Epigenesis
And Psychiatric Disorders

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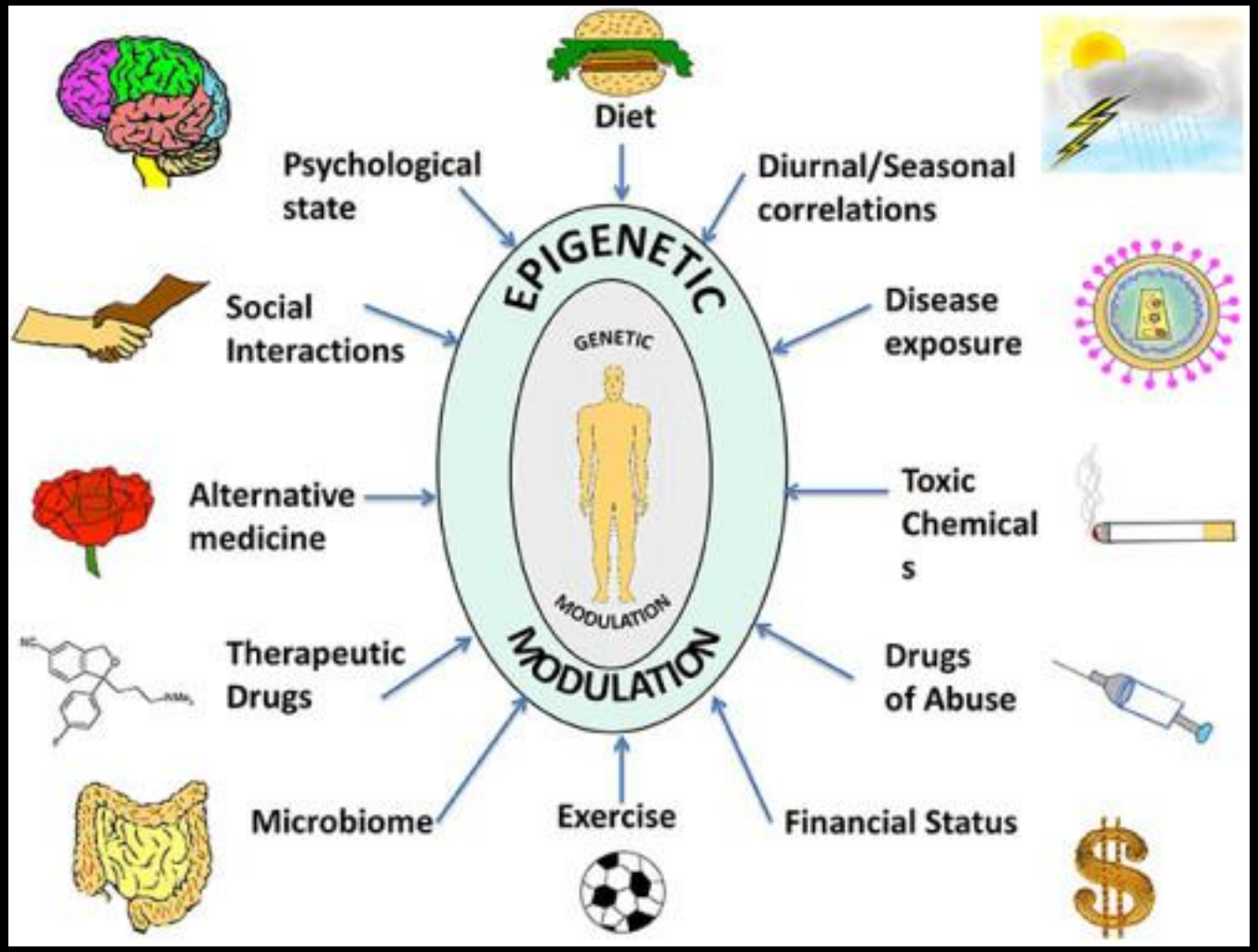
Introduction & Overview


Diet
Psychological state
Diurnal/Seasonal correlations
EPIGENETIC
GENETIC
MODULATION
MODULATION
Social Interactions
Disease exposure
Alternative medicine
Toxic Chemicals
Therapeutic Drugs
Drugs of Abuse
Microbiome
Exercise
Financial Status

Epigenetic modes of gene regulation can be grouped into **three general domains**:

1. Histone Post-Translational Modifications **(PTMs)** and histone variant exchange
2. Chromatin **Remodeling**
3. DNA **Methylation**

- A large variety of histone PTMs have been identified, including phosphorylation, acetylation, methylation, adenosine diphosphate (ADP) ribosylation, ubiquitination, crotonylation, and small ubiquitin-like modifier (SUMO)ylation.

- **<u>The Writers & The Erasers</u>**
  - HATs, HDACs – Specificity for K residues poorly understood
  - HMTs, HDMs – Specificity for K & Arginine residues clearly understood

- **<u>The Readers</u>**
  - Specific proteins that bind to the K & Arginine residues, bring about transcriptional change
  - Hundreds of proteins are thought to be recruited to a gene in concert with its activation or repression

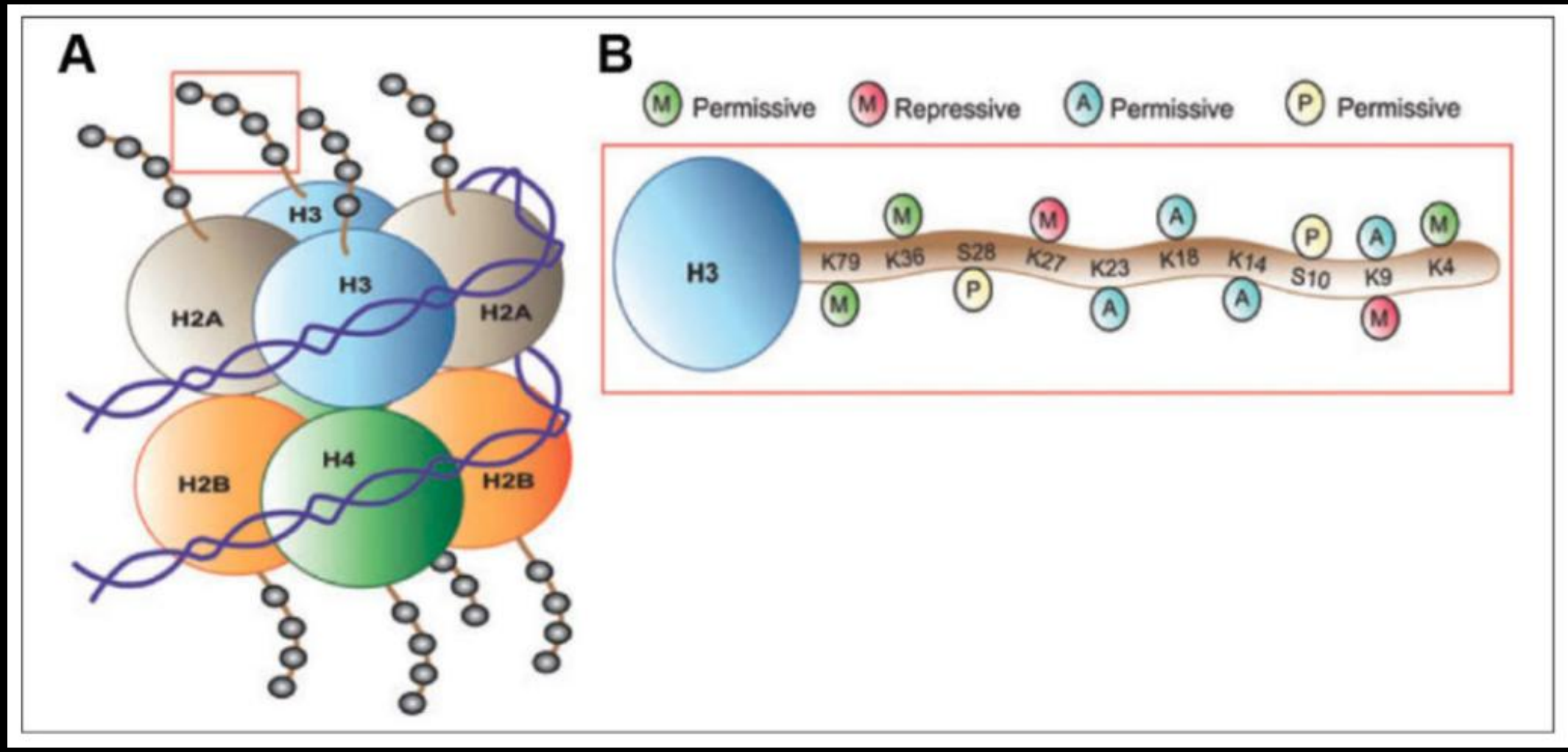
- **Acetylation** generally **promotes decondensation** of chromatin and increases gene activity by negating the positive charge of K residues in histone tails and increases spacing between nucleosomes

- **Methylation can either promote or repress** gene activity, depending on the residue undergoing methylation

- **Phosphorylation** of histones is also associated with **chromatin inhibition or activation**

- Other modifications poorly understood

- **Histone code hypothesis** - the sum of modifications at a particular gene defines a specific epigenetic state of gene activation or silencing


A
H3
H3
H2A
H2A
H2B
H4
H2B
B
M Permissive
M Repressive
A Permissive
P Permissive
H3
K79
K36
S28
K27
K23
K18
K14
S10
K9
K4
M
M
P
M
A
A
P
A
M
M

