Maternal care and Gene - Environment Interactions Defining Development

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The development of an individual is an active process of *adaptation that occurs within a social and economic context*:

- To resource (food, shelter, safety) availability.
- To social interactions.
- To independence from the parent.

Developmental Origins of Adult Disease

Early experience

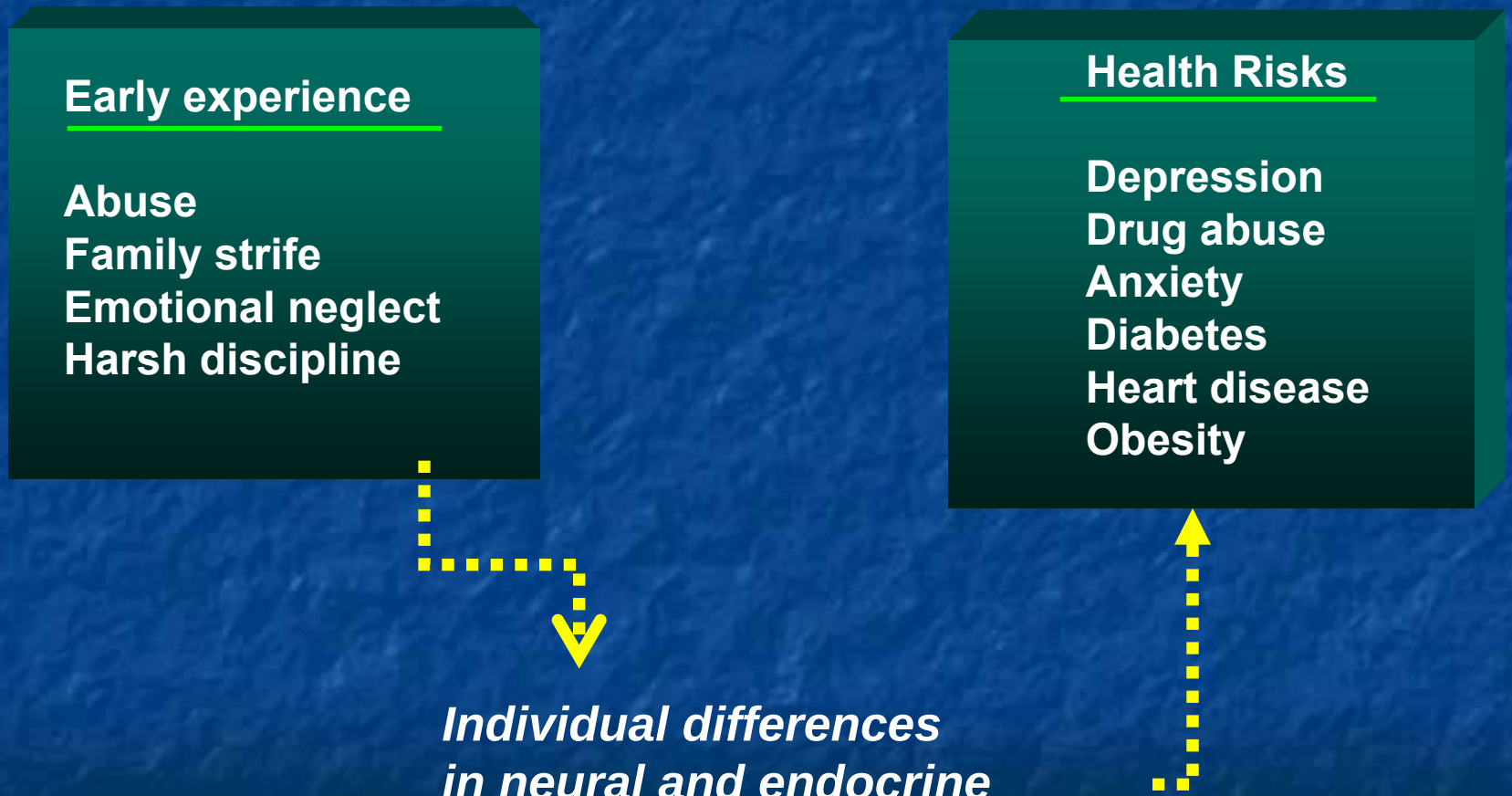
Abuse
Family strife
Emotional neglect
Harsh discipline

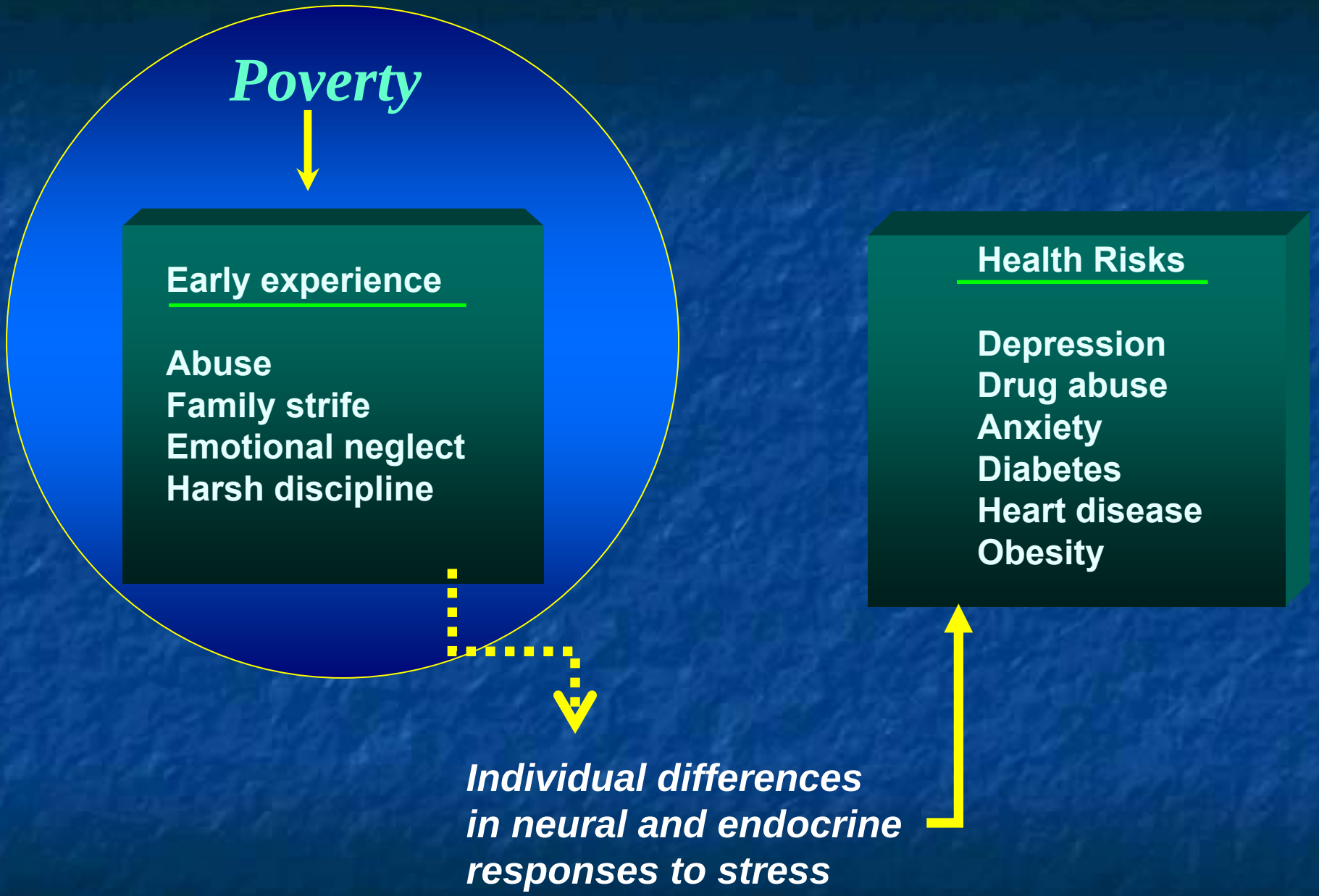
Health Risks

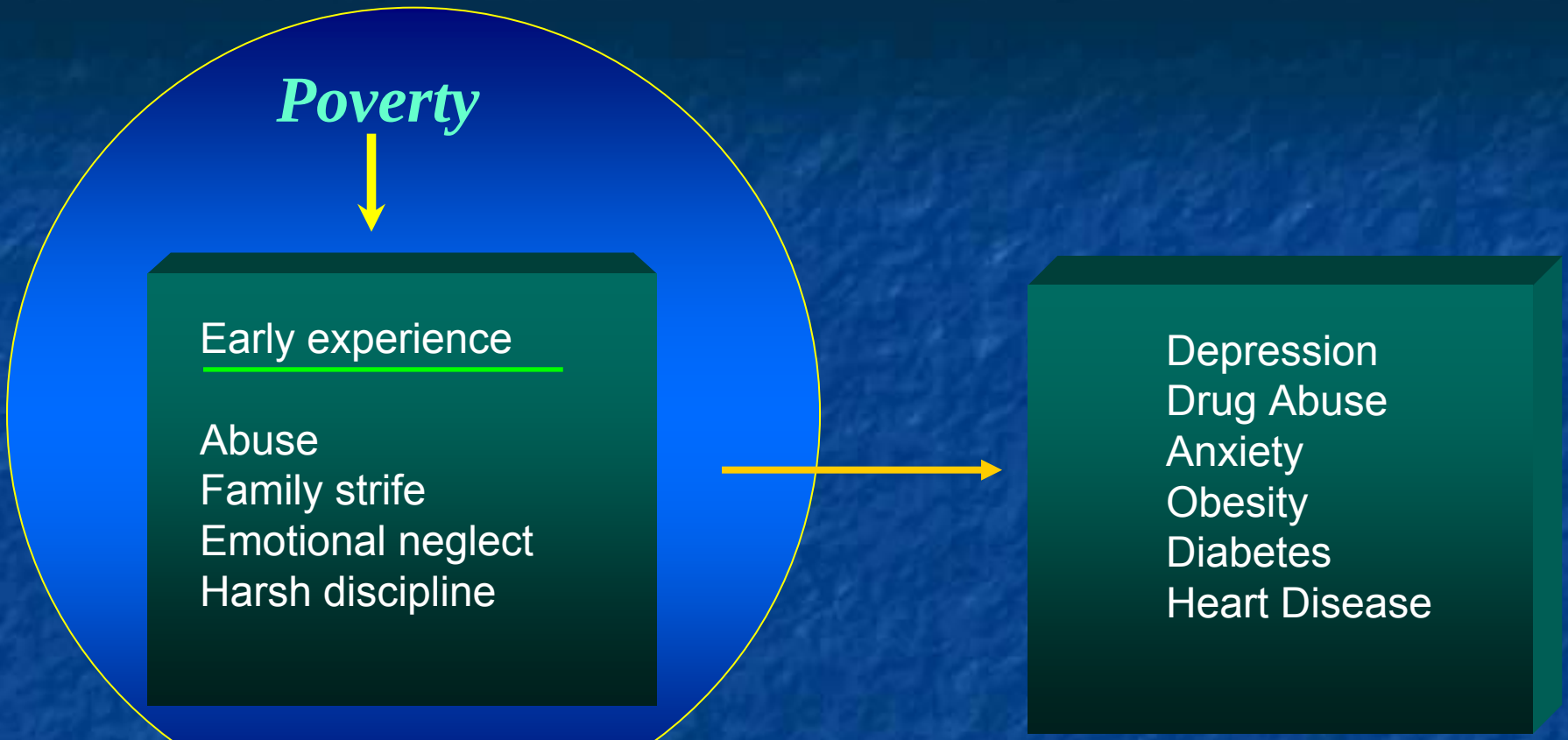
Depression
Drug abuse
Anxiety
Diabetes
Heart disease
Obesity

Mechanism?

