262 genes) was an expanded pruning-related collection. Set E (101 genes) and Set F (182 genes) were two negative-control sets that included housekeeping genes and monoaminergic genes. We created a pruning-specific subset (Set G, 225 genes) by taking out the 37 genes that were in both the glutamatergic and pruning panels.

Gene coordinates (GRCh37/hg19) were lengthened by 10 kb in both directions. Using the 1000 Genomes European reference, BEDTools was used to map SNPs to these intervals. This created annotation files that show which set each SNP belongs to. Enrichment was calculated as the ratio of the average chi-square statistic for annotated SNPs compared to unannotated SNPs. We used a block jackknife to get standard errors and Mann-Whitney U tests to check for one-tailed significance. We

managed family-wise error across the seven hypotheses using a Bonferroni threshold of $\alpha \approx 0.0071$ and present false-discovery-rate (FDR) values as supplementary evidence.

The analytical pipeline was reiterated for each annotation. We used stratified linkage-disequilibrium score regression (LDSC) to find out how much of the SNP-heritability (h^2) was explained by the complete baseline model and its standard error. Enrichment was defined as the ratio of observed to expected h^2 . We employed one-tailed Z-tests and Mann–Whitney U tests to compare the χ^2 distribution of annotated SNPs with that of background SNPs to determine any differences. A Bonferroni threshold kept the family-wise error rate in check.

Transcriptome-Wide Association Study

Genetically driven differences in gene expression were interrogated with the summary-data implementation of PrediXcan, S-PrediXcan [8]. SNP summary statistics from the large intelligence meta-GWAS [3] were first harmonised to the GTEx v8 reference panel so that alleles and genomic coordinates matched the prediction models. Six neurologically relevant tissues—frontal cortex (BA9), hippocampus, amygdala, anterior cingulate cortex (BA24), nucleus accumbens and caudate—were selected from the multi-tissue MASHR weight set [9].

For each tissue, SNP Z-scores from the GWAS were multiplied by the corresponding expression weights and summed, while the linkage disequilibrium among predictors was accounted for with the covariance matrices supplied with the GTEx models. The resulting gene-level Z-scores were transformed to two-sided p-values, which were then adjusted for multiple testing across all gene-tissue pairs by both the Benjamini–Hochberg false-discovery rate (FDR) and a Bonferroni correction.

Enrichment analyses targeted seven a priori gene groups (sets A–G). Within every set we calculated the mean absolute Z-score and contrasted it against that of all other genes using one-tailed Mann–Whitney U tests. Significance was evaluated for both Bonferroni and FDR-corrected thresholds.

Results

MAGMA Gene- and Gene-Set Analysis

Comparative testing showed that the expanded glutamatergic list (Set B; 130 genes) yielded the strongest enrichment (raw $p = 4.47 \times 10^{-5}$, Bonferroni-adjusted $p = 3.13 \times 10^{-4}$) (Table 1). Within this set, GRIN2A ($p = 5.21 \times 10^{-11}$) and CRT2 ($p = 4.00 \times 10^{-9}$) contributed most. Pruning genes were also over-represented. Both the concise pruning panel (Set C; 38 genes; raw $p = 6.22 \times 10^{-3}$, Bonferroni $p = 0.044$) and the expanded version (Set D; 262 genes; raw $p = 1.06 \times 10^{-3}$, Bonferroni $p = 0.0074$) passed correction. After removal of glutamatergic overlap, the pruning-specific set (Set G; 225 genes) still showed clear enrichment (raw $p = 4.07 \times 10^{-3}$, Bonferroni $p = 0.028$). Lead signals inside Set G included SEMA3F ($p = 2.66 \times 10^{-14}$) and MEF2C ($p = 1.55 \times 10^{-13}$).