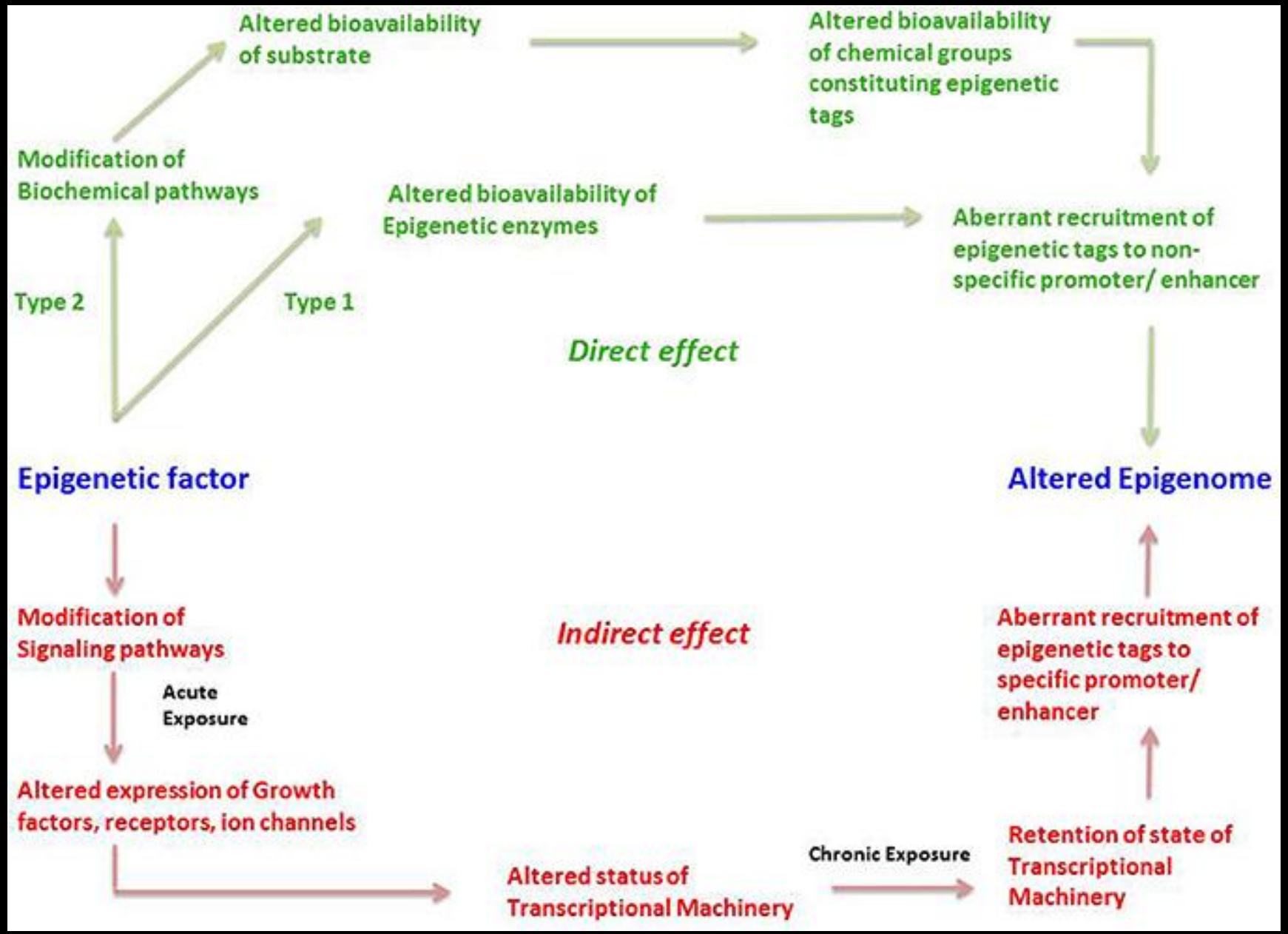

Altered bioavailability of substrate
Altered bioavailability of chemical groups constituting epigenetic tags
Modification of Biochemical pathways
Altered bioavailability of Epigenetic enzymes
Aberrant recruitment of epigenetic tags to non-specific promoter/ enhancer
Type 2
Type 1
Direct effect
Epigenetic factor
Altered Epigenome
Modification of Signaling pathways
Indirect effect
Aberrant recruitment of epigenetic tags to specific promoter/ enhancer
Acute Exposure
Altered expression of Growth factors, receptors, ion channels
Chronic Exposure
Retention of state of Transcriptional Machinery
Altered status of Transcriptional Machinery

Table 1.1 Heritability of Psychiatric Disorders

Disease	Heritability
Schizophrenia	0.81
Autism spectrum disorder	0.80
Bipolar disorder	0.75
Major depression	0.37
Attention deficit disorder	0.75
Alzheimer's disease	0.58

Source: Sullivan PF, et al. Genetic architectures of psychiatric disorders: the emerging picture and its implications. Nat Rev Genet 2012; 13: 537–52.

Table 1.2 CNVs Showing Effects on Multiple Psychiatric Phenotypes

	SZ	BP	ASD	ID
1a21.1 deletion	8.1 (4.3–15.6)	2.9 (1.2–6.9)	8.0 (3.5–18.4)	12.6 (7.4–21.3)
1q21.1 duplication	4.2 (2.1–8.6)			4.4 (2.6–7.4)
3q29 deletion	63.0 (8.1–491.7)	27.3 (2.5–301.5)	30.0 (1.9–480.4)	41.8 (5.6–311.6)
7q11.23 deletion	—	—	30.7 (3.4–275.1)	++++
7q11.23 duplication				16.5 (2.2–124.5)
VIPR2 (7q36.3) duplication	3.2 (1.5–7.1)	—	—	—
15q11.2 deletion	2.1 (1.6–2.8)	—	42.6 (15.7–115.5)	1.9 (1.6–2.3)
15q11.2 duplication	5.1 (1.4–19.1)	—	10.8 (3.5–33.1)	18.5 (7.1–47.9)
15q13.3 deletion	10.7 (5.4–21.3)	—	—	15.1 (8.4–27.4)
16p13.3 deletion	2.0 (1.1–3.5)	—	9.5 (5.2–17.4)	2.4 (1.8–3.2)
16p11.2 deletion	9.4 (5.3–16.6)	3.9 (1.9–8.2)	11.8 (6.1–22.7)	9.2 (5.8–14.7)
16p11.2 duplication				3.4 (1.8–6.5)
17p12/HNPP deletion	5.7 (2.4–13.7)	—	—	—
17q12 deletion	9.5 (2.4–38.2)	—	16.0 (2.9–87.9)	17.3 (6.1–49.0)
22q11.21 deletion	++++	++++	++++	++++
22q11.21 duplication			3.3 (1.6–6.6)	3.7 (2.3–6.1)

Note: The figures give the odds ratios with 95% confidence intervals.
Abbreviations: CNVs, copy number variants; SZ, schizophrenia; BP, bipolar disorder; ASD, autism spectrum disorder; ID, intellectual disability; ++++, found only in cases.
Source: Data from Malhotra D, Sebat J. CNVs: harbingers of a rare variant revolution in psychiatric genetics. Cell 2012; 148: 1223–41.

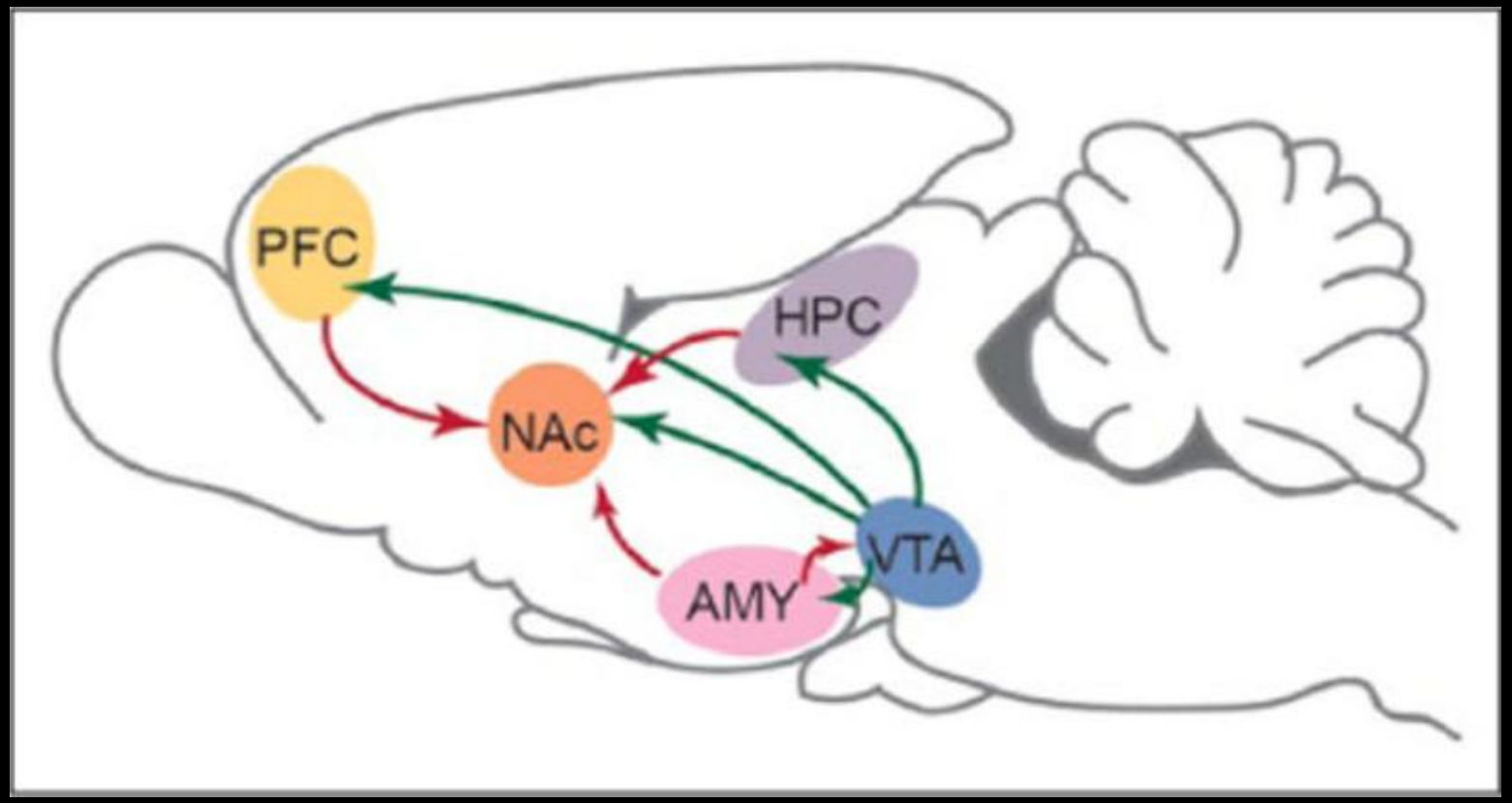
- Complex, multifactorial
- Chronic alterations in **neural circuit structure and function**, abnormalities in **glial cells**
- Relatively **high rates of discordance among identical twins** - importance of additional mechanisms
- Environmental insults - **stable changes in gene expression**, neural circuit function - ultimately behavior
- These mal-adaptations appear distinct between **developmental versus adult exposures**
- Modifications in specific brain regions
- Animal models, Postmortem patient brains

- Functional and transcriptional alterations in **several limbic brain regions** implicated in regulating **stress responses, reward, and cognition**

- Epigenetic mechanisms control the **spacing of nucleosomes** and the degree to which they are **condensed**, which thereby determines **gene activity**

- In simplified terms, chromatin exists across a continuum between an inactivated, **condensed state (heterochromatin)**, which does not allow transcription of genes, and an activated, **open state (euchromatin)**, which allows individual genes to be transcribed