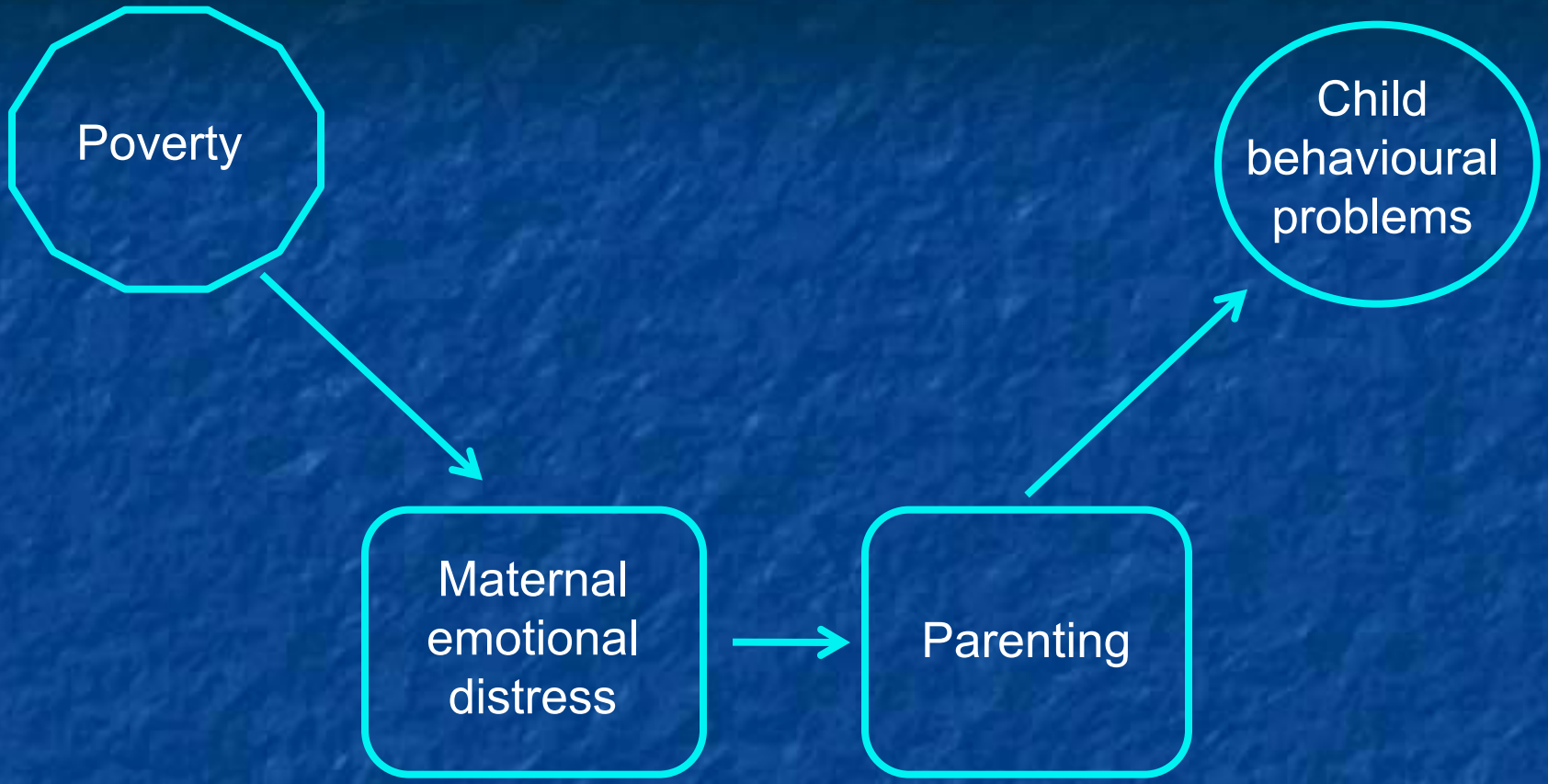
Stress Diathesis Models



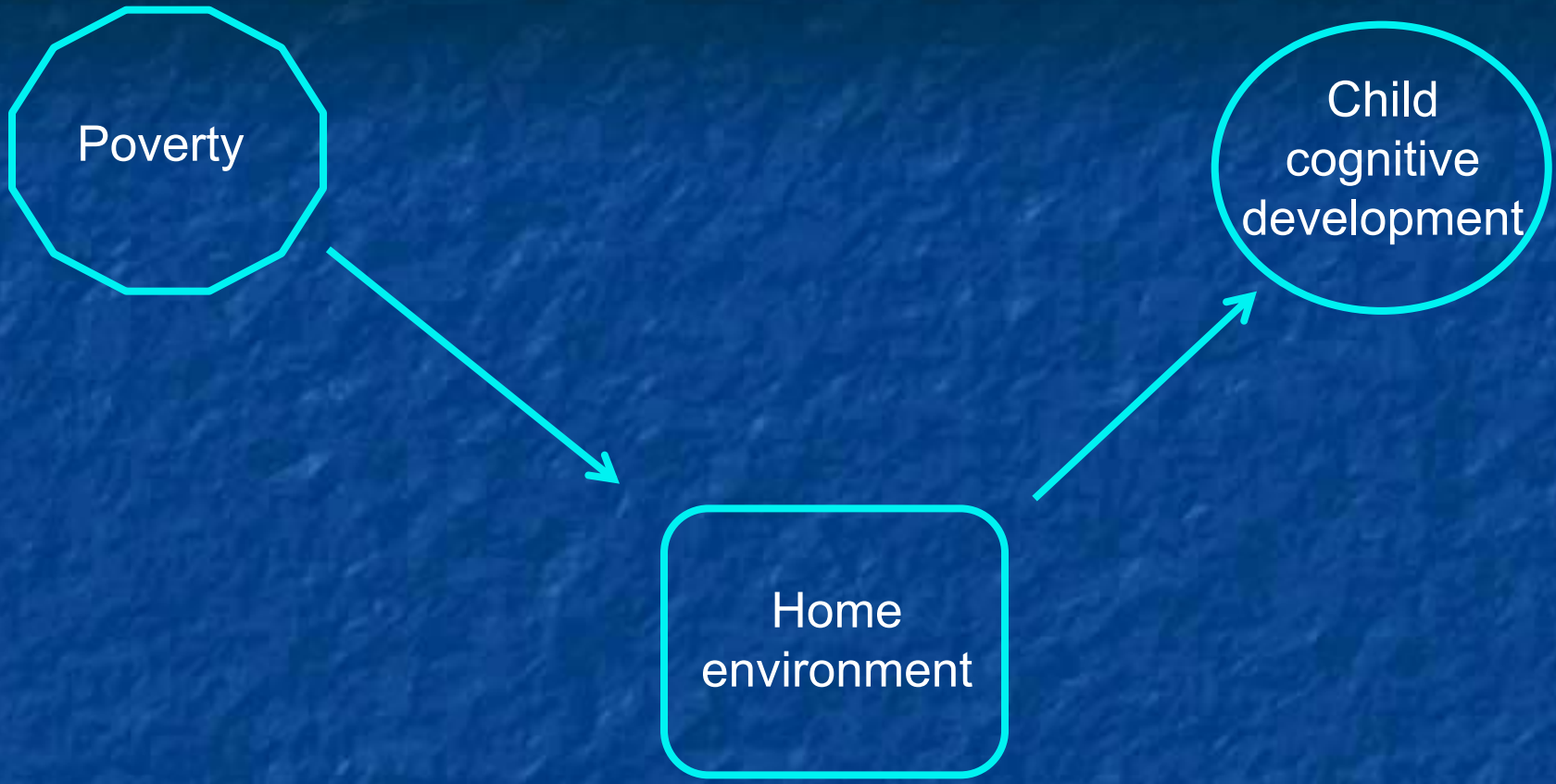

Poverty
Early experience
Abuse
Family strife
Emotional neglect
Harsh discipline
Health Risks
Depression
Drug abuse
Anxiety
Diabetes
Heart disease
Obesity
Individual differences
in neural and endocrine
responses to stress

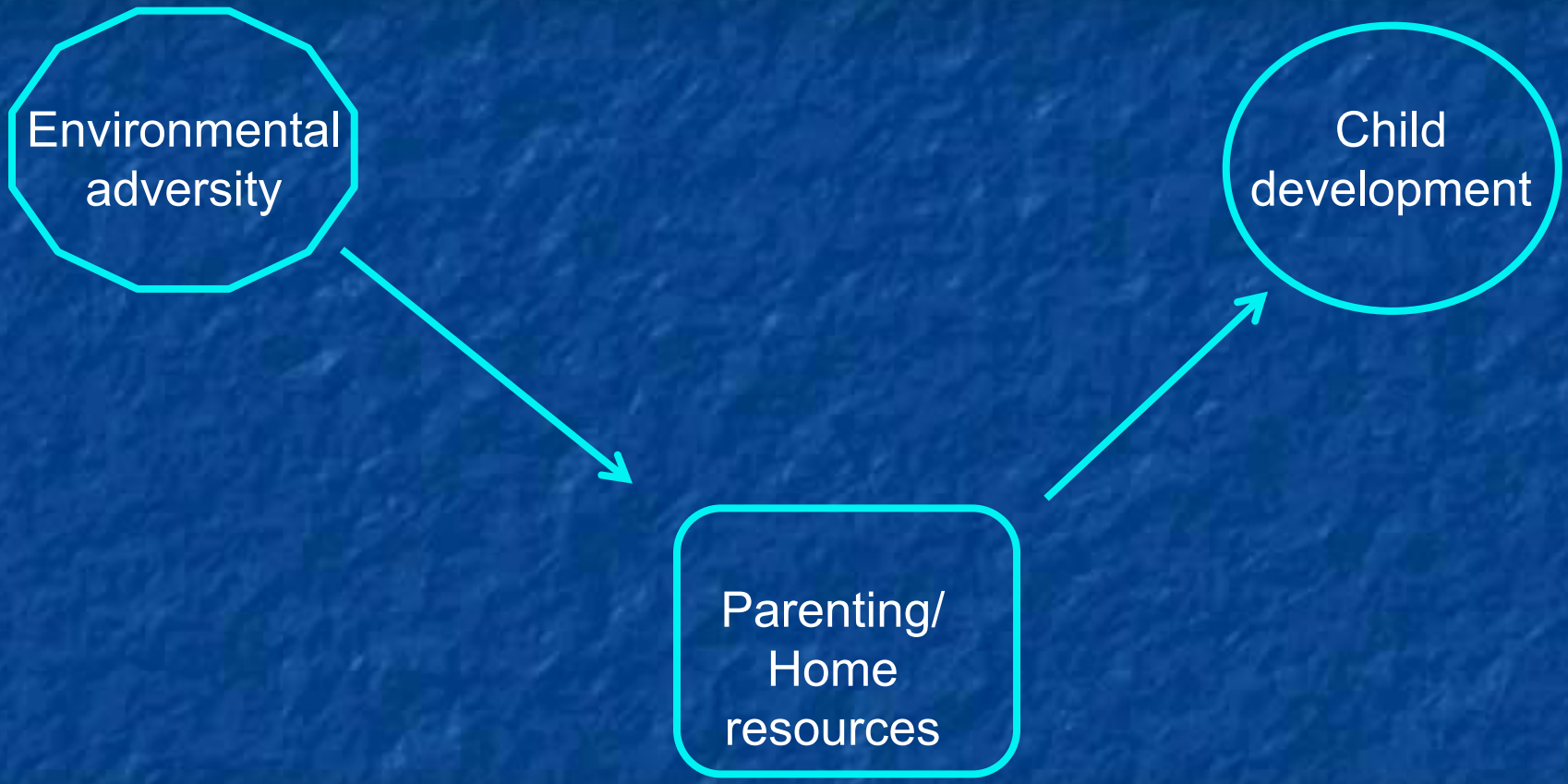


Effects of poverty on emotional and cognitive development are mediated by parental factors (Conger, McLloyd, Eisenberg).

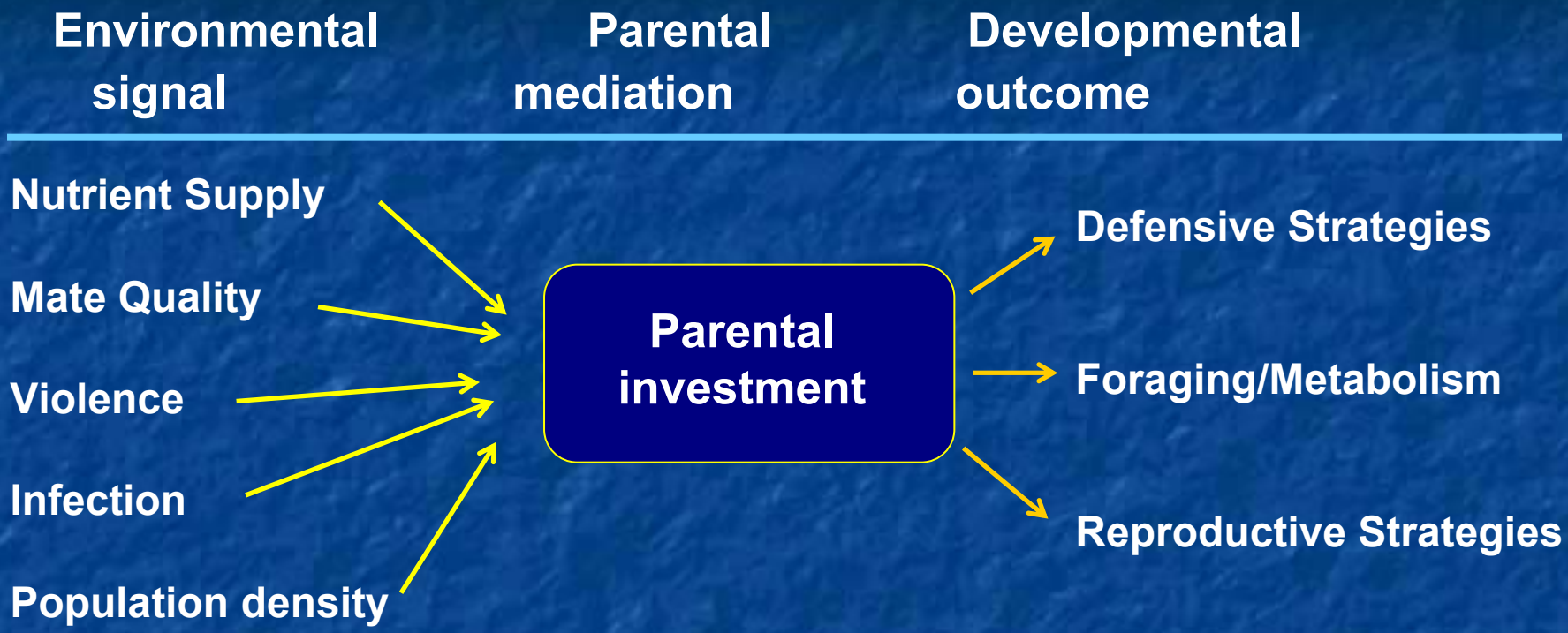


Linver, Brooks-Gunn & Kohen Dev Psychol 2002; same model predicts child cortisol levels (Lupien, McEwen, Meaney Biol Psychiatry 2000)


Poverty
Home environment
Child cognitive development

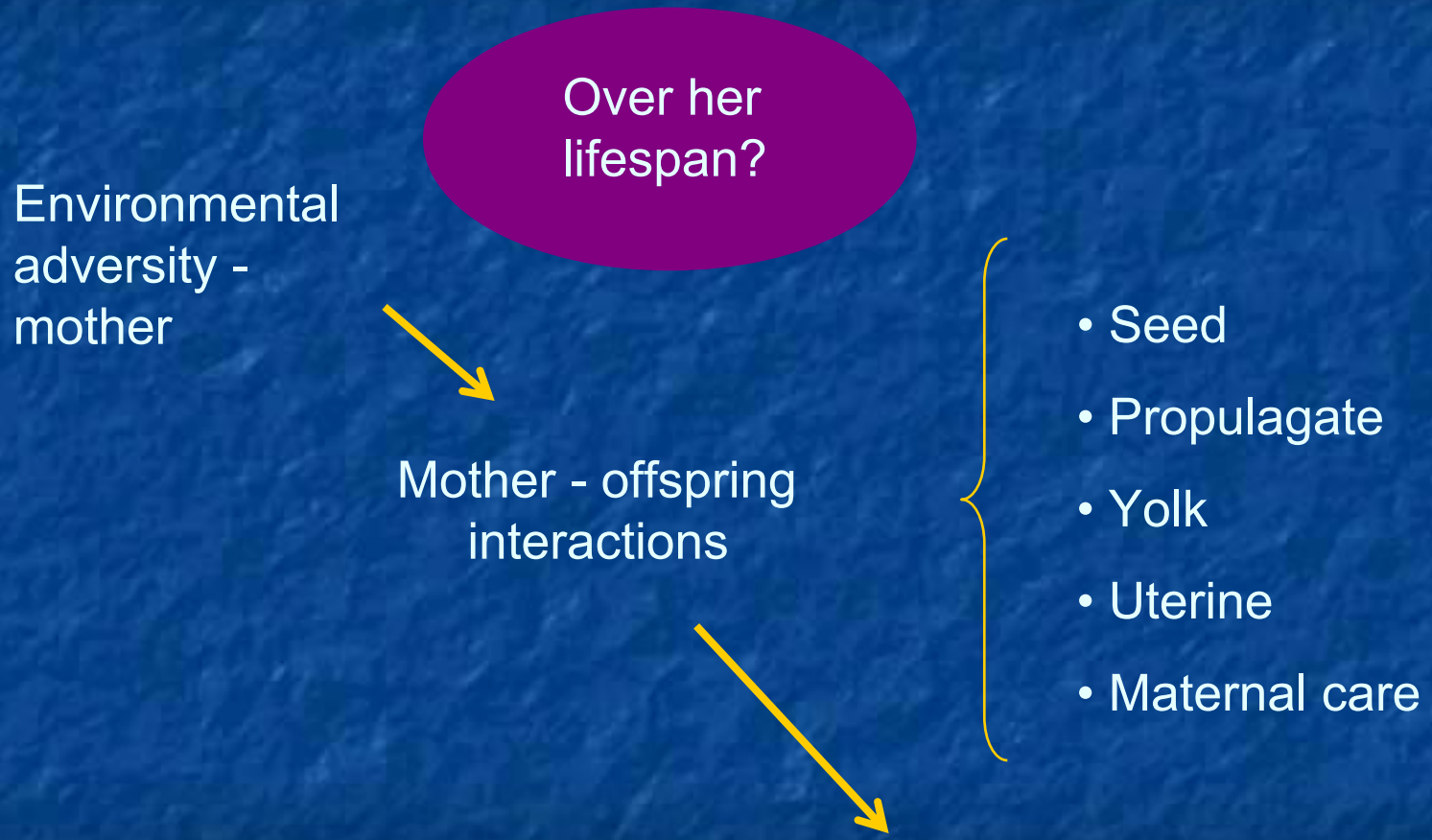

Environmental adversity
Parenting/ Home resources
Child development

Evolutionary biology - Maternal effects



Robert Hinde: Evolution has shaped the young to use parental signals as a 'forecast' of the quality of the environment into which they have been born. For most species, there is no single, optimal phenotype.