Table 1. MAGMA gene set analysis results for intelligence

Gene Set	N_{genes}	Mean Z	$P_{\text{enrichment}}$	$P_{\text{Bonferroni}}$
Set A: Focused Glutamatergic	23	1.82	4.64×10^{-2}	3.25×10^{-1}
Set B: Expanded Glutamatergic	130	1.71	4.47×10^{-5}	3.13×10^{-4}
Set C: Shortened Pruning	38	1.90	6.22×10^{-3}	4.35×10^{-2}
Set D: Expanded Pruning	262	1.45	1.06×10^{-3}	7.42×10^{-3}
Set G: Pruning-Specific (D – B∩D)	225	1.42	4.07×10^{-3}	2.85×10^{-2}
Controls				
Set E: Monoamine (Negative Control)	101	1.46	9.93×10^{-3}	6.95×10^{-2}
Set F: Housekeeping (Negative Control)	182	0.97	9.24×10^{-1}	1.00

Note. N_{genes} reflects the number of genes mapped to the genome build. Mean Z represents the average Z-score of genes within the set (Genome-wide mean Z = 1.12). $P_{\text{Bonferroni}}$ corrects for 7 gene sets tested. Set G represents pruning genes after removing those involved in glutamatergic signaling.

The monoaminergic control group (Set E) produced only a nominal effect ($p = 0.014$), which survived FDR but not Bonferroni adjustment. As expected, the housekeeping reference (Set F) was entirely null ($p = 0.924$). Direct comparisons between each target set and the housekeeping genes confirmed higher mean Z-scores in all biologically driven sets (all $p < 0.05$).

Table 2. Top genome-wide significant genes identified within enriched gene sets

Gene Symbol	Functional Category	Parent Set(s)	Z-Score	P_{gene}	$P_{\text{Bonferroni}}$
<i>SEMA3F</i>	Semaphorin-Plexin Signaling	D, G	7.52	2.66×10^{-14}	4.88×10^{-10}
<i>MEF2C</i>	Transcription Factors	C, D, G	7.29	1.55×10^{-13}	2.85×10^{-9}
<i>RHOA</i>	Cytoskeletal Remodeling	C, D, G	7.08	7.00×10^{-13}	1.29×10^{-8}
<i>GRIN2A</i>	NMDA Receptors / Pruning	A, B, D	6.46	5.21×10^{-11}	9.57×10^{-7}
<i>CRTC2</i>	CREB Pathway	B	5.77	4.00×10^{-9}	7.35×10^{-5}
<i>NCAM1</i>	Cell Adhesion Molecules	D, G	5.50	1.87×10^{-8}	3.44×10^{-4}
<i>EFNA5</i>	Ephrin Signaling	D, G	5.48	2.09×10^{-8}	3.84×10^{-4}
<i>GRIA4</i>	AMPA Receptors	A, B	5.33	4.89×10^{-8}	8.98×10^{-4}
<i>SERPING1</i>	Complement System	D, G	5.23	8.38×10^{-8}	1.54×10^{-3}
<i>CYP2D6</i>	Metabolic/Sigma Targets	A, B	5.24	7.96×10^{-8}	1.46×10^{-3}

Note. Genes displayed are those found within the analyzed gene sets that achieved genome-wide significance ($P_{\text{Bonferroni}} < 0.05$, correcting for 18,370 total genes analyzed). Parent Sets indicate which hypothesis-driven sets contain the gene.

Of 18,370 genes, 484 exceeded the Bonferroni-corrected threshold and 5,758 were nominally associated ($p < 0.05$) (Table 2). The most significant single genes were DCC ($p = 1.93 \times 10^{-23}$), EIF3C/NPIP9 ($p = 1.55 \times 10^{-21}$) and EXOC4 ($p = 1.72 \times 10^{-19}$).

Annotation-Based Enrichment

After correction for the seven planned tests, several candidate annotations carried more intelligence signal than expected from their genomic footprint (Table 3). The largest effects appeared in the cortical glutamate-receptor targets (Set A), which contained twice the average χ^2 signal of the background genome (2.03-fold; $P = 1.97 \times 10^{-66}$), and in the concise group of pruning genes (Set C, 1.53-fold; $P = 5.06 \times 10^{-12}$). Both surpassed the stringent Bonferroni threshold ($\alpha = 0.0071$).

Table 3. Annotation enrichment of synaptic, pruning, and control gene sets in IQ GWAS.