Limbic Circuitry Implicated In Psychiatric Disorders



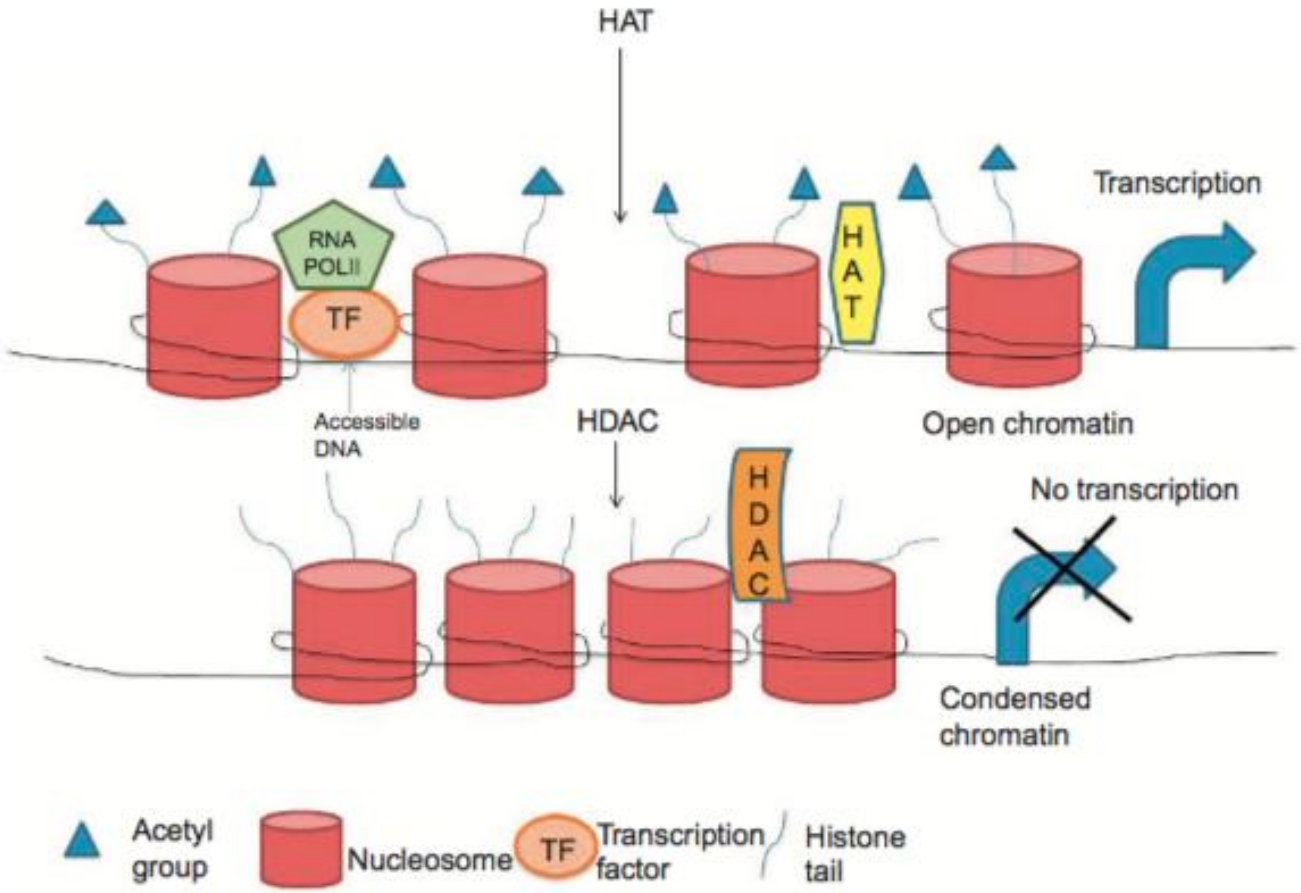
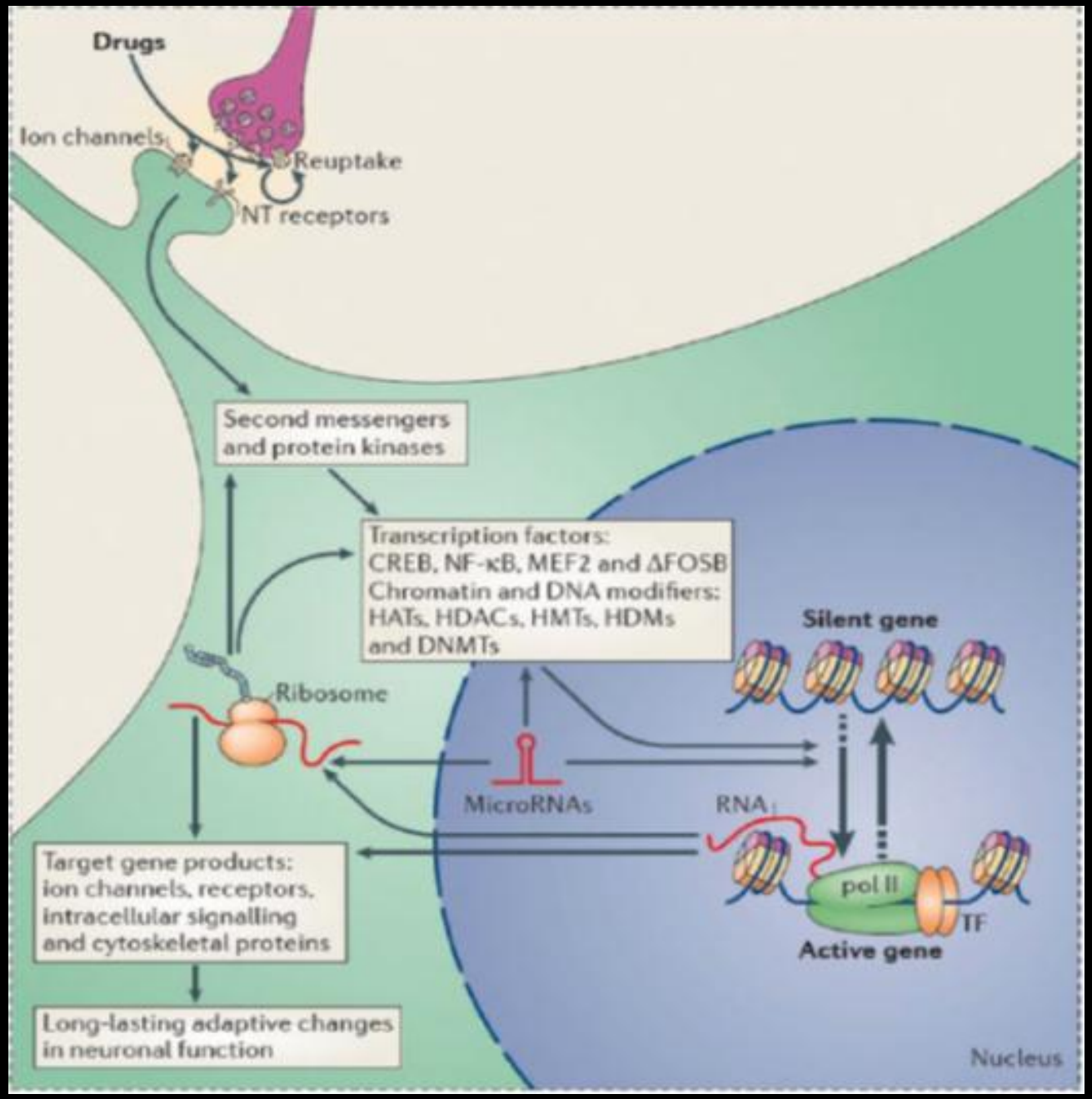
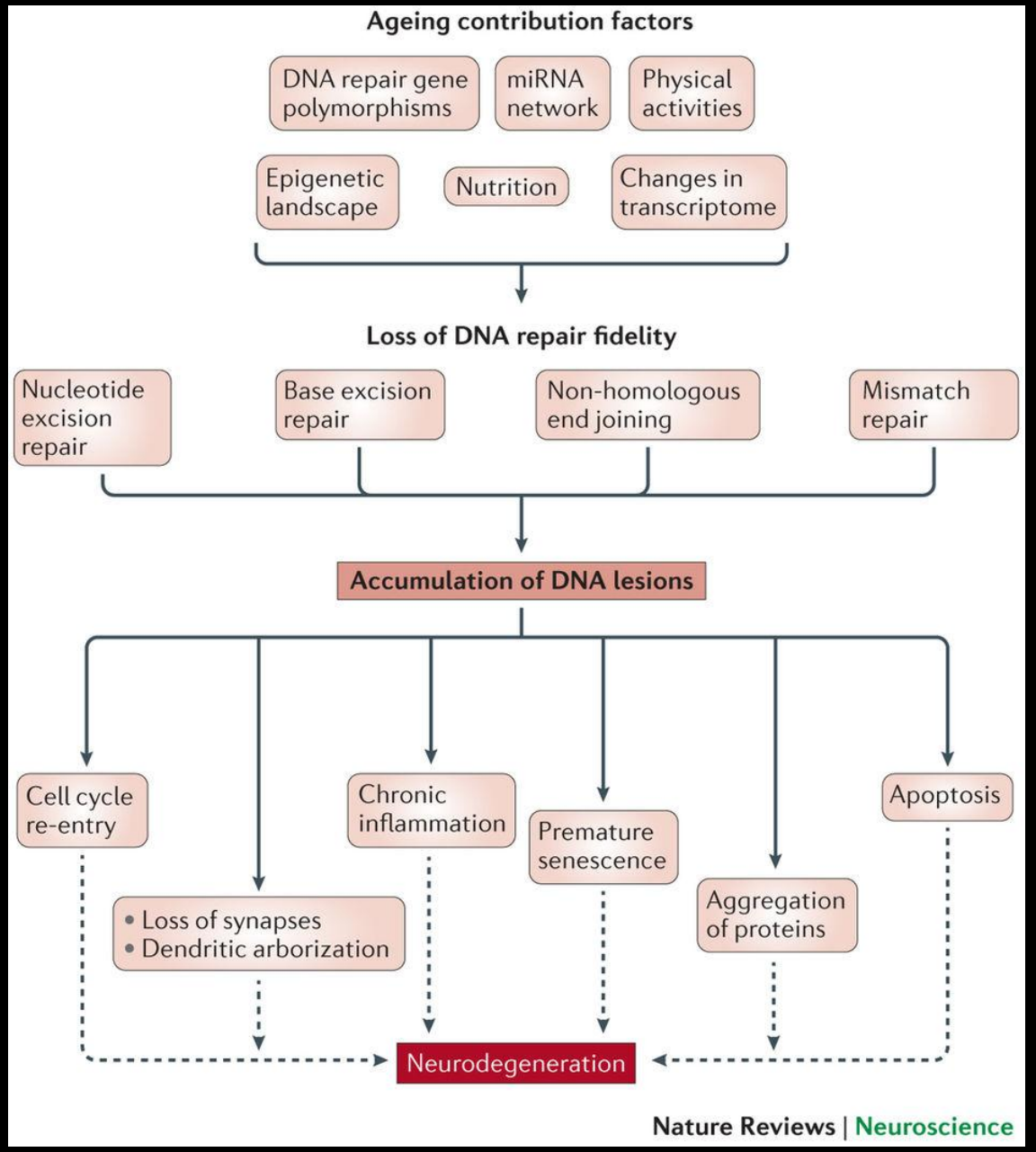


FIGURE 2.3

Acetylation of histones by histone acetyltransferase (HAT) generally leads to an open chromatin structure; thus, transcription factors and RNA polymerase can bind to DNA and activate transcription. Histone deacetylases (HDACs) deacetylate histone and lead to a closed chromatin state where the transcription factors cannot bind.


Drugs
Ion channels
Reuptake
NT receptors
Second messengers and protein kinases
Transcription factors: CREB, NF-κB, MEF2 and ΔFOSB
Chromatin and DNA modifiers: HATs, HDACs, HMTs, HDMs and DNMTs
Silent gene
Ribosome
MicroRNAs
RNA
pol II
TF
Active gene
Target gene products: ion channels, receptors, intracellular signalling and cytoskeletal proteins
Long-lasting adaptive changes in neuronal function
Nucleus


Ageing contribution factors
DNA repair gene polymorphisms
miRNA network
Physical activities
Epigenetic landscape
Nutrition
Changes in transcriptome
Loss of DNA repair fidelity
Nucleotide excision repair
Base excision repair
Non-homologous end joining
Mismatch repair
Accumulation of DNA lesions
Cell cycle re-entry
Chronic inflammation
Premature senescence
Apoptosis
• Loss of synapses
• Dendritic arborization
Aggregation of proteins
Neurodegeneration
Nature Reviews | Neuroscience

Predetermined epigenesis:

(Unidirectional structure–functional development)

genes → brain structure → brain function → experience

Probablistic epigenesis:

(Bidirectional structure–functional development)