Evolutionary Biology: Maternal Effects



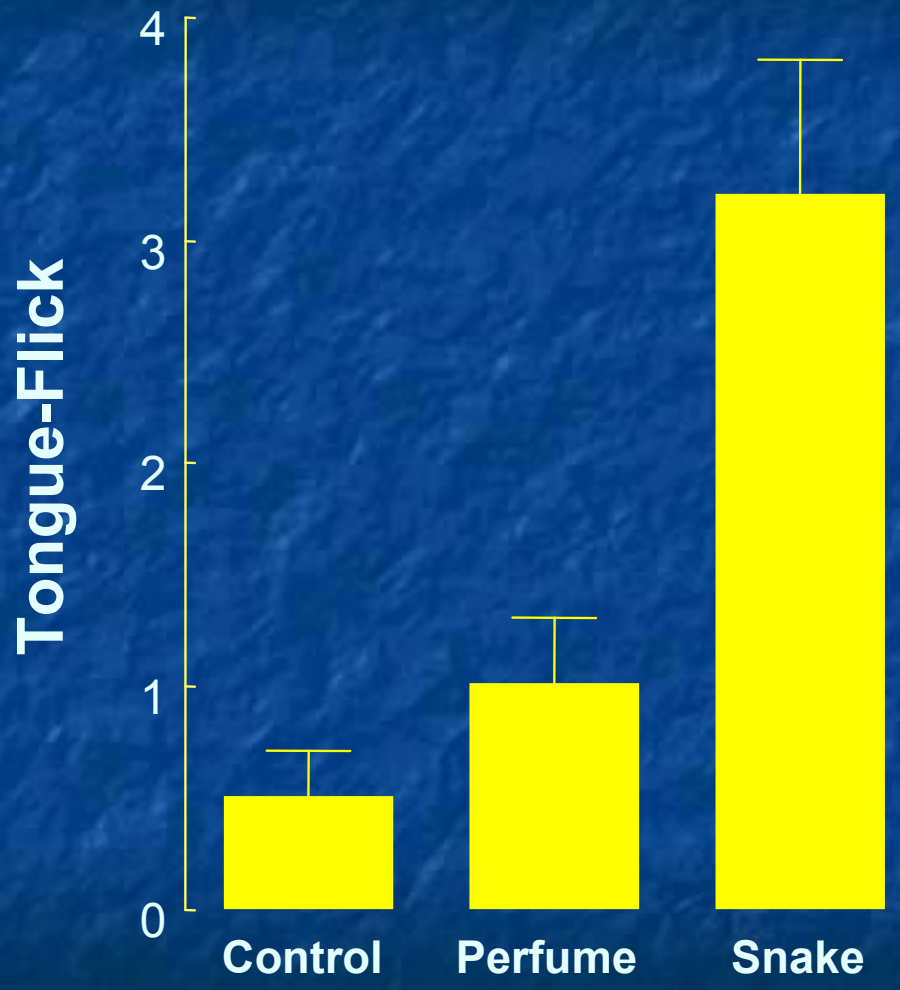
Defense to snake predation in skink lizards

Most frequent prey

- **smaller**
- **shorter tails**
- **less reactive to snake cues**

If mother has been exposed to the scent of a predatory snake then offspring are larger, with longer tails and ….

Response to snake odours



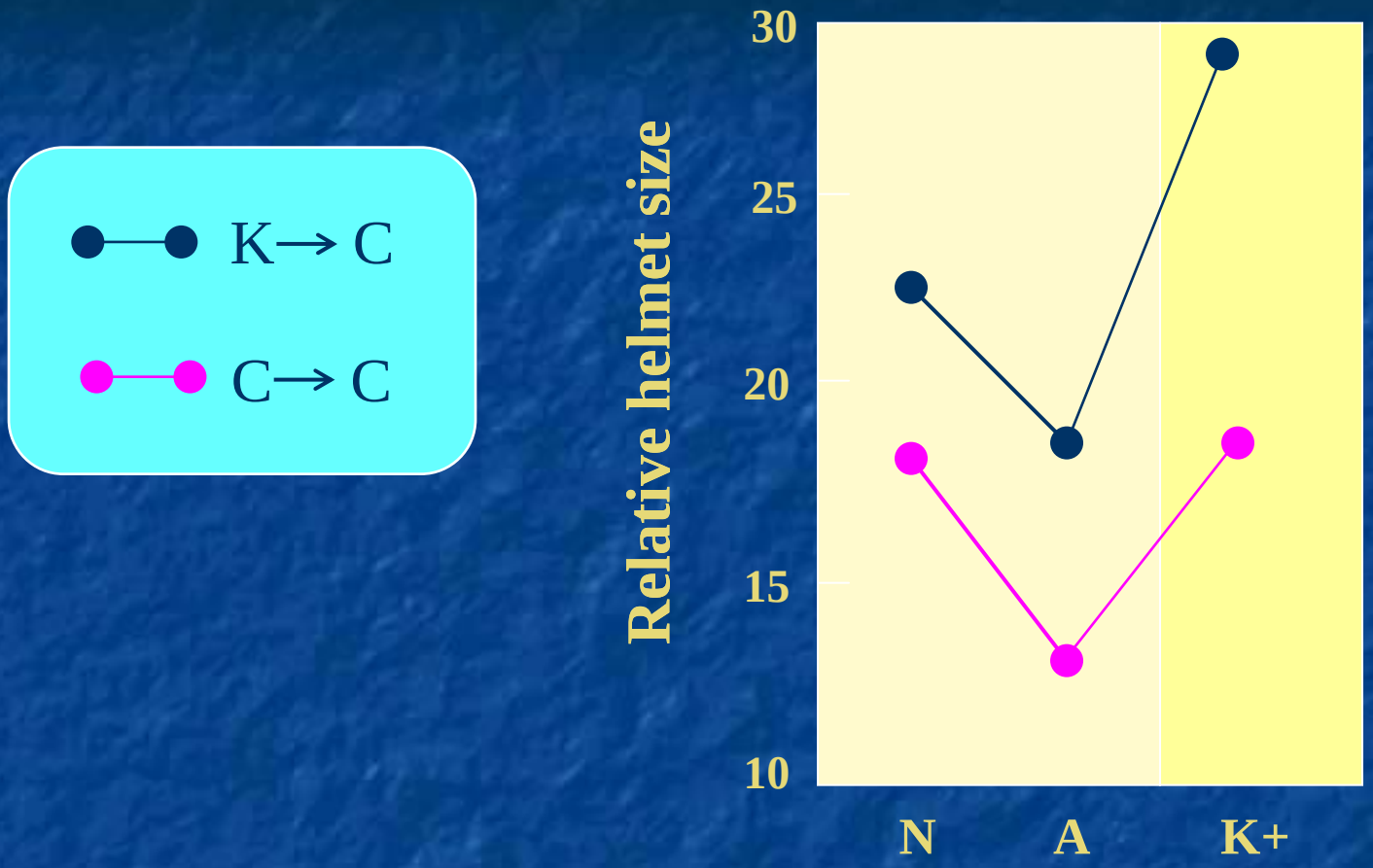
Offspring are significantly larger and with longer tails

Inducible defenses

Predator exposed

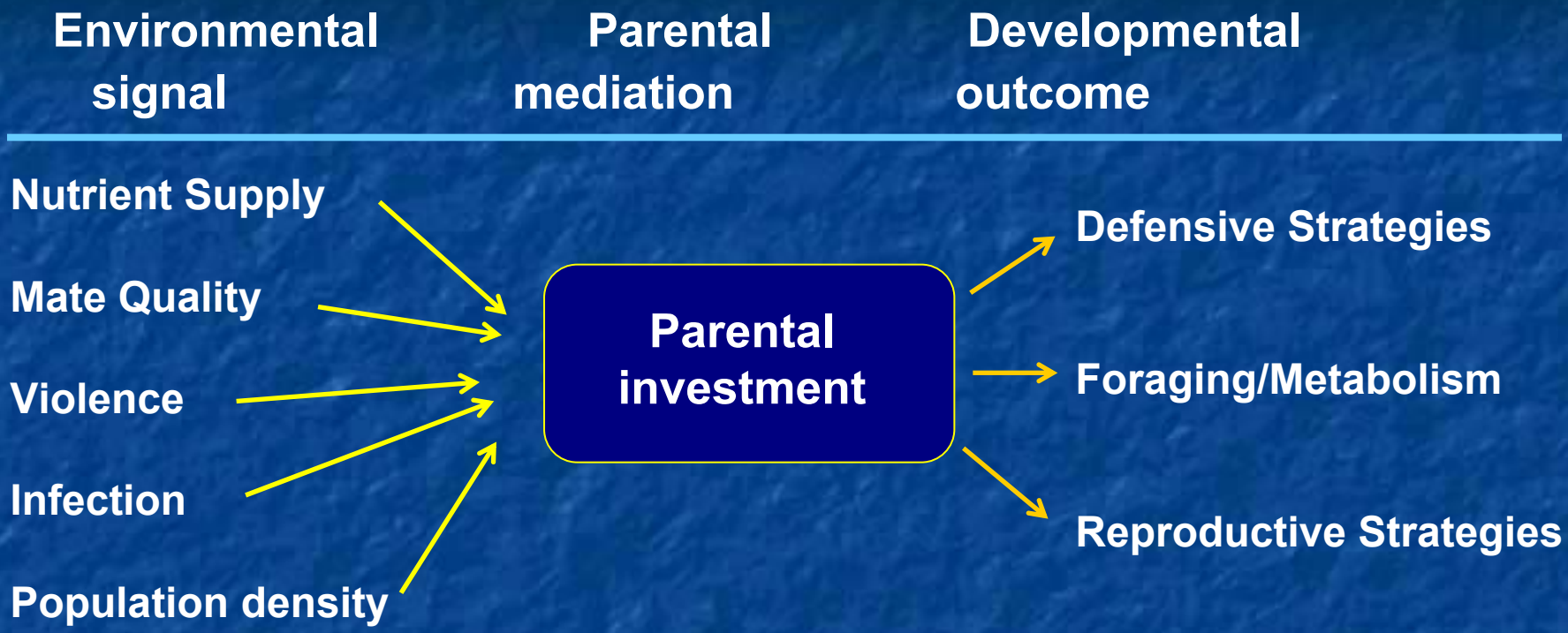
Control

Scanning electron micrograph showing typical and predator-induced morphs of Daphnia cucullata of the same clone.



Agrawal et al. *Nature* 1999

Evolutionary biology - Maternal effects



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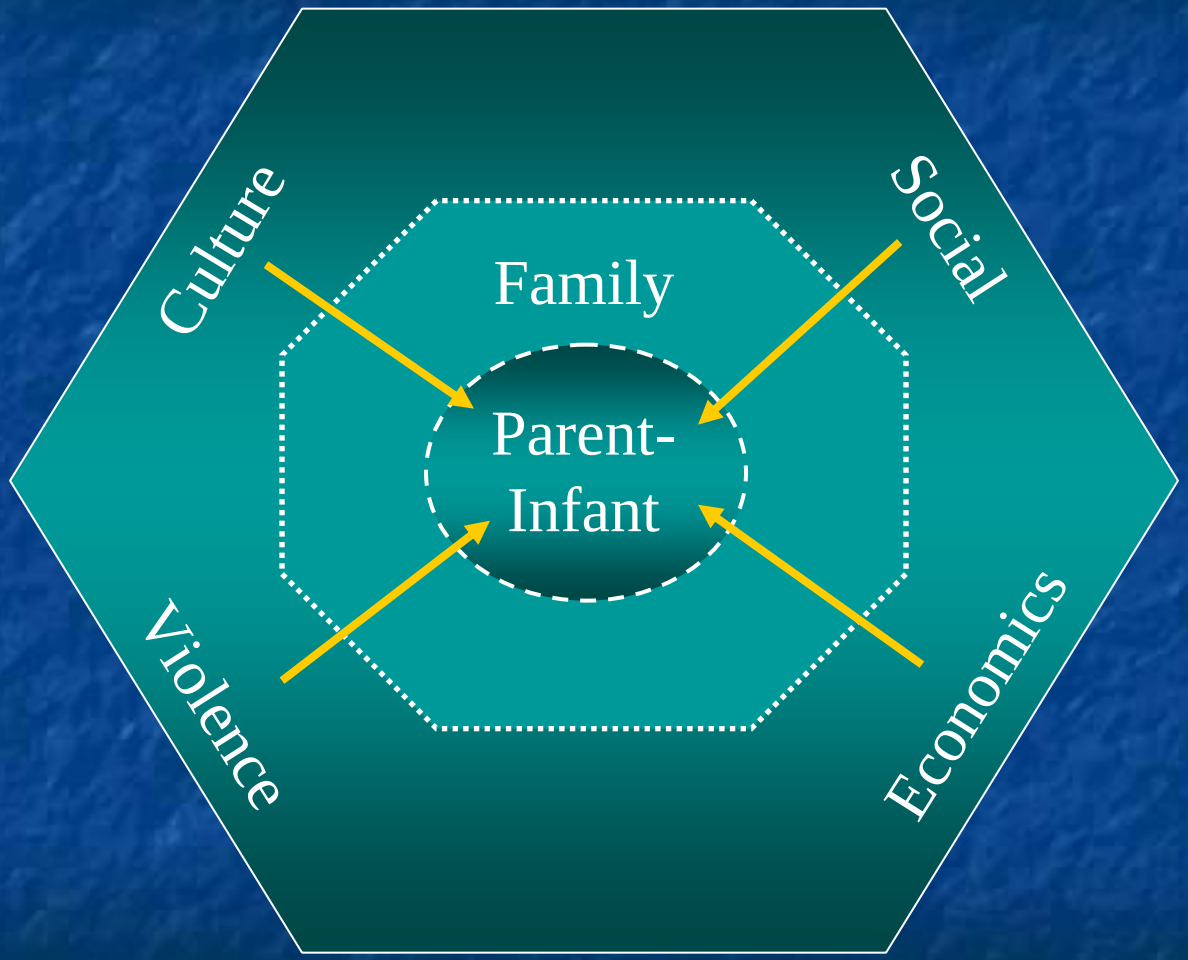