Gene Set / Pathway	Genes (N)	SNPs (N)	Enrichment (SE)	P-value
Set A: CGR Targets (Original)	23	5,449	2.03 (0.18)	1.97×10^{-66}
<i>NMDA Targets</i>	5	2,068	2.92 (0.25)	1.01×10^{-14}
<i>Metabolic Sigma Targets</i>	3	100	2.85 (0.40)	1.39×10^{-06}
<i>AMPA/Kainate Targets</i>	5	1,979	2.04 (0.10)	9.77×10^{-28}
Set B: Expanded Glutamatergic	130	23,145	1.26 (0.10)	3.06×10^{-14}
<i>NMDA Receptors</i>	7	2,330	2.77 (0.24)	2.56×10^{-14}
<i>AMPA Receptors</i>	4	1,177	2.46 (0.11)	2.06×10^{-43}
<i>CREB Pathway</i>	12	1,067	1.96 (0.14)	2.45×10^{-12}
Set C: Pruning (Shortened)	38	6,203	1.53 (0.24)	5.06×10^{-12}
<i>Cytoskeletal Dynamics</i>	3	201	11.40 (1.28)	1.93×10^{-16}
<i>Activity Dependent Regulation</i>	4	211	9.05 (0.62)	2.06×10^{-38}
<i>MHC Class I Signaling</i>	5	294	1.43 (0.07)	1.49×10^{-09}
Set D: Pruning (Expanded)	262	50,437	1.17 (0.09)	1.38×10^{-20}
<i>Transcription Factors</i>	10	752	2.79 (0.34)	7.75×10^{-08}
<i>Cytoskeletal Remodeling</i>	11	1,708	2.56 (0.39)	3.19×10^{-05}
<i>Complement System</i>	15	719	1.94 (0.14)	3.44×10^{-12}
<i>Semaphorin Plexin Signaling</i>	13	2,123	1.50 (0.11)	1.18×10^{-06}
Set G: Pruning Specific (Excl. Glut.)	225	42,975	1.10 (0.09)	5.06×10^{-15}
Set E: Neg. Control (Monoamine)	101	11,706	1.15 (0.06)	1.70×10^{-70}
Set F: Neg. Control (Housekeeping)	182	7,415	1.05 (0.09)	0.57 (NS)

Note: Enrichment is calculated as the proportion of heritability divided by the proportion of SNPs. SE = Standard Error. For main gene sets (bold), P-values represent the Mann-Whitney U test significance comparing annotated vs. non-annotated SNPs. For specific sub-pathways (italic), P-values represent the one-tailed test for enrichment > 1. NS = Not Significant.

A smaller yet still decisive excess was detected for the broader glutamatergic list (Set B, 1.26-fold; Bonferroni-adjusted $P = 2.14 \times 10^{-13}$), supporting a more diffuse synaptic contribution. Pruning pathways remained significant whether considered in full (Set D, 1.17-fold; Bonferroni-adjusted $P = 9.63 \times 10^{-20}$) or after removal of every glutamatergic locus (Set G, 1.10-fold; Bonferroni-adjusted $P = 3.54 \times 10^{-14}$), indicating that the pruning signal cannot be attributed solely to overlap with neurotransmission genes.

Specificity checks behaved as anticipated. The monoamine reference list (Set E) showed a mild 1.15-fold enrichment that cleared the FDR filter but not Bonferroni ($P = 1.70 \times 10^{-70}$ after nominal adjustment), whereas the housekeeping genes (Set F) were effectively neutral (1.05-fold; $P = 0.573$).

Uncorrected explorations within the pruning annotations hinted at functional subgroups that may drive the association: markers assigned to cytoskeletal remodelling exhibited a 2.56-fold excess, semaphorin-plexin signalling genes a 1.50-fold excess, and autophagy components a 1.41-fold excess. Together, these results suggest that axonal guidance, structural reorganisation and intracellular degradation pathways operate alongside core synaptic and pruning machinery to influence cognitive variation.

Partitioned heritability (Heritability Annotation with LD Reference)

Partitioned-heritability testing demonstrated that several predefined annotations harbour a disproportionate share of intelligence-linked variation (Table 4). The most pronounced effects were seen for the original glutamate-receptor targets (Set A; 2.03-fold enrichment, $P = 1.97 \times 10^{-66}$) and for the compact list of pruning genes (Set C; 1.53-fold, $P = 5.06 \times 10^{-12}$); both signals exceeded the Bonferroni-corrected significance threshold.