genes ↔ brain structure ↔ brain function ↔ experience

Stress

DNA Methylation

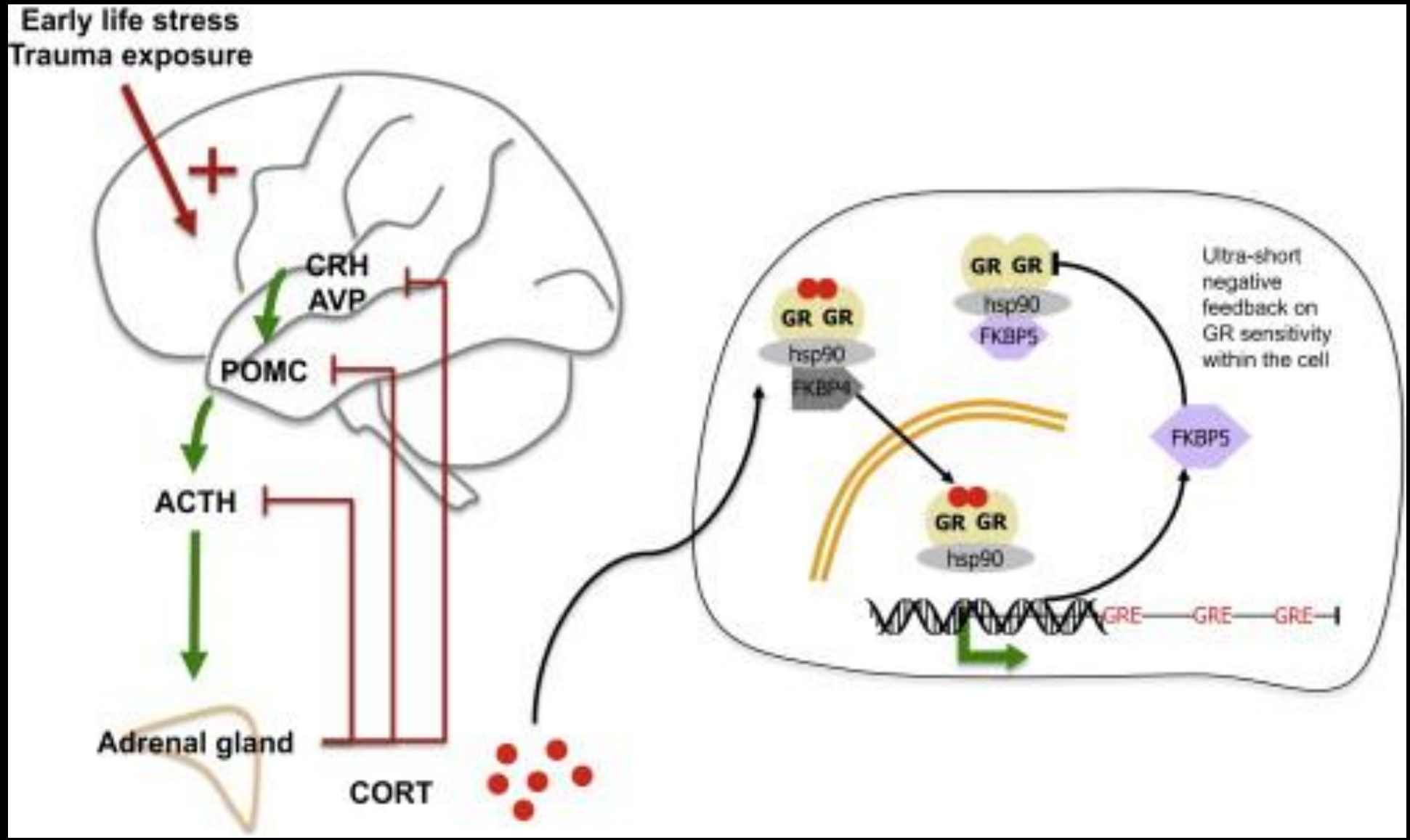
- Chronic social defeat stress **increases expression of Dnmt3a in NAc**
- Overexpressing Dnmt3a in this region increases **depression-like behavior**
- DNMT3a activity is generally associated with **transcriptional repression**
- **Expression of DNMTs is altered in limbic and brain stem regions** in depressed suicide completers
- Candidate genes studied –
  - **GDNF in NAc**
  - **CRF in Paraventricular Nucleus of Hypothalamus (PVN)** - decreased DNA methylation at the Crf promoter
  - CRF in PVN may also be associated with **sex specific HPA regulation**

DNA Methylation

- Pivotal role in **cell differentiation, imprinting, and X chromosome inactivation**
- Within gene promoters generally exerts a **repressive effect on gene transcription**
- **Prevent the association** of DNA-binding factors with their target sequence
- Bind to methyl-CpG-binding proteins to recruit transcriptional co-repressors – **Chromatin silence**
- Significant portion of DNA methylation occurs at non-CpG sites – **Activation / Suppression**

DNA Methylation

- **Stable** genetic change
- Enzymatic basis of demethylation **poorly understood**
- Most studies of DNA methylation in psychiatric disorders to date have **not distinguished between 5mC (5-methylCytosine) and 5hmC (5-hydroxy methylcytosine)**
- 5mC oxidation derivatives are expressed at highest levels in neurons, and in contrast to the generally repressive effect of 5mC on gene expression, **5hmC is more correlated with transactivation** (5mC forms can be converted to unmethylated states)


Early life stress
Trauma exposure
+
CRH
AVP
POMC
ACTH
Adrenal gland
CORT
GR GR
hsp90
FKBP5
GR GR
hsp90
FKBP4
GR GR
hsp90
FKBP5
Ultra-short negative feedback on GR sensitivity within the cell
GRE
GRE
GRE

Residue	Mark/Effect	Region	Stress induction	Age of stress	Specific genes regulated	Refs.
General H3	Ac ↑	HPC	Maternal separation	Postnatal	global	80
	Ac ↓	NAc	Chronic mild stress in stress-sensitive mice, reversed by imipramine; human depression with medication	Adult	global; *CaMKIIa*	20, 30
	Ac ↑	PFC	Social stress	Adult	global	28
	Ac ↓	HPC	Social stress, LR/HR rats	Adult	global	23, 25, 27
	Ac ↑	HPC	Social stress	Adult	*Bdnf*	13
H3K27	2Me ↑↓	NAc	Broad changes with social stress, reversed by imipramine	Adult	global	32
	2Me ↑	HPC	Social stress		*Bdnf*	13
	3Me ↑	NAc	Social stress	Adult	*Rac1*	31
	3Me ↑	PFC	Human depression/suicide		*TRKB*	40, 41
	3Me ↓	HPC	restraint stress		global	33
	3Me ↓	PFC	Human depression with medication		*BDNF*	38
H3K14	Ac ↑	AMY	Social stress	Adult	global	25
	Ac ↑↓	HPC	Temporally modulated by social stress; regulation in LR/HR rats		global	23, 25
	Ac ↑↓	NAc	Temporally modulated by social stress		global	14
H3K9	Ac ↓	HPC	Low maternal care	Postnatal	*Nr3c1, Grm1, Gad1*	17, 81-83
	1Me ↓	HPC	Restraint stress	Adult	global	33
	2Me ↓	NAc	Social stress	Adult	global	22
	2Me ↑	NAc	Fluoxetine; human depression with medication		*Camkiia*	30
	3Me ↑	HPC	Restraint stress	Adult	global; transposable elements	33, 34
H3K4	2Me ↑	HPC	Social stress and imipramine	Adult	*Bdnf*	13
	3Me ↓	HPC	Low maternal care	Postnatal	*Grm1*	81
	3Me ↓	NAc, HPC	Social stress and chronic mild stress, reversed with imipramine	Adult	global	20, 33
	3Me ↑	PFC	Human depression/suicide	Adult	*SYN1, OAZ1*	36, 37
General H2B	Ac ↓	HPC	Regulation in LR/HR rats	Adult	global	23
General H4	Ac ↑	HPC	Maternal separation	Postnatal	global, *Arc, Egr1*	80
	Ac ↓	HPC	Regulation in LR/HR rats	Adult	global	23
H4K12	Ac ↑	forebrain	Maternal separation	Postnatal	global	79

Epigenetic mark	Age of stress	Brain region/direction		Specific genes regulated
H2BAc (general)	Adult	HPC	↓	
H3Ac (general)	Postnatal Adult	HPC, PFC	↑	*Bdnf*
		PFC	↑	
		HPC, AMY	↓	
H3K9Ac	Postnatal	HPC	↓	*Nr3c1*
H3K14Ac (general)	Adult	AMY	↑	
		HPC *(transiently)*	↑	
		HPC *(chronic)*	↓	
		NAc	↓	
H3K18Ac (general)	Adult	PFC	↑	
H4Ac (general)	Postnatal	HPC	↑	
H4K12Ac (general)	Postnatal	forebrain	↑	
Hdac 1, 3, 7, 8, 10 (general)	Postnatal	forebrain, HPC	↓	
Hdac2 (general)	Adult	NAc	↓	
Hdac3 (general)	Adult	HPC	↑	
Hdac5 (general)	Adult	NAc, AMY	↓	
		NAc, *with imipramine*	↑	
		HPC, *with imipramine*	↓	
CREB Binding protein (general)	Adult	HPC	↓	
H3K4me3	Adult	PFC	↑	*SYN2, OAZ*
H3K9me1 (general)	Adult	HPC	↓	
H3K9me2	Adult	NAc	↓	*Camkiia*
		NAc, *with fluoxetine*	↑	
H3K9me3 (general)	Adult	HPC	↑	
G9a (general)	Adult	NAc	↓	
H3K27me3	Adult	NAc, HPC	↑	*Rac1 (NAc)*
		PFC	↑	*TRKB*
		PFC *(with treatment)*	↓	*BDNF*

DNA methylation

Gene	Gene region	Age of stress	Brain region or tissue/direction	
Sert (Slc6a4)	promoter	prenatal	infant cord blood	↓
Crf	promoter	prenatal adult	hypothalamus	↓
			PVN (males)	
			PVN (females)	↑
Hsd11b2	promoter/exon	prenatal	hypothalamus	↓
			placenta	↑
Gr (Nr3c1)	exon 1_7	prenatal	hypothalamus	↑
	exon 1_F	prenatal	infant cord blood	
	exon 1	postnatal	HPC	
Grm1	promoter	postnatal	HPC	↑
Gad1	promoter	postnatal	HPC	↑
Esr1	promoter/exon	postnatal	hypothalamus	↑
Avp	enhancer	postnatal	PVN	↓
Bdnf	exon IX promoter	postnatal	PFC	↓
Th	promoter	adolescent	VTA	↑
Gdnf	promoter	adult	NAc	↑
Modifier / enzyme		**Age of stress**	**Region**	
Dnmt1		prenatal	PFC, HPC	↑
		postnatal	HPC	
Dnmt3a		prenatal Adult	PFC, HPC	↑
			placenta	
			NAc	
MeCp2		prenatal	PFC, HPC	↑
		postnatal	PVN	

Table 1. Examples of studies on the role of epigenetics in social psychiatry involving the brain.

Brain tissue	Finding
Hippocampus	Early life events alter expression of many GR variants in suicide completers via effects on promoter DNA methylation
Hippocampus	Childhood adversity alters epigenetic mechanisms in promoters of many genes
Amygdala, fusiform and insula	High levels of oxytocin receptor gene methylation associated with face and emotion processing
Fusiform gyrus	DNA methylation of oxytocin receptor gene linked to many measures of human sociability
Amygdala	Lower SES in adolescence associated with increased DNA methylation of proximal promoter of SERT gene

GR: glucocorticoid receptor; SERT: serotonin transporter; SES: socioeconomic status.
Other studies on the brain on the role of epigenetics in social psychiatry are given in the text.

Table 2. Examples of studies on the role of epigenetics in social psychiatry involving peripheral tissues.

Tissue/cells	Finding
Whole blood DNA	Differential methylation of five genes in PTSD patients which correlated with abuse during childhood and TLS
Whole blood DNA	Childhood maltreatment increases methylation of GR gene
Whole blood DNA	Dynamic changes in methylation of oxytocin receptor gene after acute psychosocial stress
Lymphocytes	In PTSD patients, traumatic life events induce DNA methylation changes in GR gene promoters
Whole blood DNA	Stressful life events cause DNA methylation changes in GR gene
Whole blood DNA	In MDD patients, DNA methylation of 5HTT gene correlates with childhood trauma
Whole blood DNA	Brain function involved in processing emotional stimuli correlated with DNA methylation of 5HTT gene
Lymphocytes	GR gene methylation in normal volunteers correlates with childhood maltreatment
Leukocytes	Reduced methylation of GR gene promoter in healthy adults associated with childhood abuse

Tissue/cells	Finding
Lymphoblastoid cell lines	Unresolved loss or other trauma associated with methylation of 5HTT gene
Lymphoblastoid cell lines	Methylation of 5HTT gene affected by childhood sex abuse
Buccal cells	Severe psychosocial deprivation in childhood associated with increased methylation of promoter of CYP2E1 gene
Placental cells	Distress of mothers during pregnancy caused increased methylation of placental genes