Culture
Social
Family
Parent-
Infant
Violence
Economics

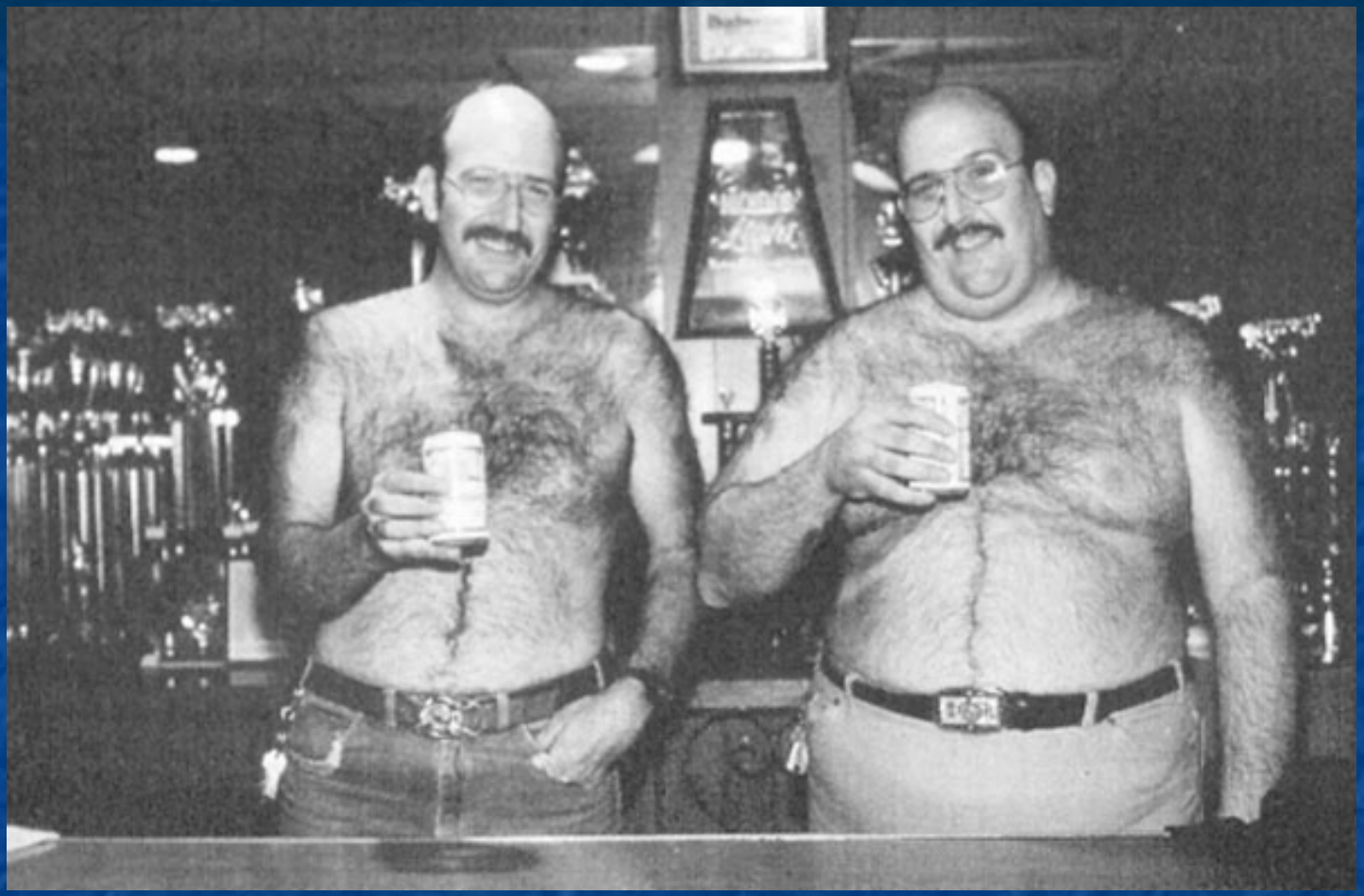
Summary

- Parental care affects the activity of genes in the brain that regulate stress responses, neural development and reproduction.

- This parental effect involves a form a “plasticity” at the level of the DNA.

Epigenetics: Any functional change in the genome that does not involve an alteration of DNA sequence.

Multiple phenotypes from a common genotype



Creating diversity in phenotype from a common genome

Genetic code is defined by the sequence of four nucleotides that produce proteins and other molecules that serve cell function.

C T A C G T A C T C G G A A T C T C G

Protein

Epigenetic effects refer to modifications of the chemistry of the DNA, but <u>not</u> to a change of sequence. Epigenetics alters the activity of the gene, but not its function.

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Warning!!!

Incomprehensible scientific jargon will follow…

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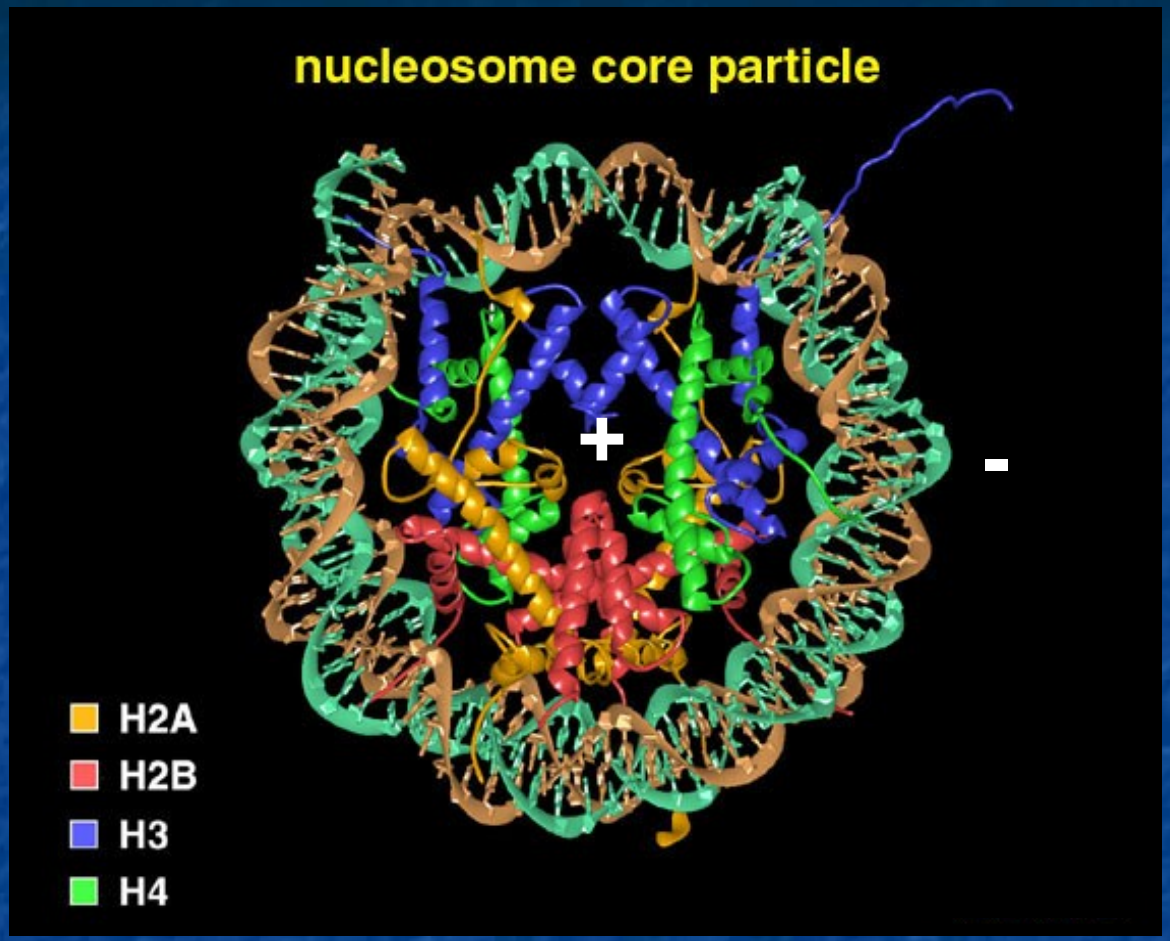
- DNA methylation: The addition of a methyl group onto a cytosine.
- DNA methylation is chemically very stable (potentially lasting for the life of the organism).
- DNA methylation silences gene expression.

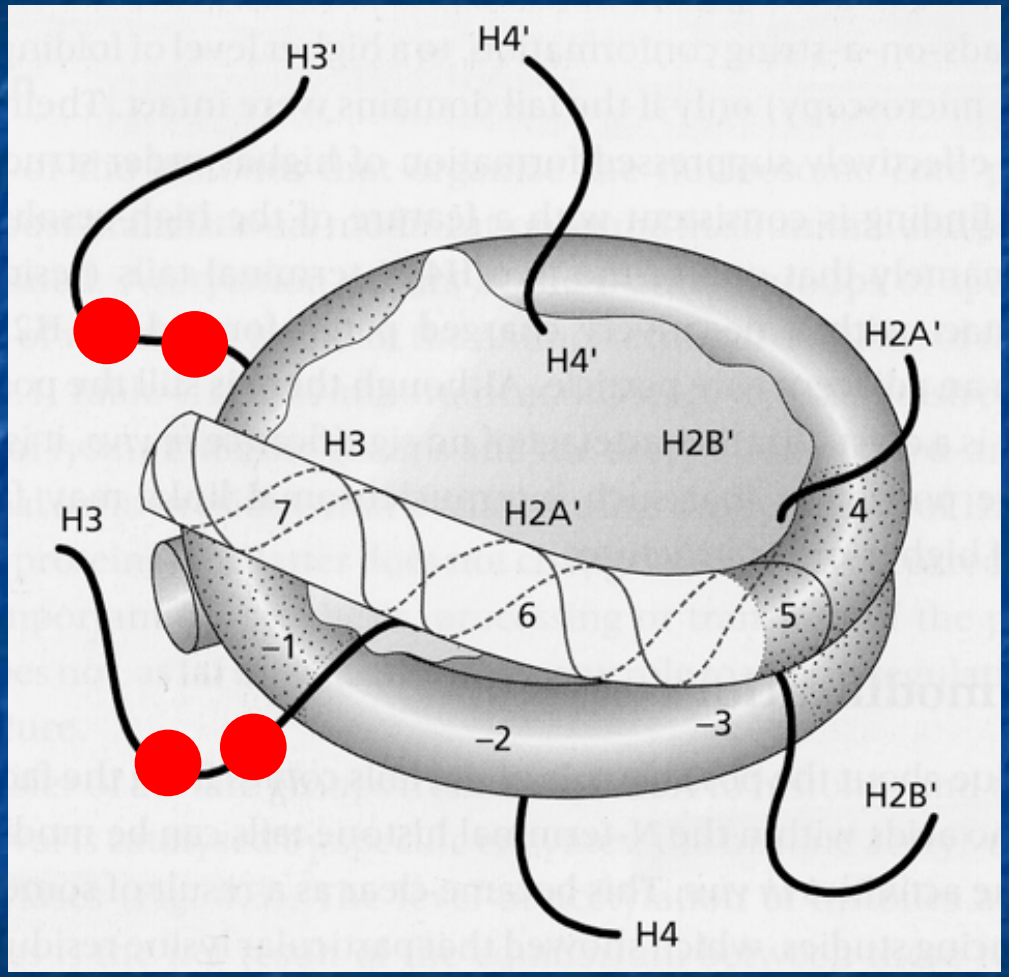


Prevents TF binding to DNA

TF binding involves alteration of chromatin structure

Nucleosome core particle: ribbon traces for the 146-bp DNA phosphodiester backbones (brown and turquoise) and eight histone protein chains (Luger et al. Nature 1997).

Acetyl group



B. Turner. Chromatin structure and gene regulation. 2001.

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C T A C G T A C T C G G A A T C T C G

RNAs, proteins

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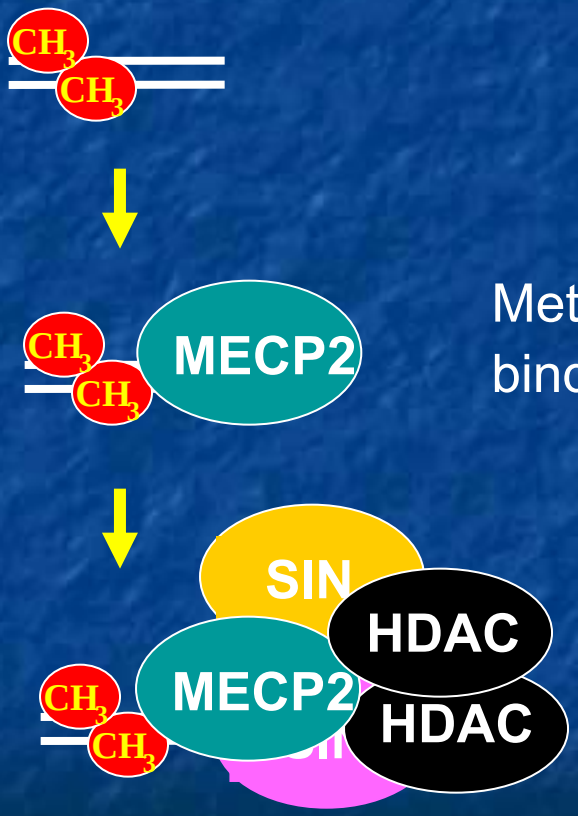
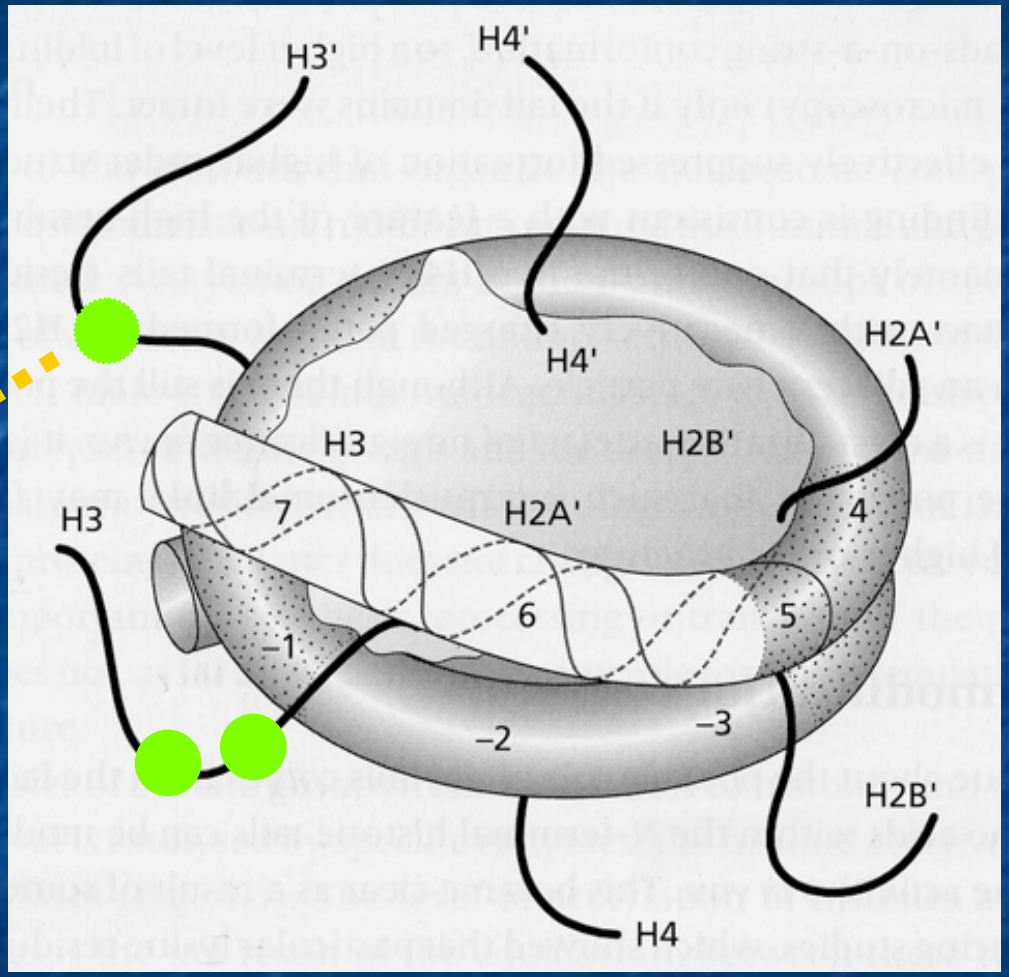
DNA Methylation can inhibit gene expression by blocking transcription factors binding



Methylated DNA binding protein

HDAC: Histone deacetylase

HDAC



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Parental care → Epigenetic mark → Gene expression → Phenotype