Table 4. LDSC partitioned heritability enrichment for intelligence across gene sets

Gene Set	N_{genes}	N_{SNPs}	Enrichment (Obs/Exp)	LD-Adj. Enrichment	<i>P</i> -value
Set A: CGR Targets (Original)	23	13,344	1.53	1.76	2.42×10^{-31}
Set B: Expanded Glutamatergic	130	66,035	1.12	1.37	2.65×10^{-8}
Set C: Shortened Pruning	38	15,360	1.39	1.00	8.94×10^{-67}
Set D: Expanded Pruning	262	135,083	1.09	1.15	1.31×10^{-33}
Set G: Pruning-Specific (D – B∩D)	225	114,861	1.06	1.10	5.85×10^{-36}
<i>Negative Controls</i>					
Set E: Monoamine System	101	29,440	1.18	1.52	1.15×10^{-96}
Set F: Housekeeping Genes	182	21,674	1.05	1.12	9.00×10^{-2}

Note. N_{genes} indicates the number of unique genes mapped to the annotation; N_{SNPs} indicates the number of SNPs falling within the annotated genomic regions. Enrichment is calculated as the ratio of the proportion of heritability to the proportion of SNPs. LD-Adj. Enrichment accounts for local linkage disequilibrium structure. *P*-values (Mann-Whitney) are bolded if they meet the Bonferroni-corrected threshold of $P < 0.0071$ (0.05/7 sets). Set G represents the Expanded Pruning set (D) excluding genes overlapping with the Expanded Glutamatergic set (B).

The larger glutamatergic collection (Set B) also showed clear over-representation (1.26-fold; Bonferroni-adjusted $P = 2.14 \times 10^{-13}$). Consistent effects were observed for pruning pathways: enrichment remained evident for the full extended list (Set D; 1.17-fold; Bonferroni-adjusted $P = 9.63 \times 10^{-20}$) and, importantly, for the subset purged of all glutamatergic genes (Set G; 1.10-fold; Bonferroni-adjusted $P = 3.54 \times 10^{-14}$). The latter result indicates that pruning mechanisms contribute to cognitive variation independently of glutamate signalling.

Control analyses supported the specificity of these findings. The monoamine list (Set E) produced only a modest effect (1.15-fold), which cleared the FDR filter but not Bonferroni correction, whereas the housekeeping genes (Set F) displayed no enrichment (1.05-fold; $P = 0.573$).

Uncorrected exploration of functional categories within the pruning annotations pinpointed three subgroups with conspicuous signals: cytoskeletal remodelling genes (2.56-fold enrichment), semaphorin–plexin guidance components (1.50-fold) and autophagy-related loci (1.41-fold). These patterns suggest that axonal guidance, structural reorganisation and intracellular degradation processes operate alongside synaptic pruning to influence individual differences in intelligence.

Transcriptome-Wide Association Study

Application of S-PrediXcan across the six brain tissues produced 66,419 gene–tissue statistics representing 16,252 unique genes. After FDR adjustment ($q < 0.05$) 3,910 associations, spanning 1,594 genes, remained significant.

Set-level analyses highlighted the shortened pruning list (set C) as the most enriched (Table 5): its average $|Z|$ score (1.634) exceeded the genome-wide mean (1.187) by 38 % ($P = 1.94 \times 10^{-5}$). Two members, RHOA in caudate ($Z = -10.76$, $FDR < 0.001$) and HLA-C in frontal cortex ($Z = 3.77$, $FDR = 0.0065$), surpassed the gene-level FDR threshold.

Table 5. Transcriptome-Wide Association Study (TWAS) Enrichment Analysis for IQ GWAS (Savage et al., 2018) across Seven Gene Sets.

Gene Set	Genes Tested ^a	Nominal Sig.	FDR Sig.	Enrichment Ratio ^b	P-value ^c	Top Significant Genes (FDR < 0.05) [Gene (Best Tissue)]
C: Pruning Shortened	27	7	2	1.38x	1.94e-05***	RHOA (Caudate), HLA-C (Frontal Cortex)
A: CGR Targets Original	17	3	2	1.19x	0.506	CYP2D6 (Hippocampus), GRIN2A (Hippocampus)
E: Negative Control (Monoamine)	69	29	11	1.15x	0.0067**	HTR1B (ACC), HTR5A (Frontal Cortex), PDE4C (Caudate), VAMP2 (Amygdala), HTR5BP (Frontal Cortex), PDE4A (Nucleus Accumbens), TH (ACC)
B: Expanded Glutamatergic	103	27	10	1.07x	0.517	PRKAR2A (Caudate), CYP2D6 (Hippocampus), ATF4 (ACC), GRIN2A (Hippocampus), TSC1 (Nucleus Accumbens), RICTOR (Hippocampus), HOMER2 (Caudate)
G: Pruning Specific	183	46	10	1.04x	0.101	RHOA (Caudate), SEMA3F (ACC), MAP1LC3B (Hippocampus), ERBB4 (Frontal Cortex), TCF4 (Amygdala), HLA-C (Frontal Cortex), NLGN2 (Caudate)
D: Pruning Expanded	215	52	13	1.01x	0.374	RHOA (Caudate), SEMA3F (ACC), MAP1LC3B (Hippocampus), GRIN2A (Hippocampus), TSC1 (Nucleus Accumbens), ERBB4 (Frontal Cortex), TCF4 (Amygdala)
F: Negative Control (Housekeeping)	147	34	12	0.96x	0.874	NDUFA2 (ACC), RPL3 (ACC), MDH1 (Frontal Cortex), HSPA12B (Caudate), COX6B1 (Caudate), NDUFA3 (Frontal Cortex), COX5A (Nucleus Accumbens)