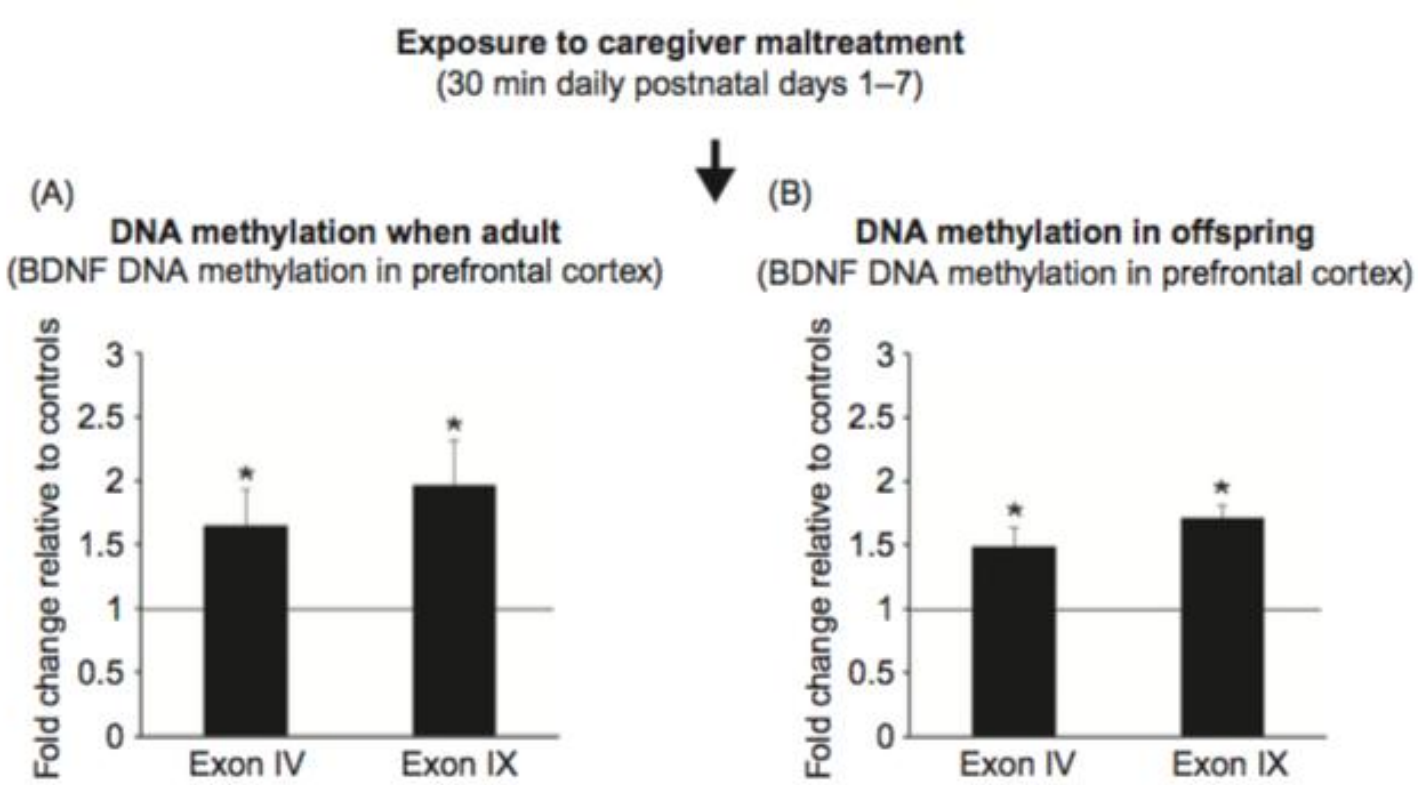
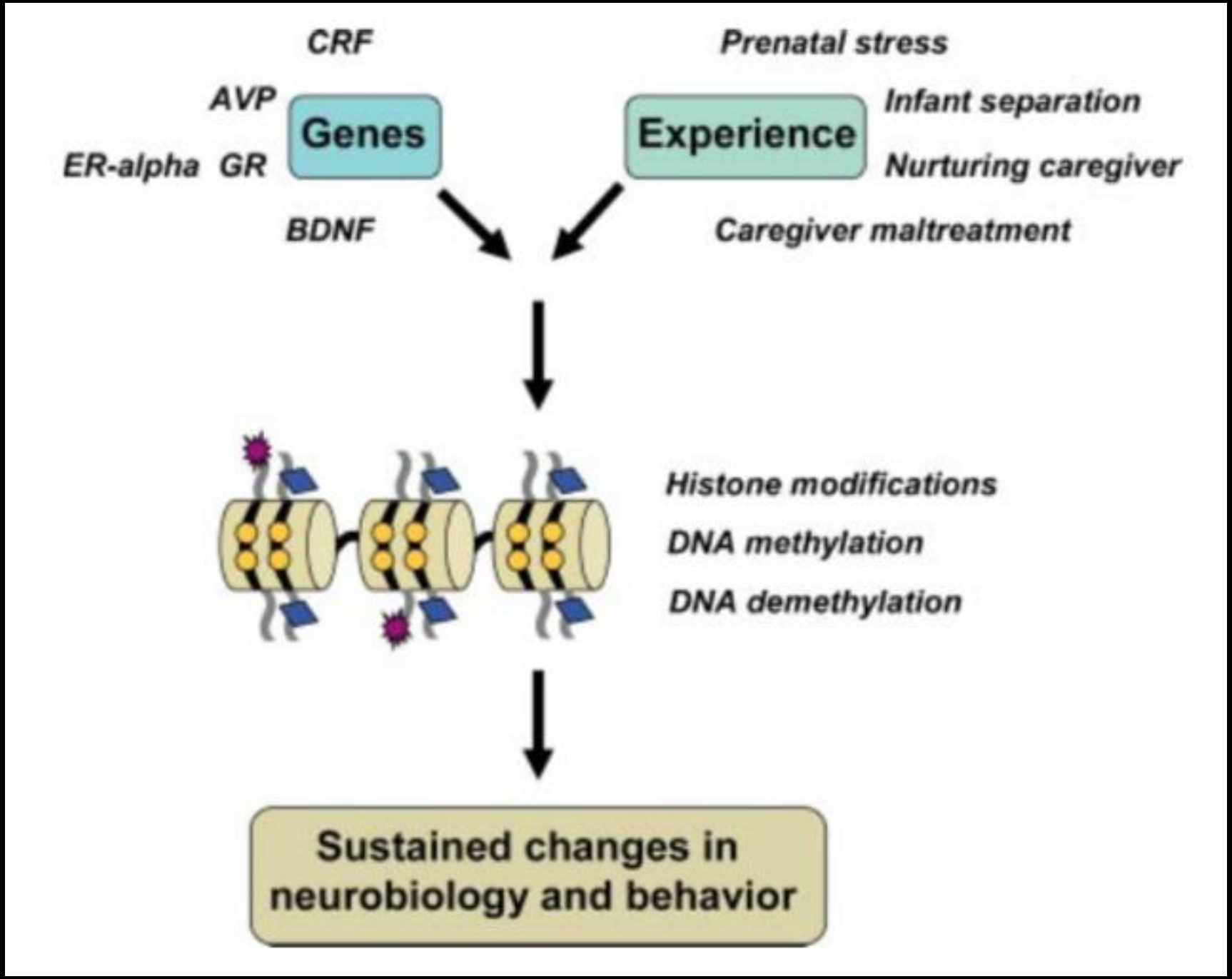


FIGURE 5.1

Epigenetic effects of early-life adversity in two generations of rats. (A) In the 2009 Roth et al. study [11], we showed that brief but repeated bouts of caregiver maltreatment during the first 7 days of life influenced adult levels of BDNF DNA methylation (increased) in the prefrontal cortex. Control groups (represented by the solid line at 1) included rats exposed to nurturing care outside of the home cage or only nurturing care from the biological mother. (B) We also took maltreated females and bred them to produce a second generation of infants. Females were observed to mistreat infants within their home cage, and both their male and female offspring likewise showed altered DNA methylation (increased on postnatal day 8).


CRF
AVP
ER-alpha GR
BDNF
Genes
Prenatal stress
Infant separation
Nurturing caregiver
Caregiver maltreatment
Experience
Histone modifications
DNA methylation
DNA demethylation
Sustained changes in neurobiology and behavior

Depression

Epigenesis In Depression

- **~40% heritable**, which emphasizes the involvement of non-genetic factors
- Animal chronic stress paradigms – **Adult models & Developmental models**
- **Adult models – Histone Acetylation**
  - **HDAC inhibition** has antidepressant effects in NAc, Hippocampus, Amygdala, or PFC, alleviates depression-like symptoms

- Genome-wide studies of NAc gene expression in defeated mice treated systemically with **fluoxetine or intra-NAc with MS275** demonstrated that both treatments **reverse a large proportion of defeat-induced differential gene expression**

- Although **each treatment also regulated subsets** of unique genes, there was also significant **overlap**, suggesting that antidepressant effects of fluoxetine may in part be mediated by affecting histone acetylation

- Precise mechanisms **unknown**. Also genome wide specific histone sites not known

Adult models – Histone Methylation

- Chronic social defeat stress **downregulates the histone methyltransferases G9a and G9a-like protein**, which catalyze H3K9me2 (a major repressive mark) in NAc

- **Overexpression of G9a in this region is antidepressant** and increased H3K9me2 at specific gene promoters is implicated in the antidepressant effect of fluoxetine

- **Reduced H3K9me2** at this gene in NAc of susceptible mice results in increased *Ras* expression, induction of ERK signaling, and, ultimately, CREB activation, which induces **depression - like behavior**

- Another repressive histone mark, **H3K27me3**, is increased upstream to the promoter of the Rac1 gene in susceptible mice and this is associated with a **sustained reduction in Rac1 expression that influences characteristic dendritic spine changes** in defeated mice

- H3K27me3 is implicated as well in the ability of chronic stress to **suppress Bdnf expression in hippocampus**

- **Stress-induced redistribution of H3K9me2 and H3K27me3** in NAc of mice subjected to chronic social defeat or protracted social isolation

- Significant and dynamic changes in repressive histone methylation were observed in upstream regulatory regions in both models, with ~20% overlap

- **H3K9me3** (another repressive histone mark) in hippocampus - dramatic induction of the mark by restraint stress at repetitive elements, non-transcribed regions of the genome - **influence genomic instability**

- **Increase in H3K9me3 after acute stress may represent an adaptive response**

- **H3K4me3 or H3K27me3 in promoter regions of several candidate genes** (e.g., OAZ, SYN2, BDNF, TRKB) in postmortem PFC

Chromatin Re-modelling In Depression

- Poorly understood
- Chronic social defeat stress **induces a repressive chromatin remodeling complex in NAc**
- Induction of the **same complex was found in NAc of depressed humans**, providing translational validation
- Induction of this repressive complex at suppressed genes correlates with **lower levels of activating histone marks (e.g., H3M4me3 and H4K16ac)** and **increased levels of certain repressive histone marks (e.g., H3K9me2)**, thus emphasizing the coordinated nature of epigenetic regulation