Naturally-occurring variations in maternal care

Expression of specific genes in brain regions

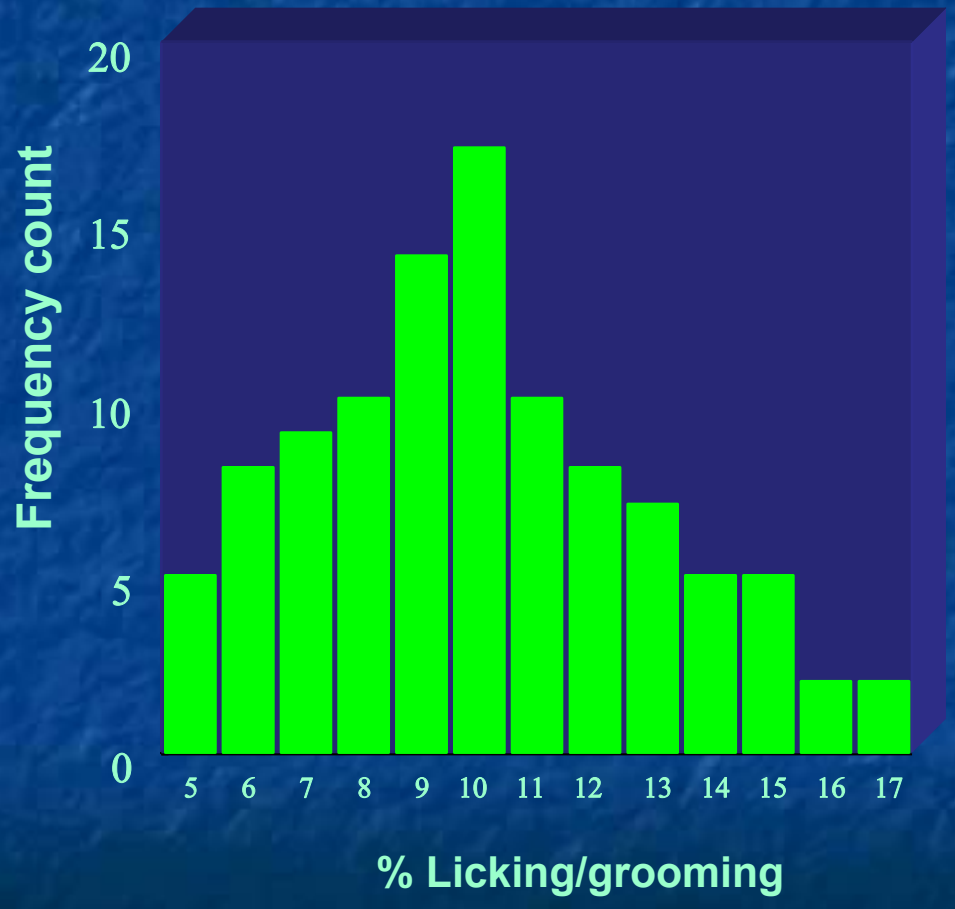
HPA function

Stable individual differences in stress reactivity

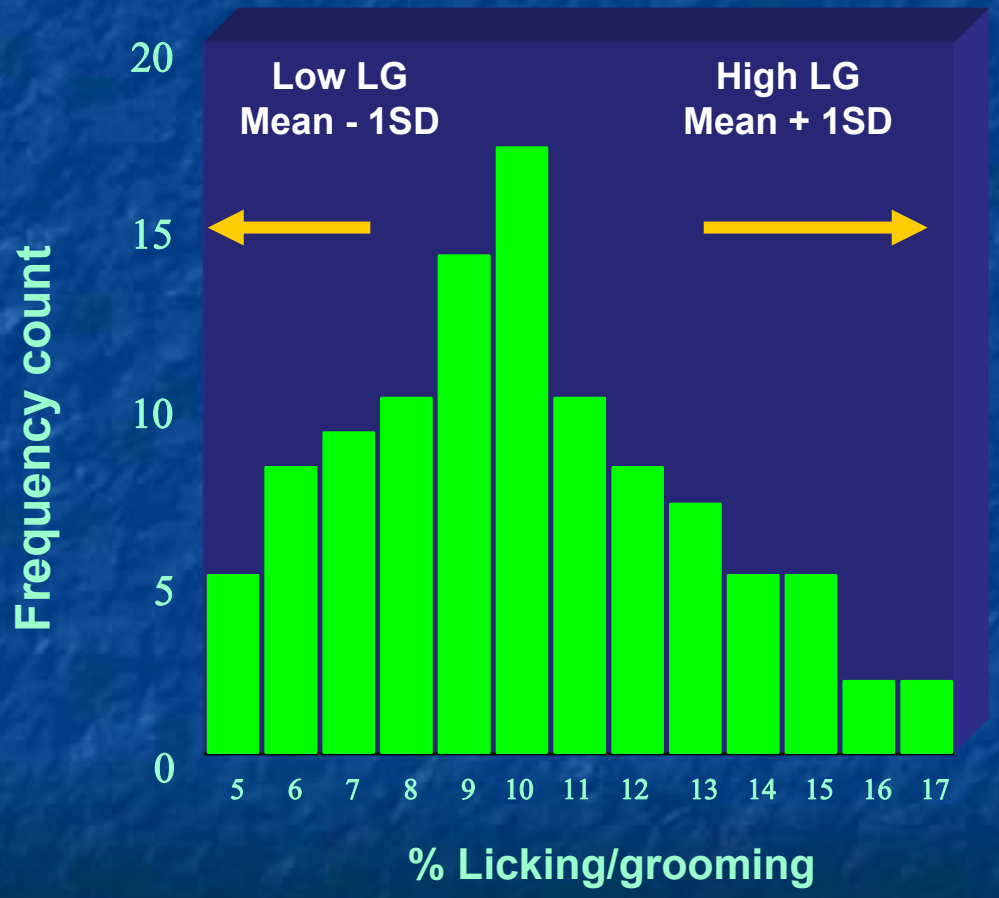
Maternal licking/grooming

Source of tactile stimulation/nurturance: Enhances activity of endocrine systems (e.g., GH/IGF) that promote somatic growth, suppresses those (glucocorticoids) that inhibit growth