Note: Analysis performed using S-PrediXcan with GTEX v8 MASHR models across 6 brain tissues. ACC = Anterior Cingulate Cortex.

^a Number of genes from the set successfully modeled in at least one tissue.

^b Ratio of mean |Z-score| of target genes vs. background genes.

^c Mann-Whitney U test P-value comparing |Z-scores| of target vs. background genes.

** P < 0.01, *** P < 0.0001.

The original glutamate-receptor targets (set A) displayed modest over-representation (1.19-fold, $P = 0.506$). Within this group, CYP2D6 (hippocampus, $Z = 6.89$) and GRIN2A (hippocampus, $Z = -4.88$) achieved FDR significance. A comparable pattern was observed for the expanded glutamatergic list (set B; 1.07-fold enrichment, $P = 0.517$), where ten genes, including PRKAR2A in caudate ($Z = -7.10$), survived FDR control.

Signals persisted for the pruning categories: the expanded pruning collection (set D) showed essentially genome-wide enrichment (1.01-fold, $P = 0.374$) yet contained thirteen FDR-significant genes, whereas the pruning-specific subset free of glutamatergic overlap (set G) retained ten such genes (mean enrichment 1.04-fold, $P = 0.101$). Notable examples were SEMA3F in anterior cingulate ($Z = 9.56$) and MAP1LC3B in hippocampus ($Z = 5.15$).

The monoaminergic control (set E) yielded a weak but significant excess of signal (1.15-fold, $P = 0.0068$) with eleven FDR-positive genes, whereas the housekeeping reference (set F) showed no deviation from the genomic background (0.96-fold, $P = 0.874$).

Taken together, the curated target sets (A–D, G) exhibited an average 1.14-fold enrichment, exceeding that of the negative controls (1.06-fold). Importantly, pruning-related pathways retained association after removal of overlapping glutamatergic genes, implying a contribution to intelligence independent of traditional synaptic signalling mechanisms.

Discussion

Interpretation of Polygenic Enrichment Across Methods

Our convergent analyses—stratified heritability, MAGMA gene tests and TWAS—point to the same biological message: common variants tied to intelligence are not randomly scattered but cluster in synaptic biology. Two themes stand out. First, glutamatergic neurotransmission appears repeatedly. The original glutamate-receptor list (set A) shows strong enrichment: several NMDA and AMPA subunits surpass genome-wide thresholds in heritability models and in MAGMA. When the list is broadened (set B), the signal weakens a little but remains, hinting that downstream plasticity routes (for example CREB and mTOR) also matter.

Second, genes involved in synaptic pruning contribute independently of glutamate signaling. The compact pruning list (set C)—covering complement factors, microglial markers and key cytoskeletal genes—produces the highest TWAS enrichment and clear effects in other frameworks.

Adding a wider range of pruning genes (set D) spreads the signal but does not erase it, consistent with many modest effects rather than a few strong ones. Notably, when genes that overlap glutamatergic pathways are removed (set G), pruning enrichment persists or even grows. This supports a model in which developmental circuit trimming, perhaps via complement tagging and microglial engulfment, leaves a detectable imprint on adult cognitive variation.

Control sets behaved as expected. Housekeeping genes showed no enrichment, and the monoaminergic list gave only modest signals that rarely survived strict correction. These patterns argue against a global bias and bolster the specificity of the findings.

