Mapping the human brain epigenome and its links to disease

Peter Hickey

Department of Biostatistics

Johns Hopkins Bloomberg School of Public Health

@PeteHaitch

Current map of human brain methylome is limited

GB-Seq

RBS

DIP-Sec

RE-Seq

	Σ	Σ	R	3
Brain				2
Brain_Angular_Gyrus			1	
Brain_Anterior_Caudate			2	
Brain_Cingulate_Gyrus			1	
Brain_Germinal_Matrix		2		1
Brain_Hippocampus_Middle				2
Brain_Inferior_Temporal_Lobe			1	
Brain_Mid_Frontal_Lobe			1	
Brain_Substantia_Nigra			2	
Brain-Frontal Lobe-Left				1
Brain-Frontal Lobe-Right				1
Brain-Temporal lobe-Left				1
Fetal_Brain	3	5	2	
Neurosphere_Cultured_Cells_Cortex_Derived	2	2		2
Neurosphere_Cultured_Cells_Ganglionic_Eminence_Derived	2	2		2
http://epigenomesportal.ca/ihec/grid.html (Bui	ld:	201	7-1	.0)

• Bulk tissue

- Limited replicates
- Few brain region-specific DMRs^{1,2}

¹Davies, M. N. *et al.* Functional annotation of the human brain methylome identifies tissue-specific epigenetic variation across brain and blood. *Genome Biol.* **13**, R43 (2012).
²Roadmap Epigenomics Consortium et al. Integrative analysis of 111 reference human epigenomes. Nature 518, 317–330 (2015). A good map requires biological replicates, multiple brain regions, and multiple cell types



Bulk tissue samples are uninformative for brain region-specificity due to variation of neuronal proportion in sampled tissue



PC1 (12.7%)



PCA: Bulk Tissue mCG

FANS + WGBS reveals brain region-specificity of mCG in NeuN+ but not NeuN- samples PCA: sorted nuclei mCG



PC1 (53.4%)

Summary of DMRs

	Cell type DMRs	Brain region DMRs		
	CG-DMRs (NeuN+ vs. NeuN-)	CG-DMRs (NeuN+)	CG-DMRs (NeuN-)	CH-DMRs (NeuN+)
n	100,875*	13,074	114	15,029+
Total size (Mb)	70.0	11.9	0.1	39.6++
Median (10-90%) width (bp)	612 (296 – 1157)	809 (671 – 3267)	767 (459 – 1789)	3558 (2421 – 9269

*21,802 novel DMRs

⁺Before merging across strand and context

⁺⁺After merging across strand and context

mCH shows little strand specificity and 'tracks' mCG (L) PCA: NeuN+ mCH (1kb bins)



PC1 (22.5%)

PC2

CG-DMRs and CH-DMRs co-occur

 $\log 2(OR)$

4

CG-DMRs are enhancer-centric, CH-DMRs are enriched over differentially expressed genes (DEGs) relative to non-DEGs

		OCR (union) H3K27ac FANTOM5 CH-DMRs (NeuN+) DEGs CG-DMRs (NeuN+) DEG promoters intronic Shelves exonic three_utr Shores promoter CGI OpenSea SINE DNA Simple_repeat Low_complexity five_utr intergenic LTR LINE Satellite
CG-DMR (NeuN+)	CH-DMR (NeuN+)	

FANS + ATAC-seq reveals brain region-specificity of chromatin accessibility in NeuN+ but not NeuN- samples PCA: sorted nuclei ATAC



PC1 (48.1%)

FANS + ATAC-seq reveals brain region-specificity of chromatin accessibility in NeuN+ but not NeuN- samples

	OCRs	DARs	DARs	DARs
	(overall)	(NeuN+ vs. NeuN-)	(NeuN+)	(NeuN-)
n	836,627	163,026	68,021	13
Total size (Mb)	619.5	275.8	118.1	0.05
Median (10-90%)	447	1176	1243	3739
width (bp)	(228 – 1459)	(659 – 3202)	(671 – 3267)	(1303 – 7541)