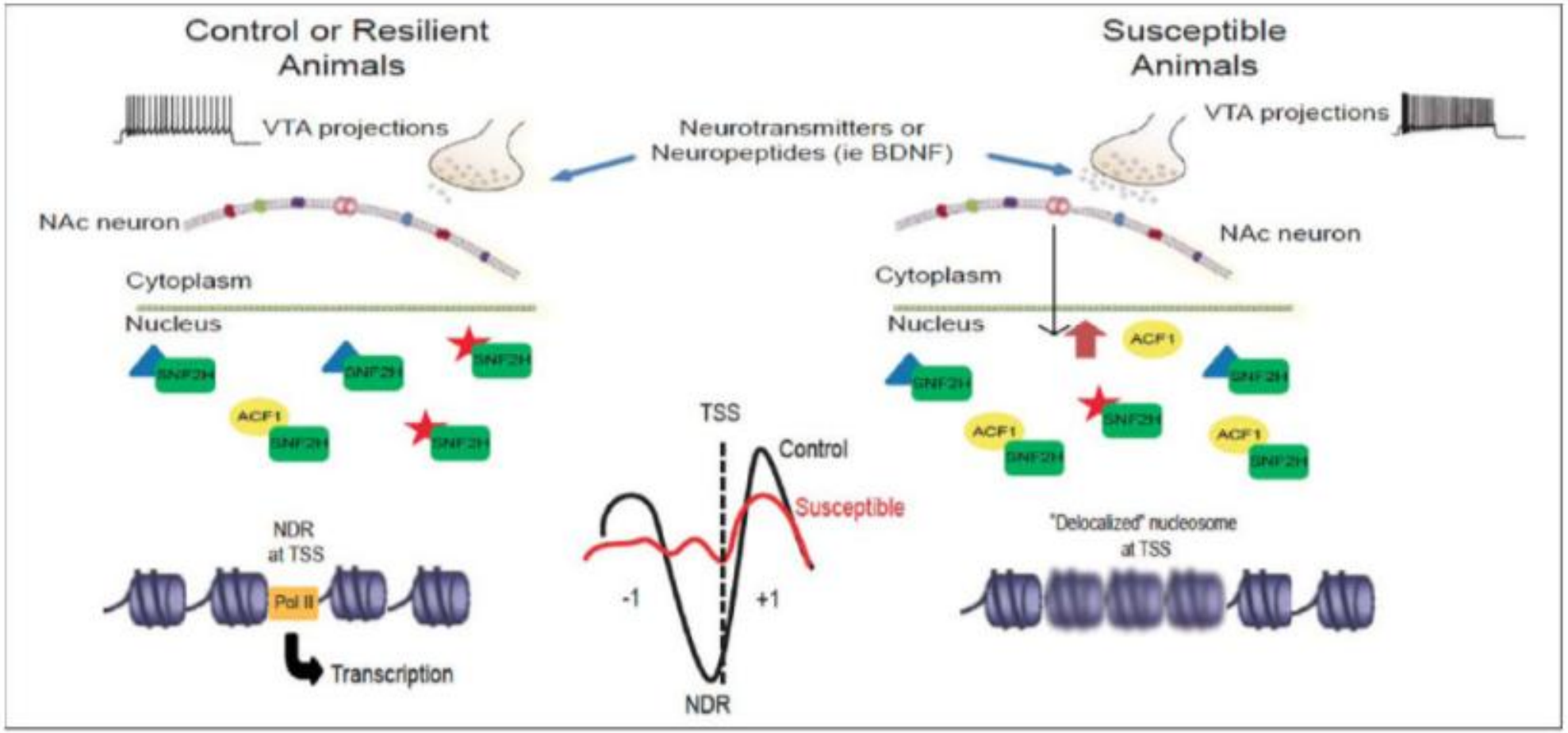


Figure 4.

Hypothesized role of chromatin remodeling ACF complex in NAc in stress susceptibility. Chronic social defeat stress CSDS, via increased burst firing of VTA neurons and BDNF release, induces ACF1 expression in NAc. The resulting upregulation of ACF complex activity, possibly through changes in TSS (transcription start site) nucleosome positioning, represses a set of genes in NAc, the reduced expression of which contributes to susceptibility. Blurry nucleosomes in the right figure represent weakly positioned or delocalized nucleosomes at TSSs. From Sun and others (2015) (permission not required).

Epigenetics – Developmental Issues In Depression

- Animal models – **Prenatal stress, Separation**
- Epigenetic changes **may last life long**
- Prenatal maternal stress –
  - **Suppresses placental expression** of 11β-hydroxysteroid dehydrogenase type 2 (11β-HSD2), which normally protects the developing fetus from maternal glucocorticoids
  - **Hypermethylation of its gene promoter** - increased stress vulnerability of offspring animals

- Elevated **DNA methylation at the NGF1-A binding region of the glucocorticoid receptor** (GR; Nr3c1) promoter exon 17 in offspring hypothalamus

- **Decreased methylation of the Crf promoter**, with no changes at Bdnf

- Mice exposed to prenatal stress had **elevated levels of Dnmt3a and Dnmt1 mRNA in PFC and hippocampus at birth**, changes that persisted into adulthood and were associated with **hypermethylation of the Reelin and Gad1 promoters, both implicated in 295**

Maternal Separation Models

- **Altered levels of expression of several HDACs in PFC and other brain regions**

- Treatment with nonselective HDAC inhibitors reverses the effects of maternal separation, while treatment with theophylline—which can activate HDACs in addition to its better described action as a phosphodiesterase inhibitor—had the opposite effect

- Early maternal separation **reduces DNA methylation in a known enhancer region for Avp expression in PVN, which is associated with increased Avp expression**

- **Alters DNA methylation and expression of Nr3c1 and Bdnf in PFC and hippocampus**, changes which could contribute to depression-like behavior observed later in life

Early Adversity

- DNA methylation is altered by extreme adversity (e.g., abuse) during early life. Maternal maltreatment of rat pups (tramping, dragging, rough handling) **leads to lasting hypomethylation at the Bdnf promoter in PFC**

- Impact of child abuse on **genome-wide DNA methylation signatures in gene promoters in hippocampus**

- DNA methylation levels in gene promoters were inversely correlated with gene expression at a genome-wide level, supporting the globally **repressive role of DNA methylation**

- Low maternal care **induces hundreds of parallel DNA methylation changes colocalized with other chromatin modifications**

- Cluster in particular at **protocadherin genes**.

- Alterations have been reported in hippocampus of suicide completers with a history of child abuse. Abused suicide completers exhibit **lower expression levels of Nr3c1**

- **Specific** to early-life adversity as Nr3c1 transcriptional modifications found in brains of depressed patients without a history of child abuse do not associate with changes in DNA methylation

- **Maternal care in rats** is reported to affect several additional genes, such as Gad1 and Grm1 in hippocampus, Esr1 in the medial preoptic area of hypothalamus - reciprocal changes in promoter methylation and in some cases with altered levels of histone acetylation or methylation

Stress beyond the early neonatal period

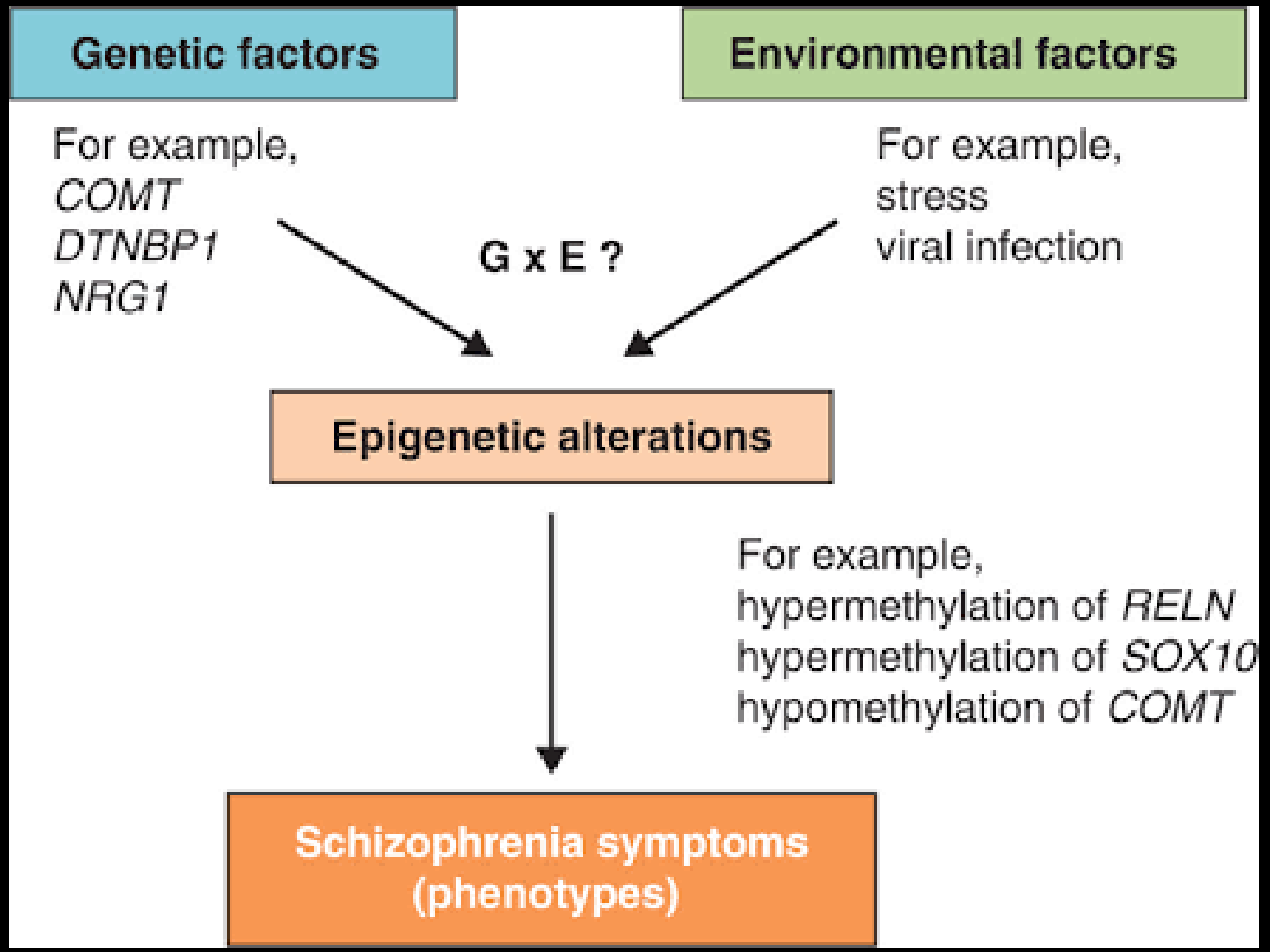
- **Hypermethylation of the tyrosine hydroxylase (Th) gene promoter** in mouse VTA

- Such hypermethylation at Th was **sustained into adulthood** and rescued by treatment with the GR antagonist RU38486

Table 1 Epigenetic modifications and MDD

Treatment	Modification	Effect
CSDS	↑ Histone methylation at *Bdnf* promoter	↓ *Bdnf* mRNA in HC
Perinatal methylmecury exposure	↑ Histone methylation at H3K27	↓ *Bdnf* mRNA in HC
CSDS	↓ pan-acH3 acetylation and ↑ methylation at H3K27 site of Rac1 promoter	↓ Rac 1 in NAc
LAC	↑ Acetylation of NF-κB	↑ mGlu2 receptor in PFC and HC
CSDS	↑ DNA methylation	↑ DNMT3a mRNA in NAc

Schizophrenia


Genetic factors
Environmental factors
For example,
COMT
DTNBP1
NRG1
G x E ?
For example,
stress
viral infection
Epigenetic alterations
For example,
hypermethylation of *RELN*
hypermethylation of *SOX10*
hypomethylation of *COMT*
Schizophrenia symptoms
(phenotypes)

Epigenetic Mechanisms Of Schizophrenia

- Characterized by **gene expression alterations in cerebral cortex** and other brain regions
- **DNA methylation** at candidate gene promoters
- **RELN, which encodes reelin, whose promoter shows increased methylation in PFC** and certain other brain regions. Reelin controls neuronal migration during development
- This hypermethylation is associated with **reduced Reln expression** and could be mediated by increased DNMT1 levels

- **DNA methylation of SOX10 in PFC** - reduced Sox10 expression and with altered expression levels of several genes associated with oligodendrocyte function – **impact on myelination**

- Polymorphisms in SOX10 are reported to **influence the age of onset of SCZ**

- **DNA methylation of HLA genes – inflammatory changes**

- Increased levels of several HMTs

- **Altered levels of H3K9K14 acetylation in PFC** - altered expression levels of the affected genes, which include **GAD1** (glutamic acid decarboxylase-1, a key enzyme for GABA synthesis), **HTR2C** (serotonin 2C receptor), and **PPM1E** (protein phosphatase 1E)

- **Robust epigenetic dysregulation of GAD1 in PFC** - including excessive levels of repressive DNA and histone methylation at the expense of certain activating histone marks such as H3K4me3

- Other SCZ risk genes - **CACNA1C** (Calcium Voltage Gated Channel Subunit Alpha Type 1C)

- Genes that show alterations in brain—for example, RELN and GAD1—are also reported to **differ in peripheral tissues**

- Abnormal methylation status has been reported in blood for several additional candidate genes, such as **BDNF** (brain-derived neurotrophic factor), **5HTR1A** (serotonin 1A receptor), and **COMT** (catechol-O-methyltransferase)