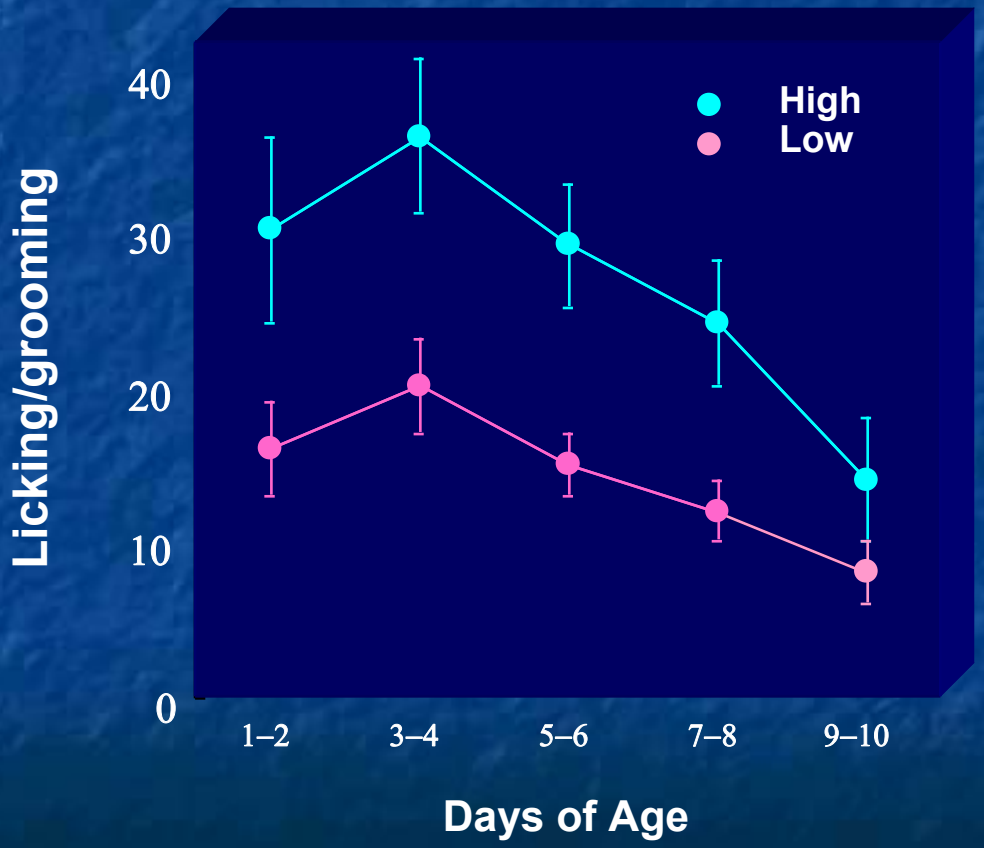
Variations in maternal care



Variations in maternal care



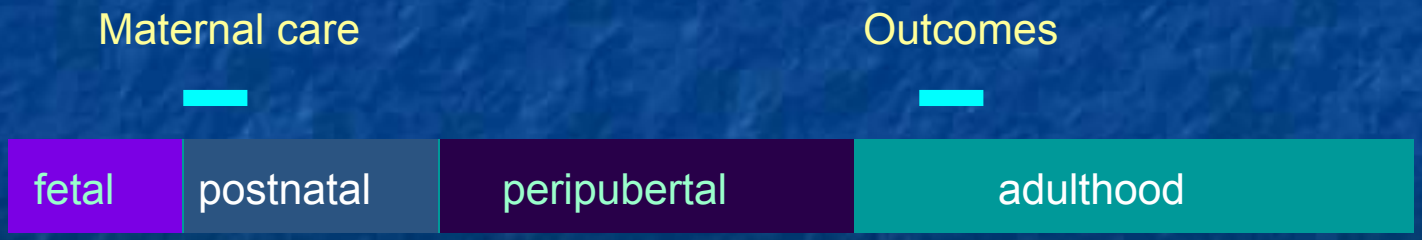
Variations in maternal care X Days

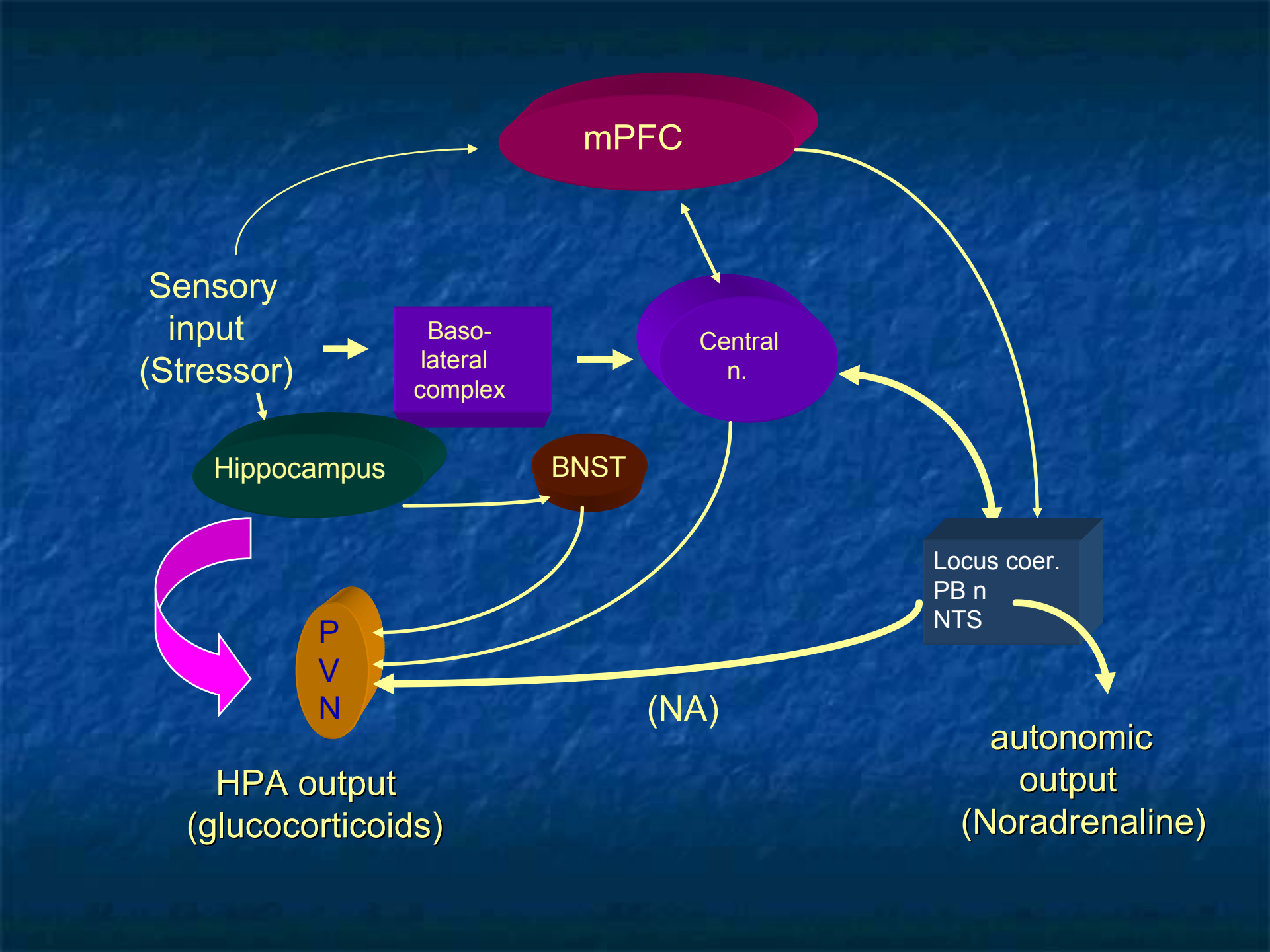


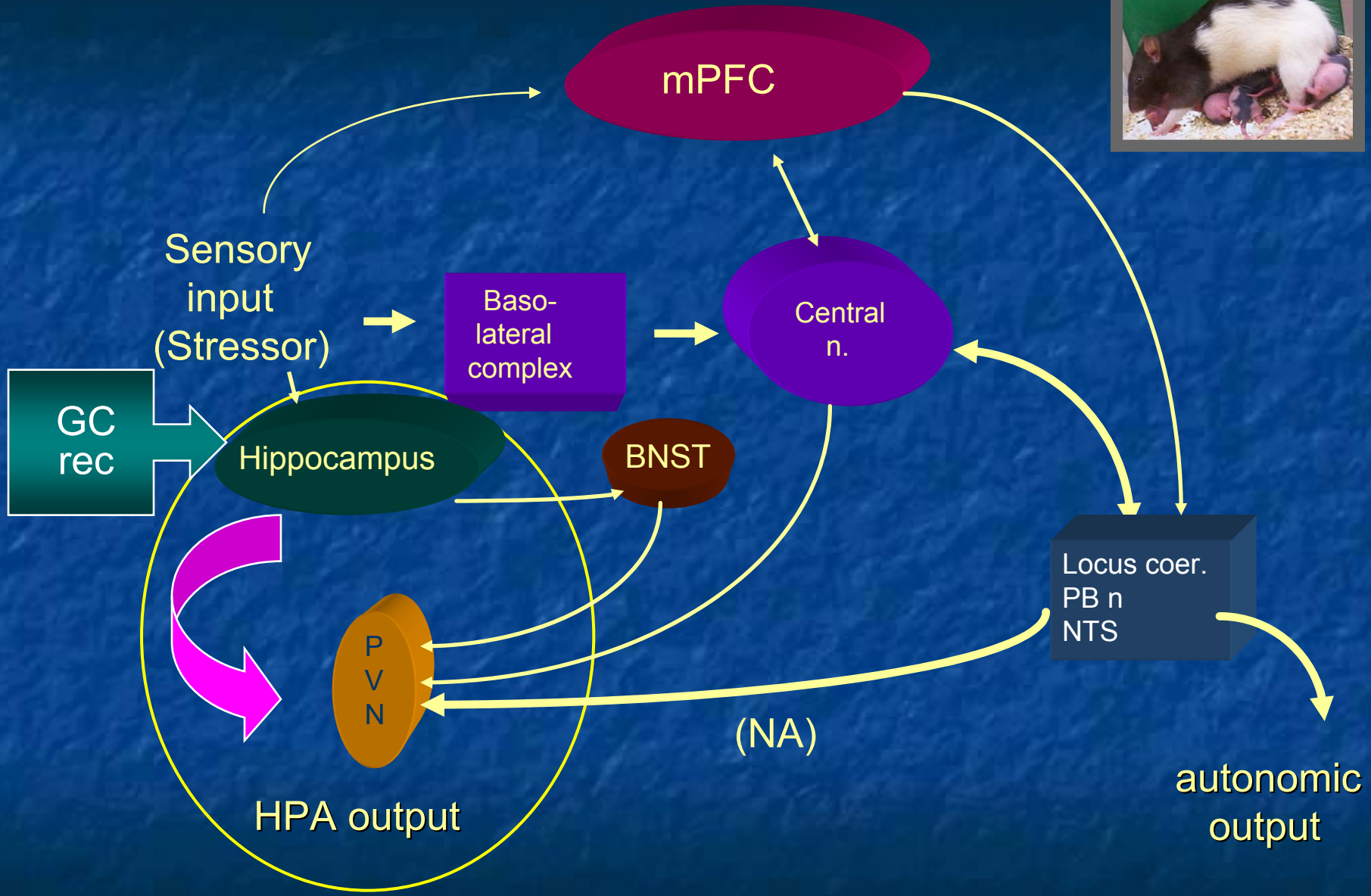
* No differences in time with pups

Are these naturally-occurring variations in maternal behaviour associated with the development of individual differences in endocrine and behavioural responses to stress?

* Effects hold for both males and females




mPFC
Sensory
input
(Stressor)
Baso-
lateral
complex
Central
n.
Hippocampus
BNST
Locus coer.
PB n
NTS
P
V
N
(NA)
HPA output
(glucocorticoids)
autonomic
output
(Noradrenaline)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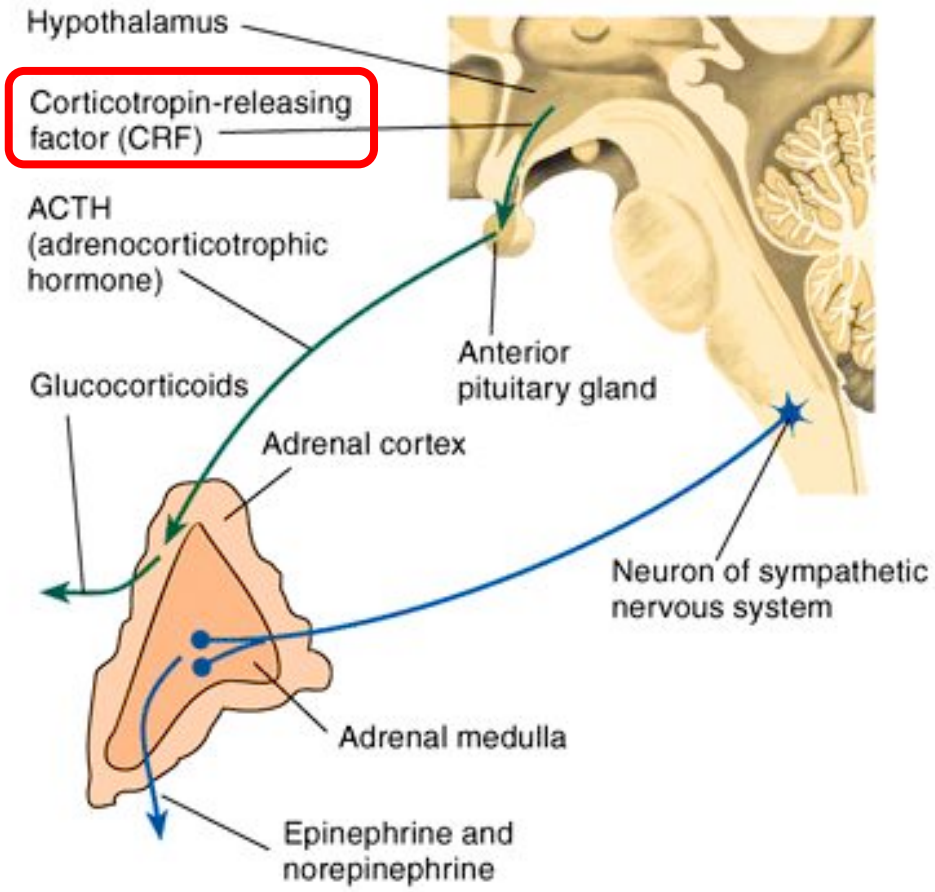
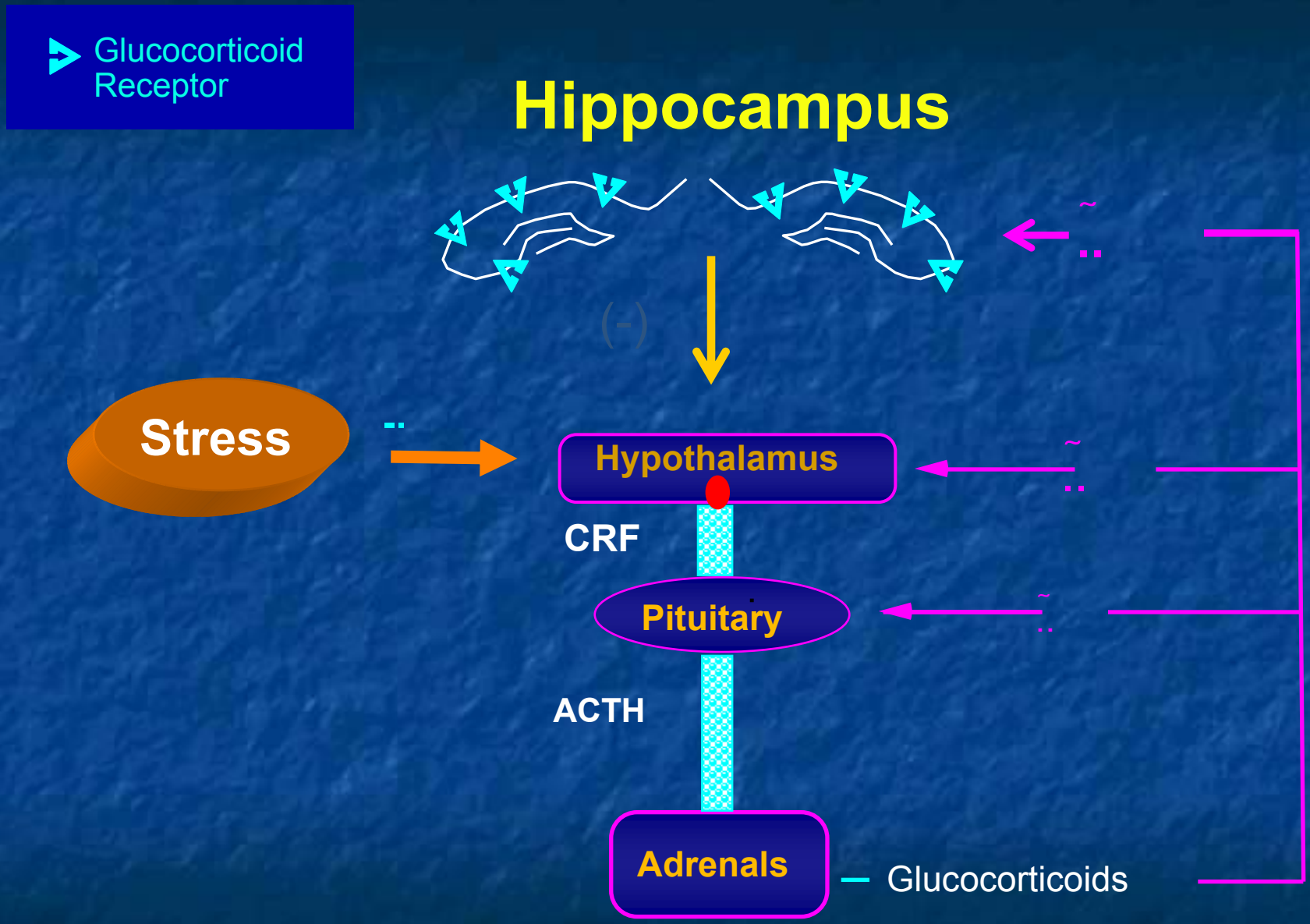
autonomic
output

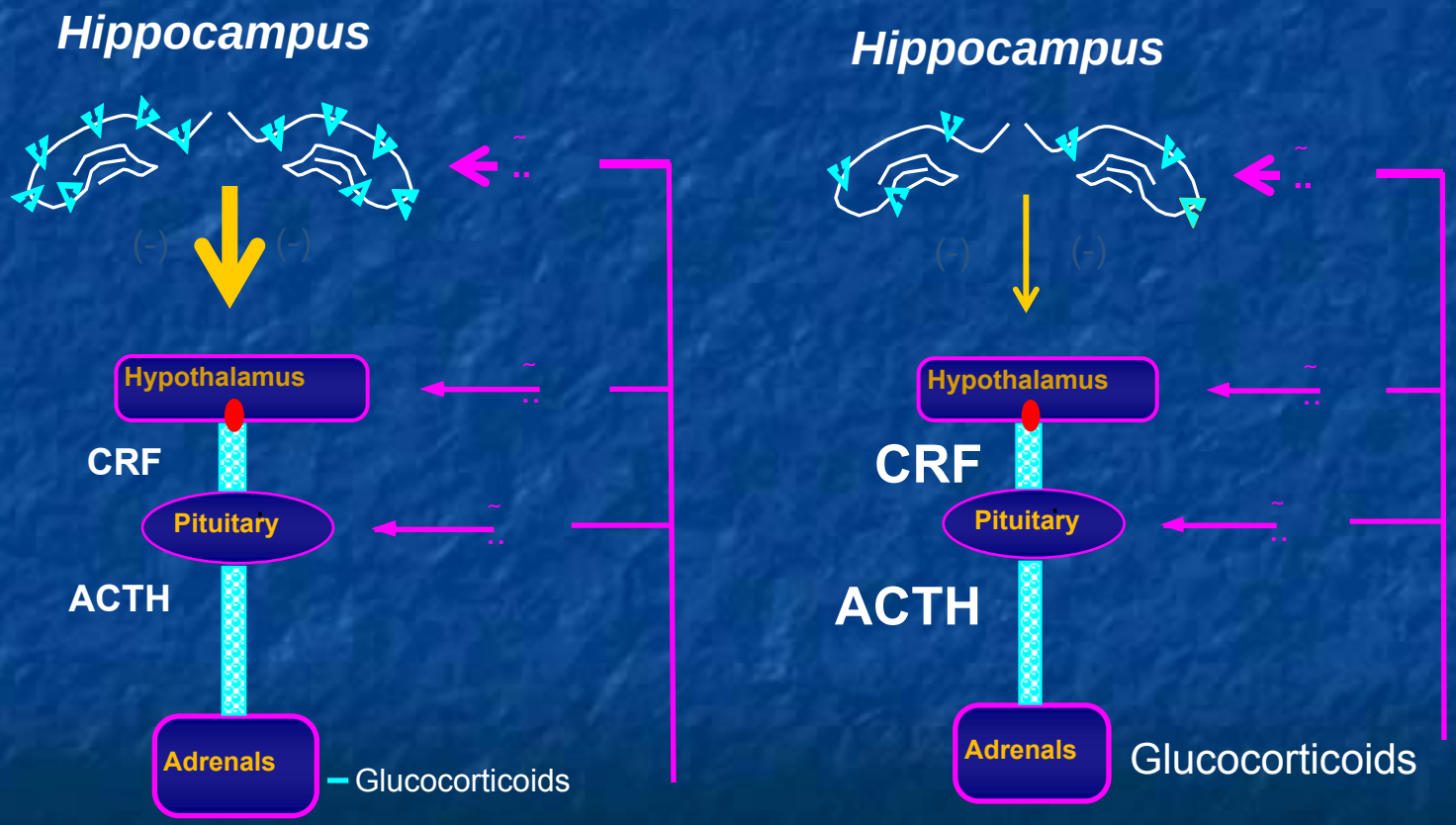
- **Control of the Secretion of Glucocorticoids by the Adrenal Cortex and of Catecholamines by the Adrenal Medulla**





CRF: corticotropin releasing factor. ACTH: adrenocorticotropin

Individual differences in glucocorticoid receptor levels lead to altered pituitary-adrenal responses to stress