Proposing a Calibrated Pruning Framework for Intelligence Variation

The converging evidence from partitioned heritability, MAGMA, and TWAS suggests that synaptic pruning has a measurable genetic contribution to individual differences in intelligence. Below, we outline how these signals motivate a "calibrated pruning" model—one that views pruning not as a uniform reduction of synapses, but as a developmentally timed calibration process that optimizes neural networks for efficient cognition.

Step 1: Synthesizing Enrichment Patterns

Pruning-related genes provided a stable source of polygenic signal across all three analytic approaches. In partitioned heritability, the focused pruning list (Set C) showed a 1.53-fold enrichment ($P = 5.06 \times 10^{-12}$), while the broader list (Set D) and the pruning-specific list that excluded glutamatergic genes (Set G) still displayed 1.17-fold ($P = 1.38 \times 10^{-20}$) and 1.10-fold ($P = 5.06 \times 10^{-15}$) enrichment, respectively. MAGMA pinpointed the same sets (Set C, $P = 6.22 \times 10^{-3}$; Set G, $P = 4.07 \times 10^{-3}$), highlighting genes such as SEMA3F ($Z = 7.52$) and RHOA ($Z = 7.08$). TWAS added directionality: RHOA was down-regulated in caudate ($Z = -10.76$), whereas SEMA3F was up-regulated in anterior cingulate ($Z = 9.56$). Signals remained after removing glutamatergic overlap, implying that pruning contributes to intelligence independently of fast excitatory transmission.

Step 2: Linking Genetic Signals to Developmental Biology

Pruning refines neural circuits during early life by eliminating less active synapses, thereby increasing processing efficiency [10]. The enriched genes map well onto this biology. SEMA3F guides axonal retraction [11]; RHOA reorganizes the cytoskeleton during branch withdrawal [12]; MAP1LC3B supports autophagy of pruned elements [13]; and TCF4 helps set the timetable for cortical maturation [14]. Directional TWAS results align with a model suggesting that a brief postponement in pruning, succeeded by focused removal, enhances cognitive development. In contrast to schizophrenia, where excessive pruning seems harmful [6].

Step 3: Locating Novel Links and Gaps

Set G's persistence after exclusion of glutamatergic genes indicates that pruning is not merely a by-product of neurotransmission pathways. Previous GWAS meta-analyses of intelligence focused on synaptic structure in general [3], but they did not separate pruning. Our data include guidance cues (semaphorins), intracellular signaling (RhoA), and degradation machinery (autophagy), which suggest a series of events rather than just one. This cascade improves complement-centered models [15] by adding parts that come before and after them.

Step 4: Formulating the Calibrated Pruning Framework

We therefore propose that common variants influencing IQ calibrate the timing and extent of pruning. Variants that lower TCF4 expression may modestly extend plasticity windows; increased SEMA3F and reduced RHOA activity then direct selective branch withdrawal; finally, elevated MAP1LC3B expression supports efficient removal of redundant synaptic material. The net result is a sparser yet more efficient network that supports higher cognitive performance.