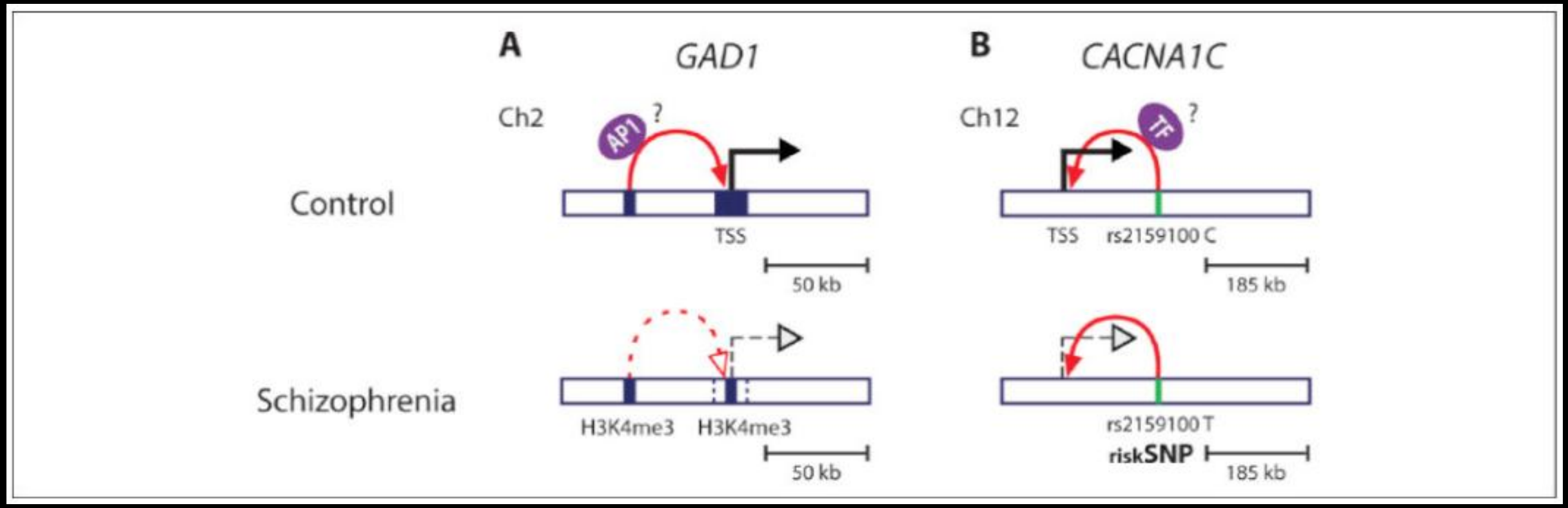
- The epigenetic status of genes is **highly cell type-dependent**
- **Genome-wide epigenetic marks not yet** available
- PsychENCODE
- Most studied parts – **PFC**
- Hot areas – **Cerebellar cortex, Subcortical areas**
- Mutations in perhaps up to **50 genes**, each encoding a different chromatin regulator, have been linked to a wide range of neurodevelopmental syndromes, including rare monogenic forms of SCZ

- **Chromatin defects** in brain were traditionally **considered static lesions** of early development – now also implicated in **adult-onset neurodegenerative disease**

- **Gene duplication** of the HMTs, KMT1D and KMT2F, or the MYTL1 and ZNF804A transcription factors, have been linked to some cases of SCZ

- The biggest difficulty – Lack of adequate animal models, unlike the wide variety of animal models used to study depression

- Epigenetic factors & **miRNA**


A
GAD1
Ch2
AP1
?
Control
TSS
50 kb
Schizophrenia
H3K4me3
H3K4me3
50 kb
B
CACNA1C
Ch12
TF
?
TSS
rs2159100 C
185 kb
rs2159100 T
riskSNP
185 kb

Higher order chromatin structure and schizophrenia. The role of higher order chromatin structure in transcriptional regulation of SCZ relevant genes has been shown for *GAD1* (A) and *CACNA1C* (B). (A) *GAD1*, encoding GABA synthesis enzyme, is located on chromosome 2 (Ch2) and frequently down-regulated in cerebral cortex of SCZ patients (dashed black arrow) and this is associated with lower levels of active histone marks, including H3K4me3 (blue square) at the *GAD1* transcription start site (TSS). The TSS region of *GAD1* has been shown to physically interact with an AP1 motif-enriched enhancer region located 50 kb further upstream (also enriched in H3K4me3 mark, blue square). Evidence has been presented that a chromatin loop (red arrow), that may carry a cargo such as AP1 transcription factors (purple oval) into close proximity to the core promoter region facilitating *GAD1* gene transcription, is weakened in brains of SCZ patients brain (dashed red arrow). This could contribute to lower *GAD1* expression. (B) Several single nucleotide polymorphisms (SNPs) residing in noncoding regions of the *CACNA1C* gene on chromosome 12 (Ch12) have been associated with lower *CACNA1C* expression and SCZ risk. The rs215100 T SCZ risk allele (green bar) resides in an intronic enhancer region, 185 kb downstream from the *CACNA1C* TSS, which has been shown to physically interact with the *CACNA1C* TSS (solid red arrow). The T allele confers lower transcriptional activity (dashed black arrow) as compared to C allele (solid black arrow), presumably by affecting the binding of transcription factors (TF, purple oval) and their interaction via chromosomal loops with the promoter *CACNA1C* region.

Table 3 Epigenetic modifications and schizophrenia

Tissue/Species	Treatment	Effect
Postmortem brain tissue	Patients diagnosed with SZ	↑ Methylation at reelin promoter
Human cell line	aza-2′-deoxycytidine	↑ Reelin MRNA
Reelin mice	Chronic L-methionine administration	↓ Reelin via hypermethylation of reelin promoter
N2T cells	MS-275	↓ Methylation at GAD67 promoter and ↑ GAD67 expression
Postmortem brain tissue	Patients diagnosed with SZ (some on antipsychotics)	↑ DNMT1, 3a mRNA in cortical GABA neurons
Mice	Chronic treatment with clozapine	↓ mGlu2 receptor expression via acetylation at the promoter site
Mice	Chronic treatment with SAHA in PFC	↑ mGlu2, mGlu3 mRNA in PFC via H3 acetylation at respective promoters

ASD

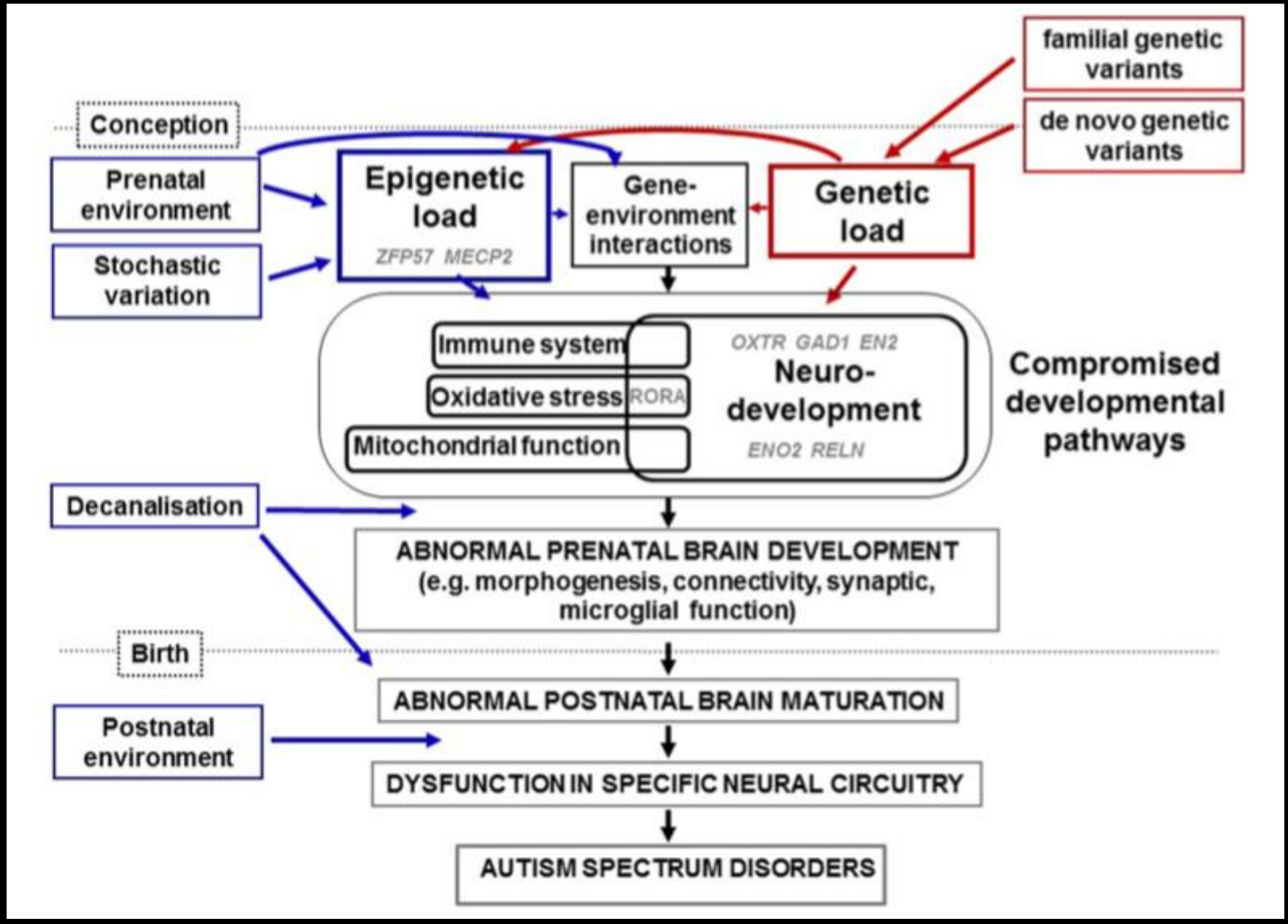

familial genetic variants
de novo genetic variants
Conception
Prenatal environment
Stochastic variation
Epigenetic load
ZFP57 MECP2
Gene-environment interactions
Genetic load
Immune system
Oxidative stress
Mitochondrial function
OXTR GAD1 EN2
Neuro-development
RORA
ENO2 RELN
Compromised developmental pathways
Decanalisation
ABNORMAL PRENATAL BRAIN DEVELOPMENT
(e.g. morphogenesis, connectivity, synaptic, microglial function)
Birth
ABNORMAL POSTNATAL BRAIN MATURATION
Postnatal environment
DYSFUNCTION IN SPECIFIC NEURAL CIRCUITRY
AUTISM SPECTRUM DISORDERS

TABLE 1 | Genes for which there is high confidence of association with ASD from genetic evidence (https://gene.sfari.org/ (13)).