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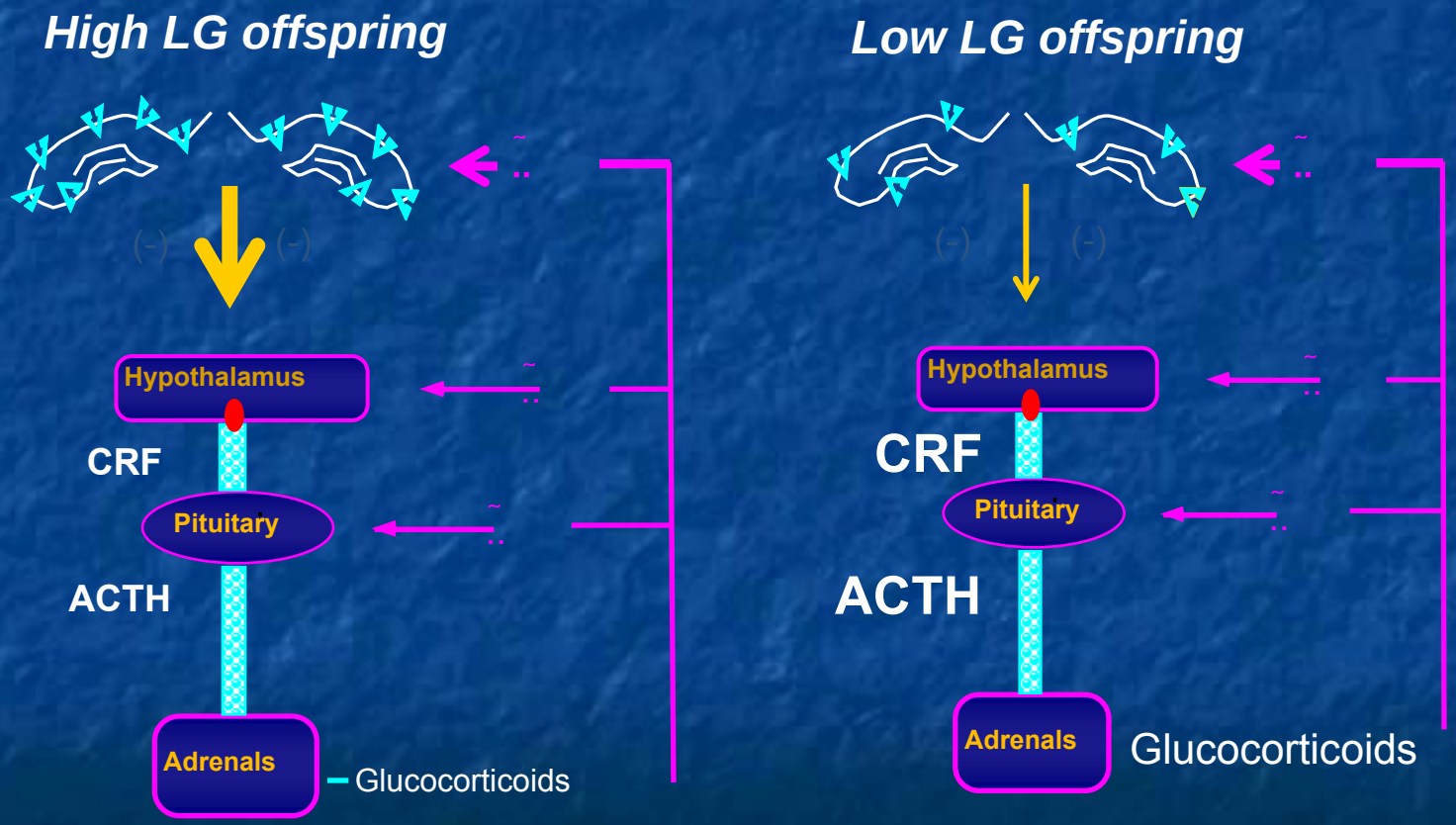
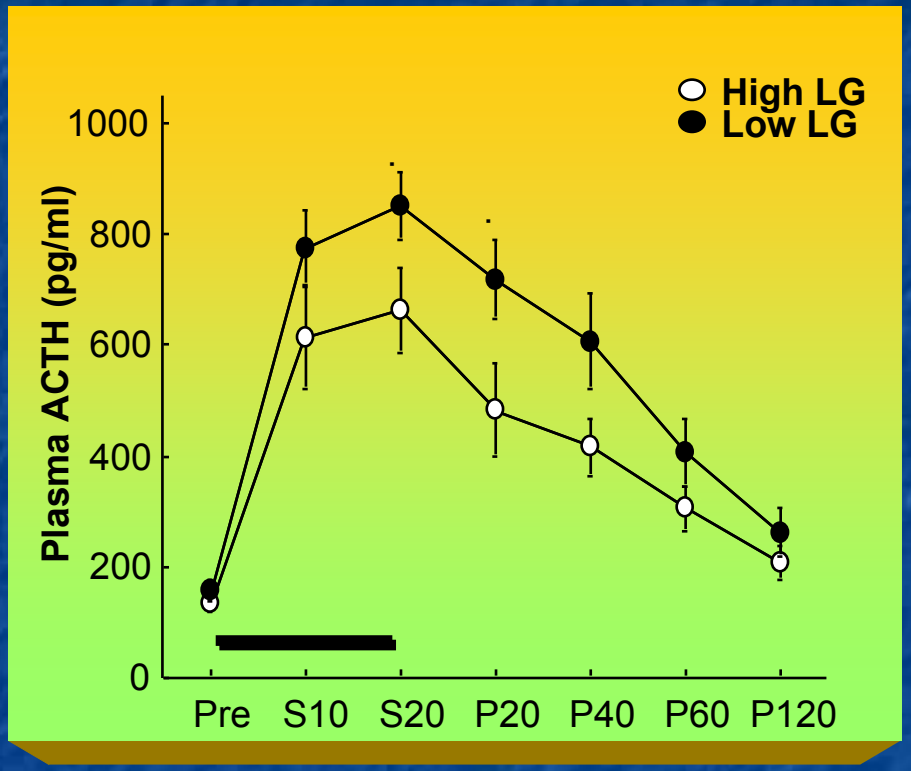
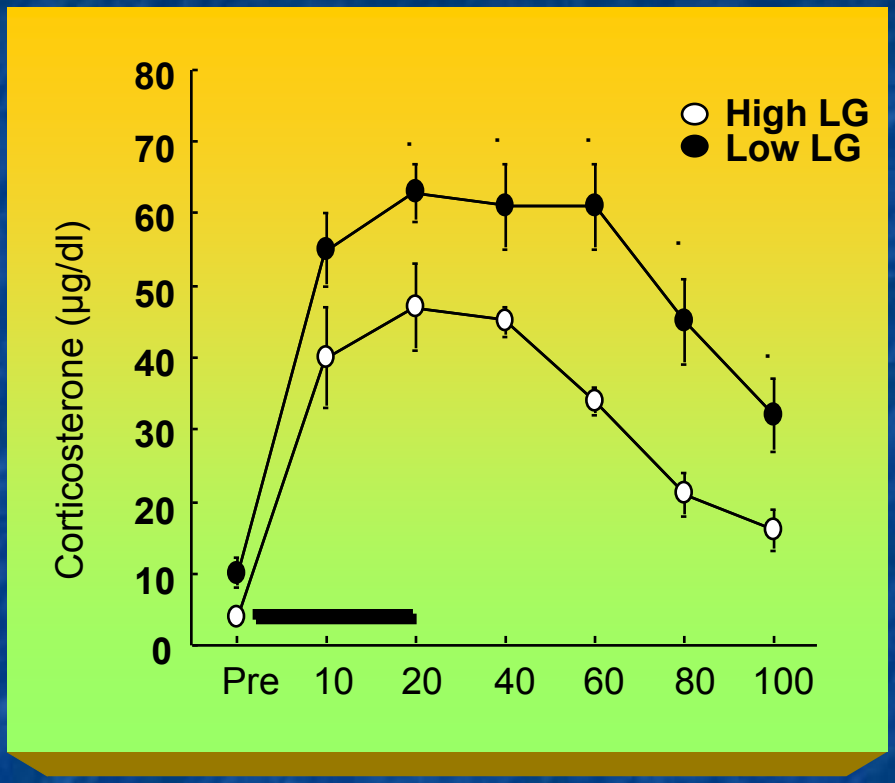


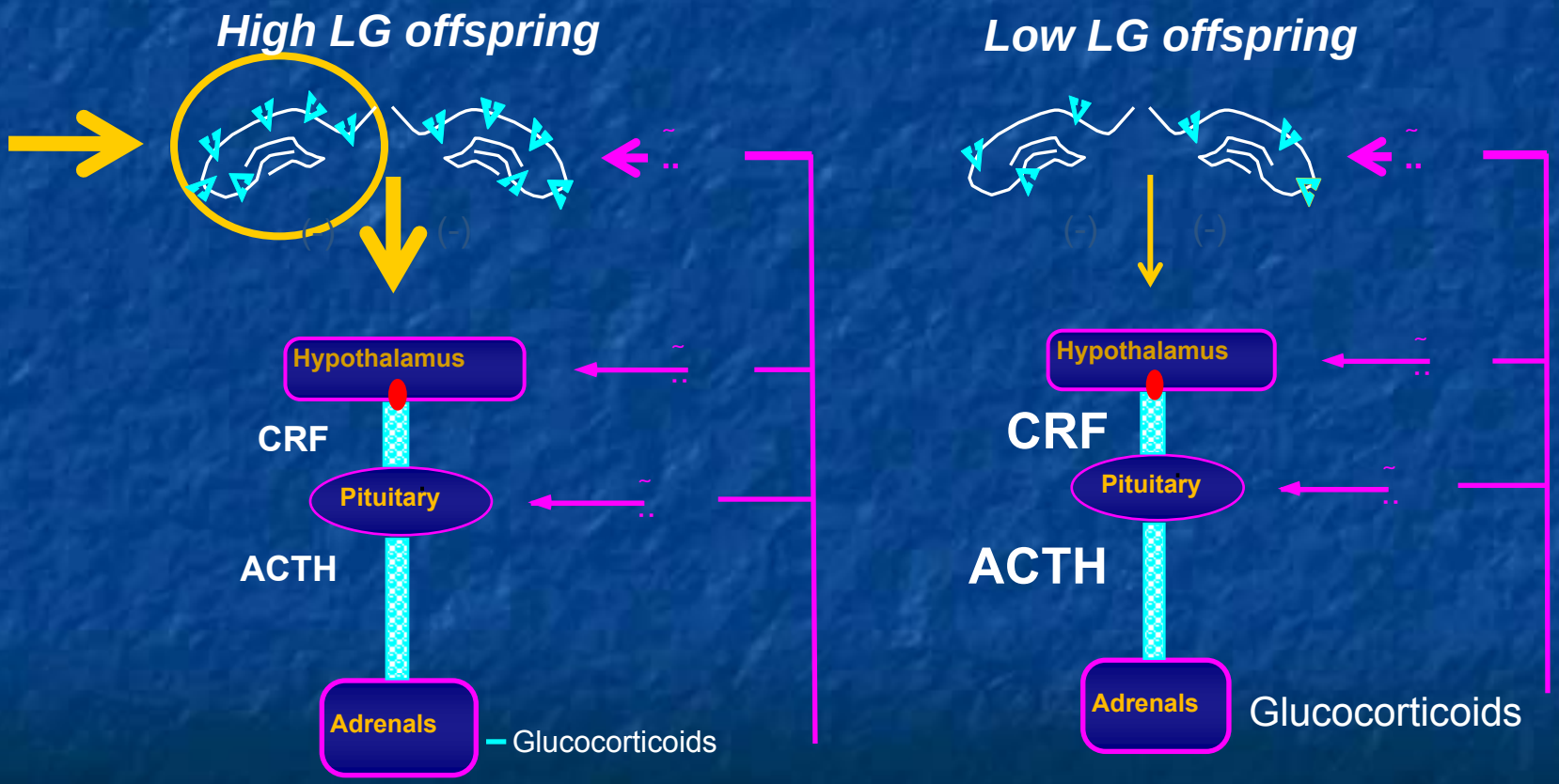
Adult offspring of High LG mothers show more modest HPA responses to stress





Intra-hippocampal infusion of a GR antagonist completely eliminates the maternal effect on HPA responses to stress

Individual differences in glucocorticoid receptor levels lead to altered pituitary-adrenal responses to stress