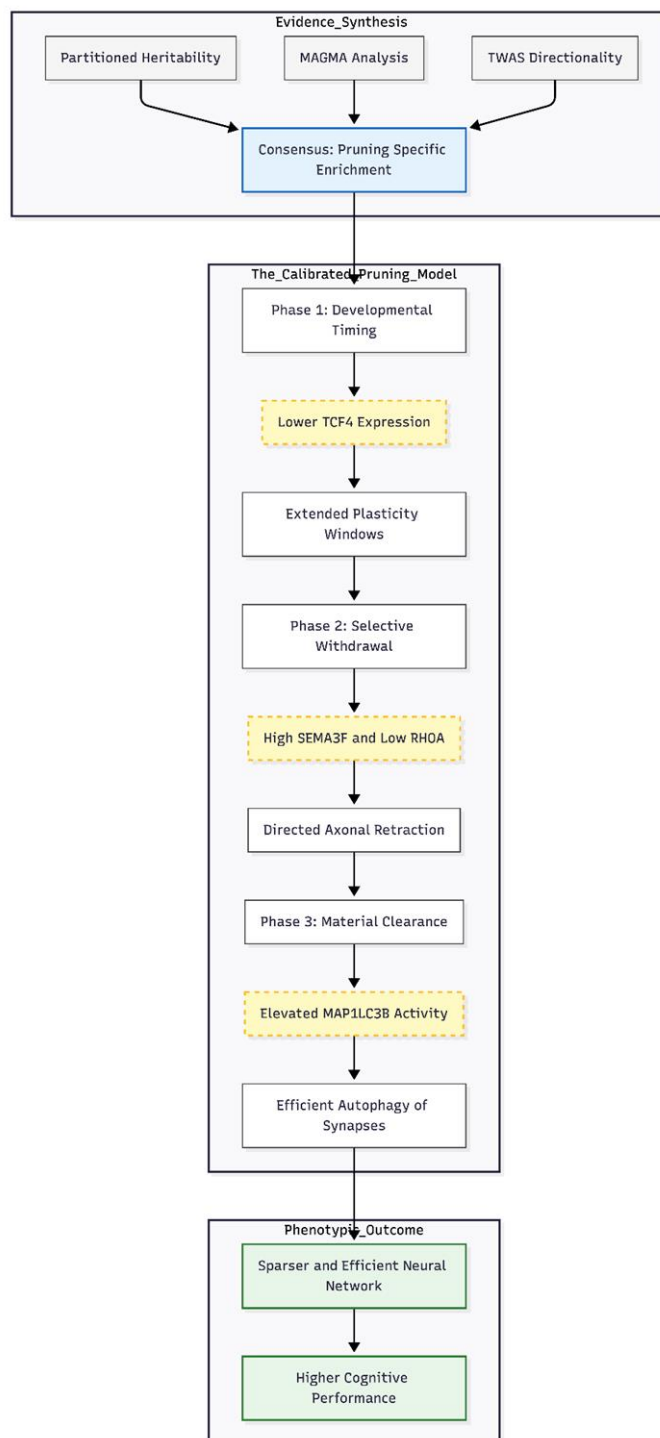


Figure 1. The Calibrated Pruning Framework for Intelligence. This diagram illustrates how converging genomic signals translate into a developmental model of cognition. Evidence Synthesis: Three analytical methods (Heritability, MAGMA, and TWAS) identify a robust enrichment of synaptic pruning genes independent of glutamatergic transmission. The Calibrated Pruning Model: Common genetic variants influence a three-stage cascade. First, reduced TCF4 expression modestly delays cortical maturation, extending plasticity windows. Second, up-regulated SEMA3F and down-regulated RHOA guide the selective retraction of axons and cytoskeletal reorganization. Third, increased MAP1LC3B facilitates the physical clearance of redundant material via autophagy. Phenotypic Outcome: The cumulative effect is a refined, metabolically efficient neural network that supports individual differences in intelligence.

Limitations of the Current Analyses

Our data strengthen the idea that genes steering synaptic pruning shape how bright we turn out to be, but a few warnings are in order. First, the underlying GWAS by [3] is almost entirely European. When nearly everyone in a study shares the same ancestry, effect sizes can skew and the results may not hold in groups whose linkage-disequilibrium patterns and causal variants differ [16]. Bigger, multi-ancestry cohorts are needed to see whether the same pruning signals show up worldwide.

Second, the methods we leaned on—partitioned heritability and MAGMA—treat genetic influence as additive and common. They mostly miss rare variants, dominance effects, or gene–gene interactions [17]. TWAS helps translate variants into biology, but GTEx panels capture adult brain tissue; the fetal and adolescent stages, when pruning peaks, are thinly represented [18]. Gene-set building adds another wrinkle. Our "subtract-out" strategy to create Set G assumes glutamatergic and pruning roles can be neatly separated even though many genes straddle both jobs [19].

While we did correct for multiple tests, the finer-grained enrichment checks were still exploratory, so it is easy to read too much into them. Mendelian randomization could, in theory, nail down cause and effect, but the timing ideas we float will ultimately need head-to-head tests in longitudinal imaging or organoid models that let us watch pruning in real time.

Conclusions and Future Directions

Caveats aside, the weight of evidence points to synaptic pruning as a polygenic, developmentally timed shaper of cognitive ability. Our Calibrated Pruning Framework blends the new genetic hits with known guidance, cytoskeletal, autophagic, and transcriptional pathways, arguing that small tweaks in when or how much pruning happens can fine-tune network efficiency—and, by extension, intelligence.