Abbreviation	Name	Protein function
ADNP	Activity-dependent neuroprotector homeobox	Vasoactive intestinal peptide, neuroprotective factor, transcription factor (E)
ANK2	Ankyrin 2, neuronal	Cytoskeletal and cell membrane protein
ARID1B	AT rich interactive domain 1B (SWI1-like)	ATP-dependent chromatin remodeller (E)
ASH1L	Ash1 (absent, small, or homeotic)-like (Drosophila)	Transcriptional activator, cell-cell tight junctions (E)
ASXL3	Additional sex combs like 3 (Drosophila)	Possible regulator of transcription (E)
CHD8	Chromodomain helicase DNA binding protein 8	Transcriptional repressor involved in early development (E)
DYRK1A	Dual-specificity tyrosine-(Y)-phosphorylation regulated kinase 1A	Protein kinase involved in signaling and early development
GRIN2B	Glutamate receptor, inotropic, *N*-methyl D-aspartate 2B	Glutamate receptor involved in long-term potentiation and synaptic transmission
POGZ	Pogo transposable element with ZNF domain	Possible transposase and transcription factor (E)
PTEN	Phosphatase and tensin homolog (mutated in multiple advanced cancers 1)	Tumor suppressor involved in signaling and mitochondrial function
SCN2A	Sodium channel, voltage-gated, type II, alpha subunit	Sodium channel expressed in the brain
SETD5	SET domain containing 5	Likely chromatin protein (E)
SHANK3	SH3 and multiple ankyrin repeat domains 3	Postsynaptic density synapse scaffold protein
SUV420H1	Suppressor of variegation 4-20 homolog 1 (Drosophila)	Likely chromatin protein (E)
SYNGAP1	Synaptic Ras GTPase activating protein 1	Postsynaptic density synapse protein
TBR1	T-box, brain, 1	Likely transcription factor associated with early cortical development (E)

Pathways identified
Neurodevelopment
Immune and inflammatory response (mediated by NK cells), cytotoxicity
Cell communication, immune and inflammatory response
Steroid hormone metabolism
Steroid hormone metabolism, circadian rhythm
immune and inflammatory response (mediated by NK cells)
Neurodevelopment, synaptic function (long-term potentiation)
Synaptic function
Immune and inflammatory response
Synaptic function, immune and inflammatory response
Microglial function, immune response, neuronal activity
Neurodevelopment; signaling; skeletal development
Ribosome function, spliceosome function, mitochondrial, immune and inflammatory response, calcium signaling
Neurotrophic signaling, notch signaling; synaptic function (long-term potentiation)
Immune and inflammatory response; hemoglobin metabolism
Neurodevelopment, skeletal development, gastrointestinal development, steroid hormone metabolism, circadian rhythm

TABLE 3 | Summary of evidence for potential ASD-specific methylation biomarkers.

Gene	Genetic evidence	Methylation reference	Diagnostic method	Tissue	Samples[a]	Largest effect size[b]	Expression	Protein	Other data
OXTR	Weak	(95)	DSM-IV, ADI-R	PBLs	20/20 10/10[c]	+23% +38.9%	No	No	Endophenotype[d]
				Temporal cortex	10/10[c]	+41.6%[c]	yes	No	
GAD1	Weak	(113)	Not given	Cerebellum	10/10	+3%[e]	Yes[f]	No	Animal models, MECP2 binding
EN2	Minimal	(120)	DSM-IV	Cerebral cortex	13/13	+10–20%[g]	Yes	Yes	
RELN	Strong	(113)	Not given	Cerebellum	10/10	Not quantifiable	Yes	Yes	MECP2 binding
MECP2	Syndromic	(127, 128)	ADI-R, ADOS	Frontal cortex	14/14	+12%, +10%[h]	Yes	Yes	Animal model

For abbreviations and references see the main text.

[a] *Cases/controls.*

[b] *% methylation difference, cases minus controls.*

[c] *Males only.*

[d] *Methylation correlated to endophenotype.*

[e] *Hydoxymethylation only.*

[f] *In Purkinje neurons and cerebellum.*

[g] *Methylation and hydroxymethylation.*

[h] *Two separate sets of MECP2 CpG sites regions measured.*

TABLE 4 | Summary of genome-wide studies of methylation in ASD.

Reference	Samples[a]	Tissue	Participant age (years)	Diagnostic method	Method of analysis	DMR/DMP analysis[b]	Effect size cut off[c]	Adjustment for multiple testing	Validation[d]	Expression data[e]
(131)	9/9[f]	Occipital cortex Cerebellar hemispheric cortex	1–60	DSM-IV, ADOS, and/or ADI-R	HM27	DMP	No	Yes	No[g]	Yes
(132)	12/21 16/21 13/21	Prefrontal cortex Temporal cortex Cerebellum	17–35 21–40 14–17	ADI-R and/or ADOS	HM450	DMR	No	Yes	No Yes[h] No[i]	No
(133)	11/11 12/12	Anterior cingulate gyrus Prefrontal cortex	16–51	ADI-R	HM450	DMP	>5% difference	Yes	Yes	Yes
(134)	3[f], 10	LCL	2–19	ADI-R	MIRA	DMR	No	Yes	Yes	Yes
(135)	5/5	PBLs	6–12	DSM-IV, MINI instrument	MeDIP	DMR	>1.5-fold change	No	Yes	Yes
(136)	6[j] 16/22 6/10[k] 50[l, m]	PBLs	15	CAST	HM27	DMP	No	No	No Yes No No	No
(130)	47/48	Buccals	1–28	Not stated	HM450	DMR	No	Unclear[n]	Yes	No

Bipolar Disorders

Bipolar Disorders

- **Considerable overlap** with SCZ

- Genome-wide and candidate gene DNA methylation mapping in postmortem brains of individuals with SCZ or bipolar disorder with psychosis revealed **a similar degree of subtle (but significant) changes at many gene promoters**

- **HLA9**, which showed aberrant DNA methylation patterns in multiple postmortem brain cohorts and in peripheral blood and, surprisingly, also in sperm of subjects diagnosed with bipolar disorder

- **GAD1**

- **Regulators of H3K4 methylation -** one of the strongest links to the genetic risk architecture of bipolar disorder

Addiction

Treatment	Modification	Effect
Chronic cocaine exposure	↑ H3 acetylation	↑ Induction of BDNF at promoter region
Acute cocaine exposure	↑ H4 acetyation	↓ *fosB*
Co-administration of sodium butyrate and cocaine	↑ H3 acetylation	↑ cFos mRNA in striatum
Chronic infusion of MS-275	↑ global H3 acetylation	Blocks cocaine induced locomotor sensitization

PTSD

Kertes et al. Clinical Epigenetics (2017) 9:68
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Clinical Epigenetics

RESEARCH Open Access

BNDF methylation in mothers and newborns is associated with maternal exposure to war trauma

Darlene A. Kertes[1*], Samarth S. Bhatt[2], Hayley S. Kamin[2], David A. Hughes[3], Nicole C. Rodney[4] and Connie J. Mulligan[5]

Abstract

Background: The *BDNF* gene codes for brain-derived neurotrophic factor, a growth factor involved in neural development, cell differentiation, and synaptic plasticity. Present in both the brain and periphery, BDNF plays critical roles throughout the body and is essential for placental and fetal development. Rodent studies show that early life stress, including prenatal stress, broadly alters *BDNF* methylation, with presumed changes in gene expression. No studies have assessed prenatal exposure to maternal traumatic stress and *BDNF* methylation in humans. This study examined associations of prenatal exposure to maternal stress and *BDNF* methylation at CpG sites across the *BDNF* gene.

Results: Among 24 mothers and newborns in the eastern Democratic Republic of Congo, a region with extreme conflict and violence to women, maternal experiences of war trauma and chronic stress were associated with *BDNF* methylation in umbilical cord blood, placental tissue, and maternal venous blood. Associations of maternal stress and *BDNF* methylation showed high tissue specificity. The majority of significant associations were observed in putative transcription factor binding regions.

Conclusions: This is the first study in humans to examine *BDNF* methylation in relation to prenatal exposure to maternal stress in three tissues simultaneously and the first in any mammalian species to report associations of prenatal stress and *BDNF* methylation in placental tissue. The findings add to the growing body of evidence highlighting the importance of considering epigenetic effects when examining the impacts of trauma and stress, not only for adults but also for offspring exposed via effects transmitted before birth.

Keywords: Brain-derived neurotrophic factor, *BDNF*, Stress, Trauma, War, Prenatal, DNA methylation, Transcription factor, Blood, Placenta

Other Disorders

REVIEW

The Plasticity of Development: How Knowledge of Epigenetics May Advance Understanding of Eating Disorders

Michael Strober, PhD, ABPP[1,2]*
Tara Peris, PhD[1]
Howard Steiger, PhD[3,4]

ABSTRACT

Objective: To depict the processes through which animals and human beings engage their environment in continuously evolving states of conflict and cooperation.

Method: Descriptive literature review.

Results: Life history outcomes are more relative than they are absolute. Genetic variations play a crucial role, but heavily influencing behavioral outcomes, psychopathology included, are external cues that epigenetically remodel DNA along experience-dependent signaling pathways. The result is phenotypes that either optimize adjustment, or constrain it.

Discussion: Knowledge of epigenetic mechanisms may help shed new light on the origin of maturational phenotypes underlying eating disorders and why adjusting treatments to these realities warrants our attention.

Keywords: development; genes; environment; epigenetics

(Int J Eat Disord 2014; 47:696–704)

Concluding Comments

Future & Questions

- **Genome-wide assays** for numerous chromatin mechanisms
- **Animal models** & paradigms
- DNA methylation—both 5mC and **5hmC**
- **Noncoding RNAs & microRNAs**
- **Translation into transcriptional change**, not only steady state alterations in expression but also **altered inducibility** in response to a subsequent challenge
- **No single modification** examined to date is deterministic for a change in gene activity
- Modifications that are most clearly associated with a functional change are associated with **no change or even opposite changes** in transcription at many genes

- **Methodologies** innovations
- **To relate chromatin modifications** to a host of transcription factors
- **3D organization** of the genome in neurons and glia
- Exploration of **regulatory DNA elements** in the context of chromosomal loopings and higher order chromatin
- **Intergenic and intronic DNA**, **chromosome conformation capture assays**, and other techniques that measure interaction and spatial proximity of noncontiguous DNA elements in brain cells

- **Difficulty in relating chromatin modifications** at a given gene to a functional outcome

- To overcome this limitation is to use **engineered zinc finger proteins (ZFPs), sequence-specific transcription activator-like effectors (TALEs), or CAS9/CRISPR**, coupled to an enzymatic moiety, to target a particular chromatin modification to a given gene of interest within a region of adult brain

- Virtually **no mention of sex differences** in epigenetic regulation

References

Epigenetics in Psychiatry

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Epigenetic Basis of Mental Illness

Eric J. Nestler[1], **Catherine J. Peña**[1], **Marija Kundakovic**[1], **Amanda Mitchell**[1], and **Schahram Akbarian**[1]

Neurotherapeutics (2013) 10:734–741
DOI 10.1007/s13311-013-0213-6

REVIEW

Epigenetics and Psychiatry

Melissa Mahgoub · Lisa M. Monteggia

State of the art

Epigenetic signaling in psychiatric disorders: stress and depression

Rosemary C. Bagot, PhD; Benoit Labonté, PhD; Catherine J. Peña, PhD; Eric J. Nestler, MD, PhD

Dialogues in Clinical Neuroscience - Vol 16 · No. 3 · 2014

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Epigenetics of Neurobiology and Behavior during Development and Adulthood

ABSTRACT: *Gene–environment interactions have long been recognized for their important role in mediating the development and functions of the central nervous system (CNS). The study of DNA methylation and histone modifications, biochemical processes collectively referred to as epigenetic mechanisms, is helping to elucidate how gene–environmental interactions alter neurobiology and behavior over the course of the lifespan. In this review, landmark and recent studies that highlight the role of epigenetic mechanisms in the sustained effects of early-life experiences on gene activity and behavioral outcome will be discussed. Likewise, studies that implicate epigenetics in CNS and behavioral plasticity in the adult animal will be discussed. As our current understanding of epigenetics in these capacities is still evolving, epigenetic research will continue to be of considerable interest for understanding the molecular mechanisms mediating neurobiology and behavior both within and outside of sensitive periods of development.* © *2012 Wiley Periodicals, Inc.* Dev Psychobiol

Keywords: *early-life experience; stress; memory; epigenetic; DNA methylation; histone modification*

Prog Biophys Mol Biol. 2015 July ; 118(0): 1–7. doi:10.1016/j.pbiomolbio.2015.04.008.

Epigenetic Mechanisms in Schizophrenia

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Lamarck rises from his grave: parental environment-induced epigenetic inheritance in model organisms and humans

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Review Article

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The role of epigenetics in social psychiatry

Jacob Peedicayil

frontiers in Neurology

REVIEW
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The role of epigenetic change in autism spectrum disorders

Yuk Jing Loke[1], *Anthony John Hannan*[2] *and Jeffrey Mark Craig*[1]*

Kertes et al. Clinical Epigenetics (2017) 9:68
DOI 10.1186/s13148-017-0367-x

Clinical Epigenetics

RESEARCH Open Access

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REVIEW

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Thank You