OCRs = Open Chromatin Regions are enriched over genic and regulatory-like features DARs = Differentially Accessible Regions are enriched over CG-DMRs

Linking brain region-specific epigenetic differences to disease

- Hypothesis: Regulatory regions in relevant cell types contain 'GWAS signal'
- Stratified Linkage Disequilibrium Score Regression (SLDSR)¹
 - Estimate per-SNP heritability of trait from genome wide association study data
 - Partition the heritability by genomic features
- Traits (n = 30): E.g., schizophrenia, neuroticism, ADHD,
- Baseline features (n = 53): E.g., conserved regions, promoters, DHS
- Brain-derived features (n = 5): E.g., CG-DMRs, DARs, H3K27ac³
- Questions:
 - Does adding the brain-derived feature explain additional heritability over the 53 baseline features?
 - Are the brain-derived features enriched for heritability of the trait?

¹ Finucane, H. K. *et al.* Partitioning heritability by functional annotation using genome-wide association summary statistics. *Nat. Genet.* **47**, 1228–1235 (2015).
²Vermunt, M. W. et al. Large-scale identification of coregulated enhancer networks in the adult human brain. Cell Rep. 9, 767–779 (2014)

Traits with a brain-derived feature that explains additional heritability over baseline features

Psychiatric

CNS (LDSC)

CG-DMRs (NeuN+

Brain H3K27ac DARs (NeuN+) chromHMM (union)

CNS (LDSC



ADHD

BMI

College_attainment

Ever_smoked



- Sorting is critical to identify brain region-specific epigenomic and transcriptomic changes
- More diverse brain regions brings lots to the party
- Little brain region-specificity of NeuN- data (WGBS, ATAC, RNA)
 - Additional sorting will help but not currently feasible
- CG-DMRs enriched for heritability of brain traits
- Data will be available as custom track hub on UCSC

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http://biorxiv.org/content/early/2017/03/24/120386

Summary

- Sorting is critical to identify brain region-specific epigenomic and transcriptomic changes
- More diverse brain regions brings lots to the party
- Little brain region-specificity of NeuN- data (WGBS, ATAC, RNA)
 - Additional sorting will help but not currently feasible
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Supplementary slides

G-DMRs are enriched over regulatory-like regions



*Vermunt, M. W. et al. Large-scale identification of coregulated enhancer networks in the adult human brain. Cell Rep. 9, 767–779 (2014).

OCRs enriched over genic and regulatory-like features DARs enriched over CG-DMRs







mCH is restricted to NeuN+ and shows little strand specificity PCA: NeuN+ mCH (1kb bins)

5

0



Consistent changes in chromatin accessibility and mCG within CG-DMRs and DARs

(g)

0.004 0.003 0.002 0.001 0

0.5

0 -0.5 -1.0





OCRs enriched over genic and regulatory-like features DARs enriched over CG-DMRs





No significant feature Alzheimer's disease Anorexia nervosa Anxiety disorder Autism spectrum disorder Childhood cognitive performance Cigarettes per day Conscientiousness Extraversion Intracerebral hemorrhage Ischemic stroke Major depressive disorder Openness PTSD Subjective well-being

> Coronary artery disease Crohn's disease Height

Up in NAcc_pos



Down in NAcc_pos



GO:0048666: neuron development GO:0099536: synaptic signaling GO:0007417: central nervous system development R-HSA-112316: Neuronal System R-HSA-422475: Axon guidance GO:0007264: small GTPase mediated signal transduction GO:0043269: regulation of ion transport ko04024: cAMP signaling pathway GO:0044708: single-organism behavior GO:0048646: anatomical structure formation involved in morphogenesis GO:0042391: regulation of membrane potential GO:0009611: response to wounding GO:0035295: tube development GO:0090066: regulation of anatomical structure size GO:0042327: positive regulation of phosphorylation GO:0050808: synapse organization GO:0051046: regulation of secretion hsa04360: Axon guidance GO:0099504: synaptic vesicle cycle GO:0051050: positive regulation of transport