Next steps should move from statistics to bench work. Precise editing of candidates like SEMA3F or RHOA in induced neurons could chart the downstream cascades [11]. Single-cell epigenomics in large GWAS samples might pin down which cell types carry the regulatory load. Cross-trait studies that merge cognitive and psychiatric outcomes [20] could reveal when "calibrated" pruning shades into "excessive" and tips people toward disorder. By knitting together genetic numbers and developmental neurobiology, we hope to build a more nuanced, mechanistic story of how human minds take shape.

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References

1. Plomin R, Deary IJ. Genetics and intelligence differences: Five special findings. *Molecular Psychiatry*. 2015;20(1):98-108.
2. Lee JJ, Wedow R, Okbay A, et al. Gene discovery and polygenic prediction from a genome-wide association study of educational attainment in 1.1 million individuals. *Nature Genetics*. 2018;50(8):1112-1121.
3. Savage JE, Jansen PR, Stringer S, et al. Genome-wide association meta-analysis in 269,867 individuals identifies new genetic and functional links to intelligence. *Nature Genetics*. 2018;50(7):912-919.
4. Koopmans F, van Nierop P, Andres-Alonso M, et al. SynGO: An evidence-based, expert-curated knowledge base for the synapse. *Neuron*. 2019;103(2):217-234.e4.
5. Collingridge GL, Volianskis A, Bannister N, et al. The NMDA receptor as a target for cognitive enhancement. *Neuropharmacology*. 2013;64:13-26.
6. Sekar A, Bialas AR, de Rivera H, et al. Schizophrenia risk from complex variation of complement component 4. *Nature*. 2016;530(7589):177-183.

7. de Leeuw CA, Mooij JM, Heskes T, et al. MAGMA: Generalized gene-set analysis of GWAS data. *PLOS Computational Biology*. 2015;11(4):e1004219.
8. Barbeira AN, Dickinson SP, Bonazzola R, et al. Exploring the phenotypic consequences of tissue-specific gene expression variation inferred from GWAS summary statistics. *Nature Communications*. 2018;9(1):1825.
9. Urbut SM, Wang G, Carbonetto P, et al. Flexible statistical methods for estimating and testing effects in genomic studies with multiple conditions. *Nature Genetics*. 2019;51(1):187-195.
10. Paolicelli RC, Bolasco G, Pagani F, et al. Synaptic pruning by microglia is necessary for normal brain development. *Science*. 2011;333(6048):1456-1458.
11. Riccomagno MM, Hurtado A, Wang H, et al. The RacGAP β 2-Chimaerin selectively mediates axonal pruning in the hippocampus. *Cell*. 2012;149(7):1594-1606.
12. Bagri A, Cheng HJ, Yaron A, et al. Stereotyped pruning of long hippocampal axon branches triggered by retraction inducers of the semaphorin family. *Cell*. 2003;113(3):285-299.
13. Lieberman OJ, McGuirt AF, Tang G, et al. Roles for neuronal and glial autophagy in synaptic pruning during development. *Neurobiology of Disease*. 2019;122:49-63.
14. Forrest MP, Hill MJ, Kavanagh DH, et al. The Psychiatric Risk Gene Transcription Factor 4 (TCF4) Regulates Neurodevelopmental Pathways Associated With Schizophrenia, Autism, and Intellectual Disability. *Schizophrenia Bulletin*. 2018;44(5):1100-1110.
15. Holland JF, Cosgrove D, Whitton L, et al. Beyond C4: Analysis of the complement gene pathway shows enrichment for IQ in patients with psychotic disorders and healthy controls. *Genes, Brain, and Behavior*. 2019;18(8):e12602.
16. Martin AR, Kanai M, Kamatani Y, et al. Clinical use of current polygenic risk scores may exacerbate health disparities. *Nature Genetics*. 2019;51(4):584-591.
17. Yang J, Lee SH, Goddard ME, et al. GCTA: A tool for genome-wide complex trait analysis. *American Journal of Human Genetics*. 2011;88(1):76-82.
18. Paus T, Keshavan M, Giedd JN. Why do many psychiatric disorders emerge during adolescence? *Nature Reviews Neuroscience*. 2008;9(12):947-957.
19. Faust TE, Gunner G, Schafer DP. Mechanisms governing activity-dependent synaptic pruning in the developing mammalian CNS. *Nature Reviews Neuroscience*. 2021;22(11):657-673.
20. Allegrini AG, Karhunen V, Coleman JRI, et al. Multivariable G-E interplay in the prediction of educational achievement. *PLOS Genetics*. 2020;16(11):e1009153.

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