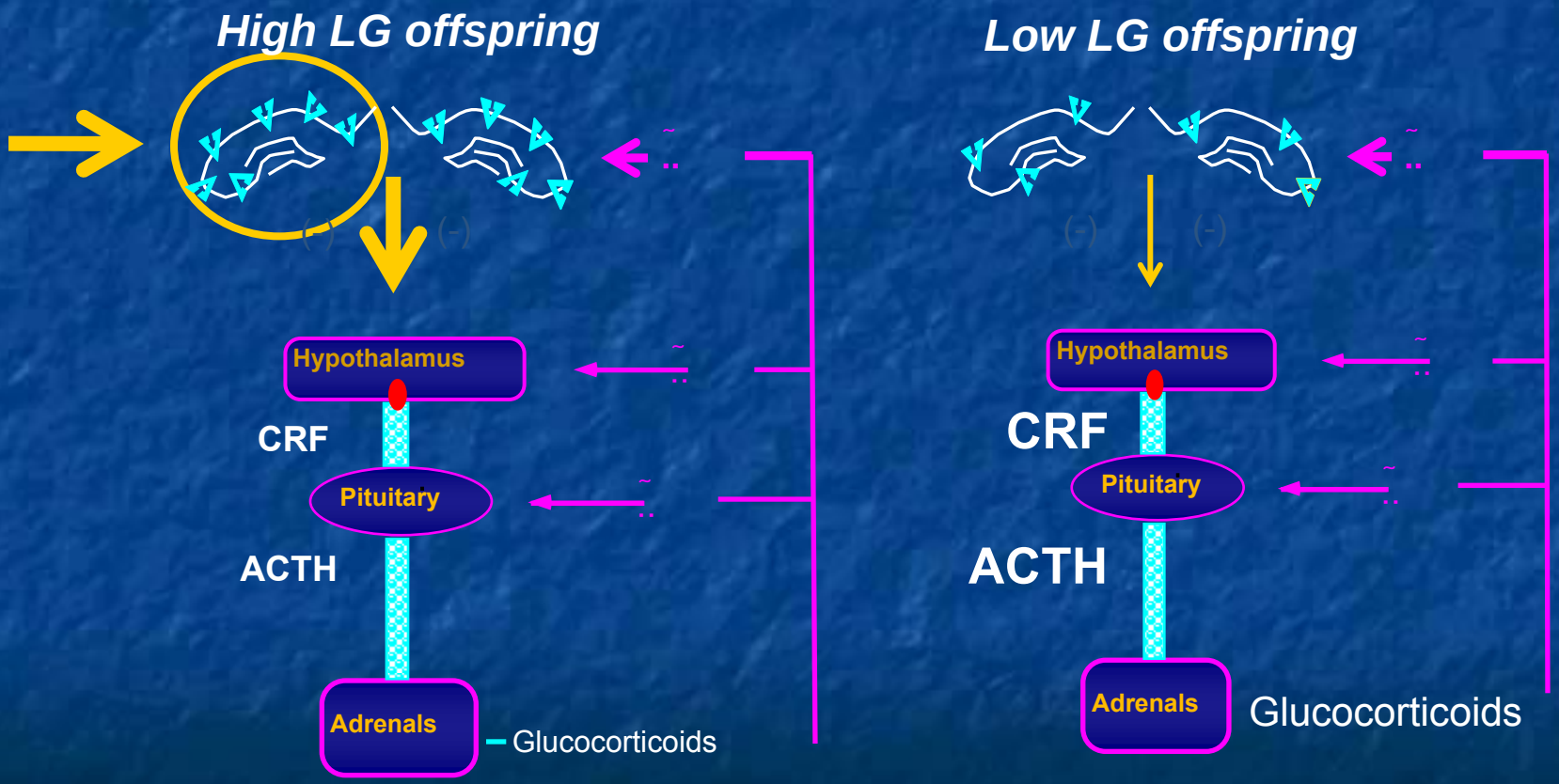


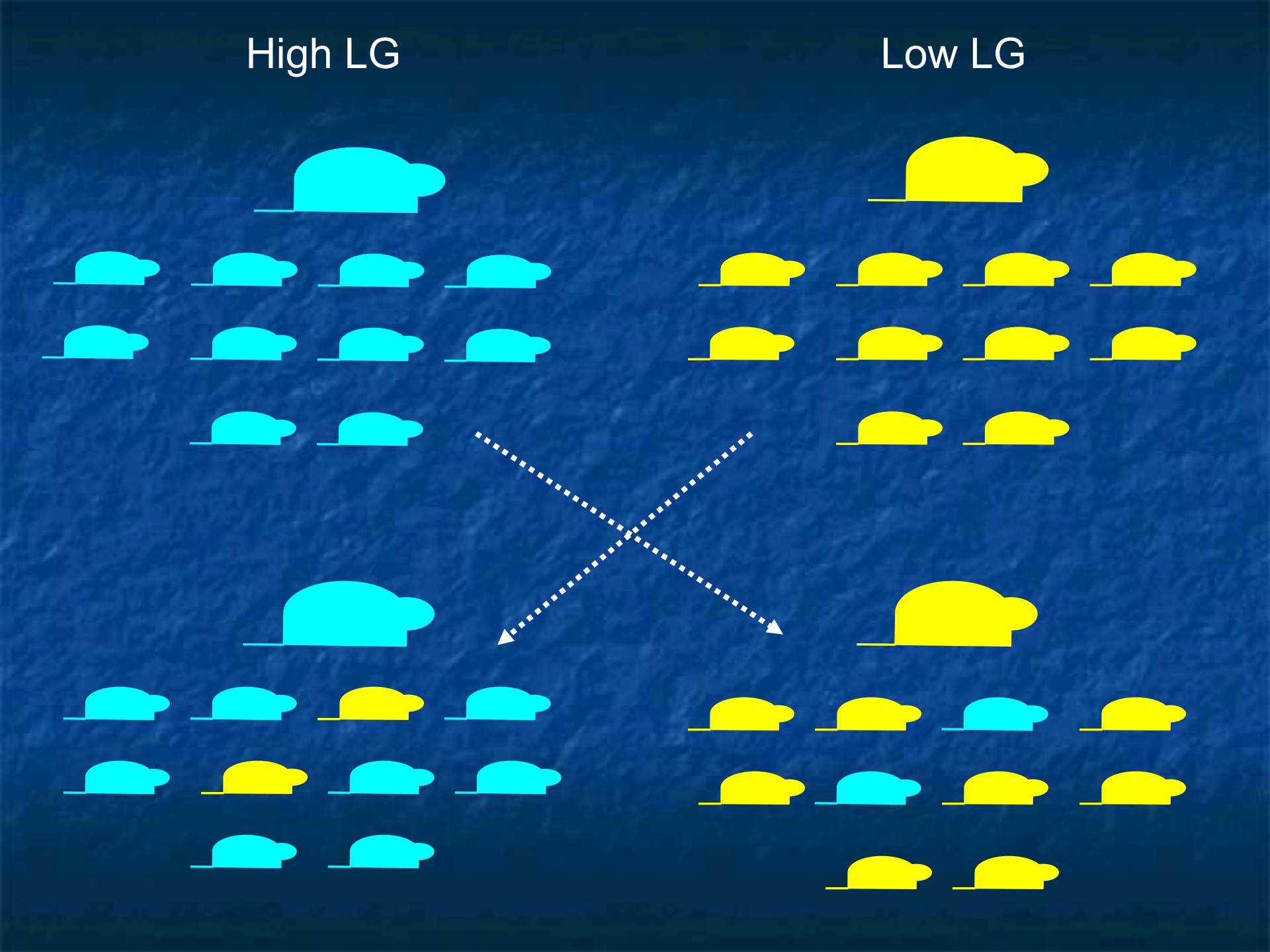
Adaptive advantages of *increased* stress reactivity (Central CRF systems, HPA axis, Catechols)

- Increased resistance to sepsis (infection).
- Increased resistance to famine.
- Decreased mortality due to aggressive conflict.

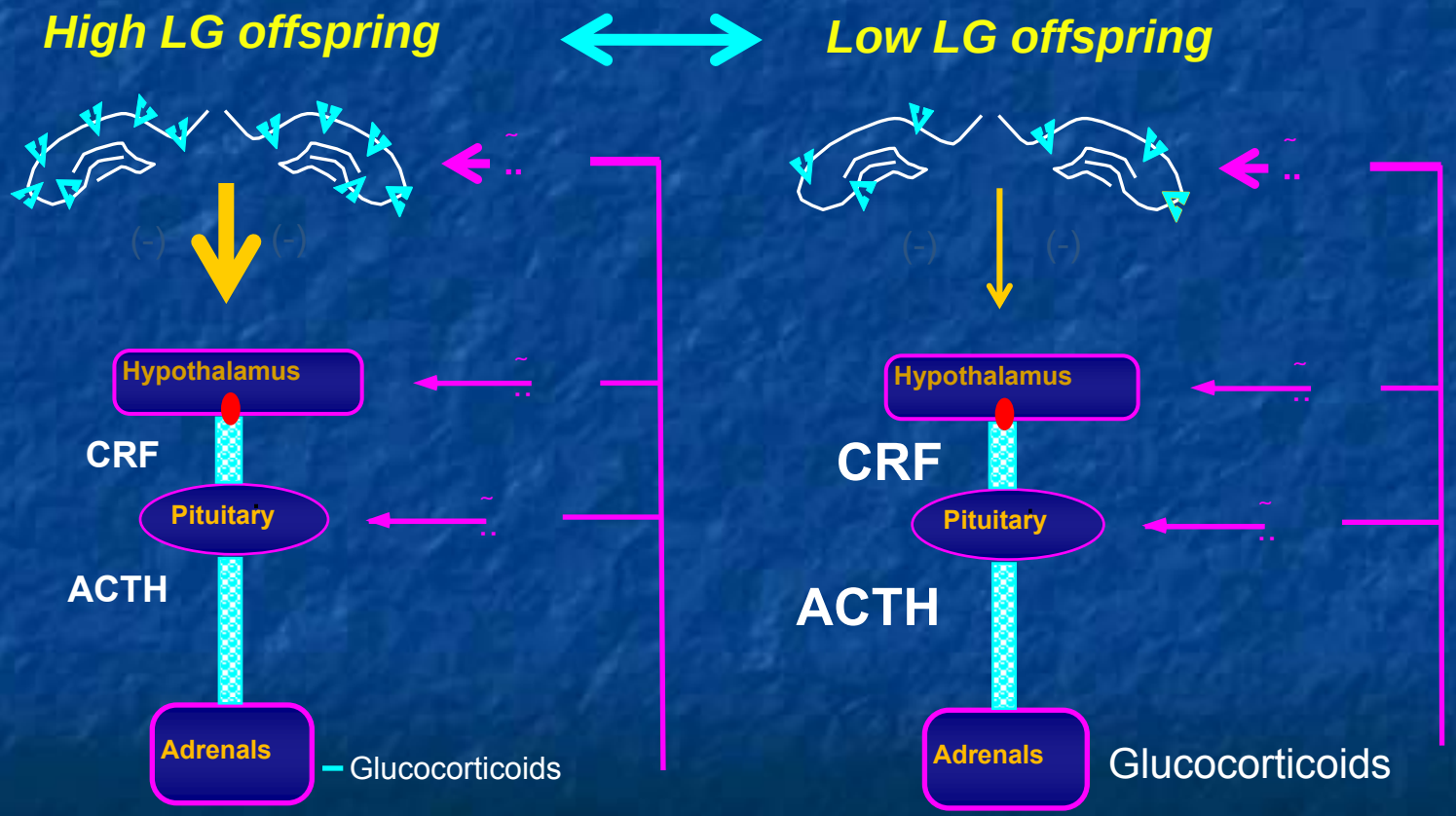
Poverty: Pathogens, nutritional deprivation and violence

Individual differences in glucocorticoid receptor levels lead to altered pituitary-adrenal responses to stress

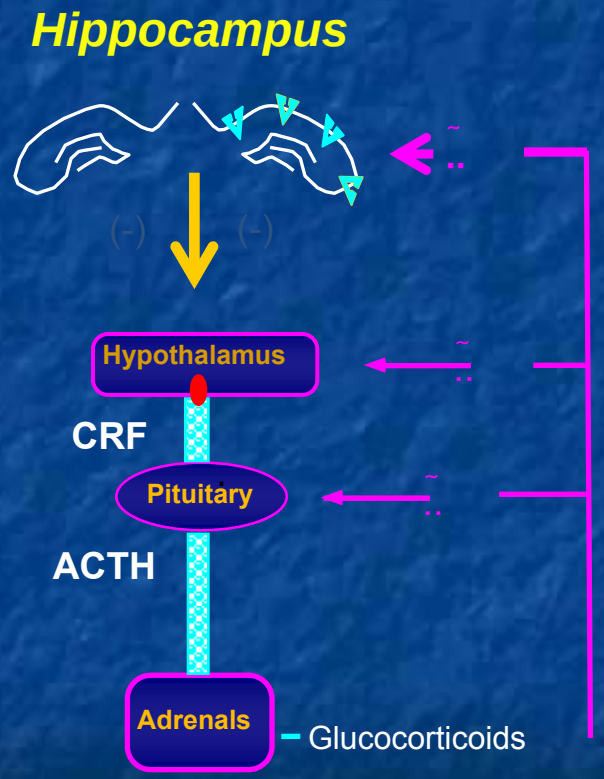



High LG
Low LG

Cross-fostering reveals evidence for direct, postnatal effects of maternal care



?


Hippocampus
(-)
(-)
Hypothalamus
CRF
Pituitary
ACTH
Adrenals
Glucocorticoids

How might maternal licking/grooming regulate hippocampal glucocorticoid receptor gene activity and HPA function?

And how do these effects persist over the lifespan of the offspring?

Relevant gene - environment interaction

Maternal Care

Extracellular signal

Signal Receptor

Intracellular signal

Transcription Factors

gene

DNA

Summary of *in vivo* and *in vitro* studies

Tactile stimulation
(maternal LG)

(H-P-T)

(T3)

5-HT

5-HT7 rec

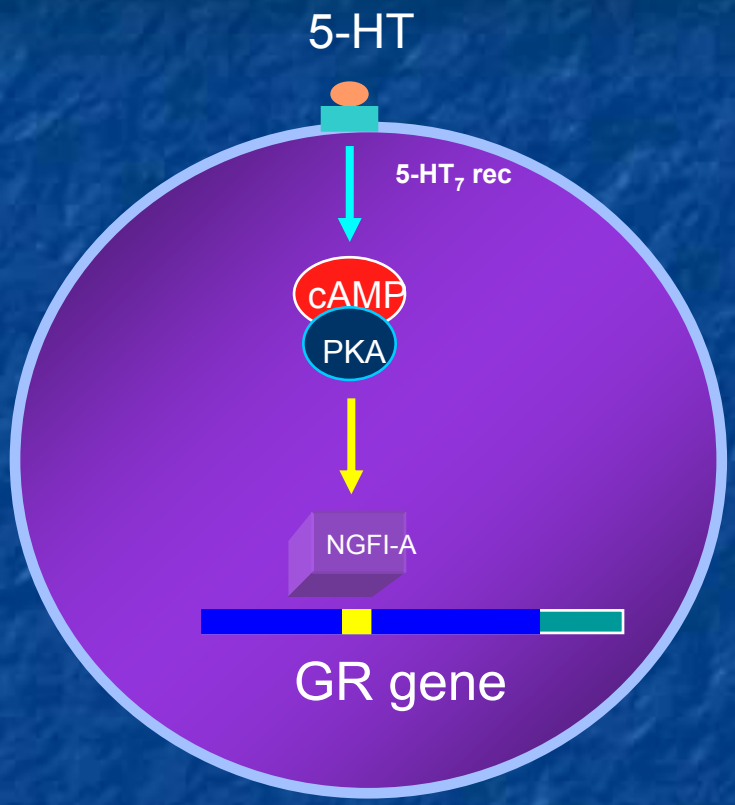
cAMP

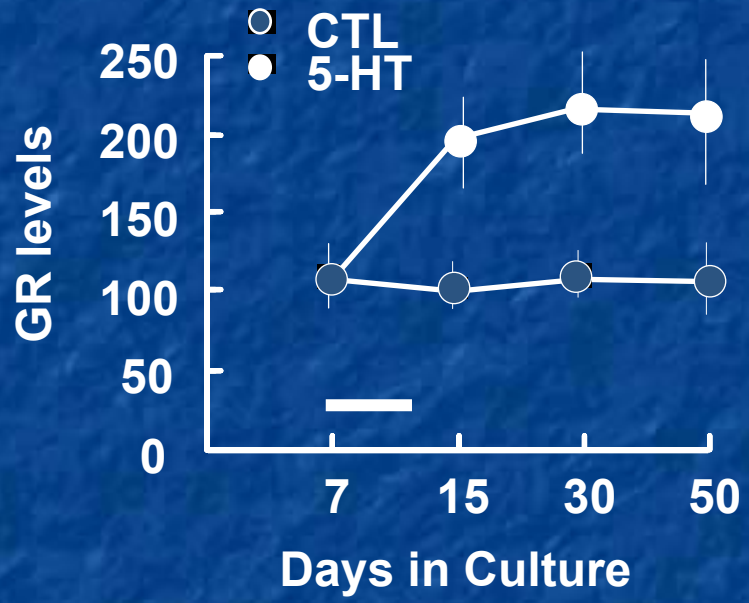
PKA

Transcription
Factor

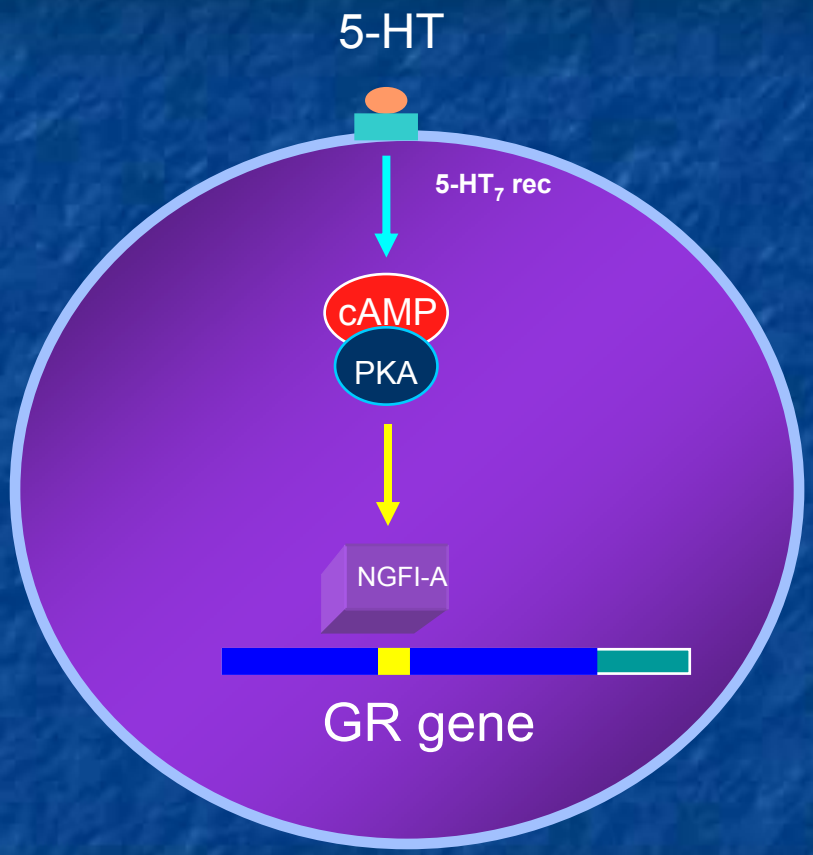
NGFI-A

GR gene





— Represents period of 5-HT exposure

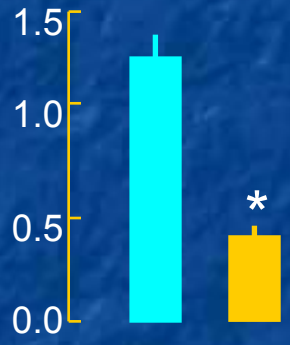


Day 6 pups

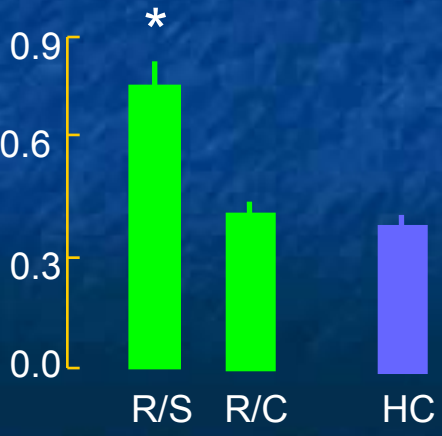
Low LG

High LG

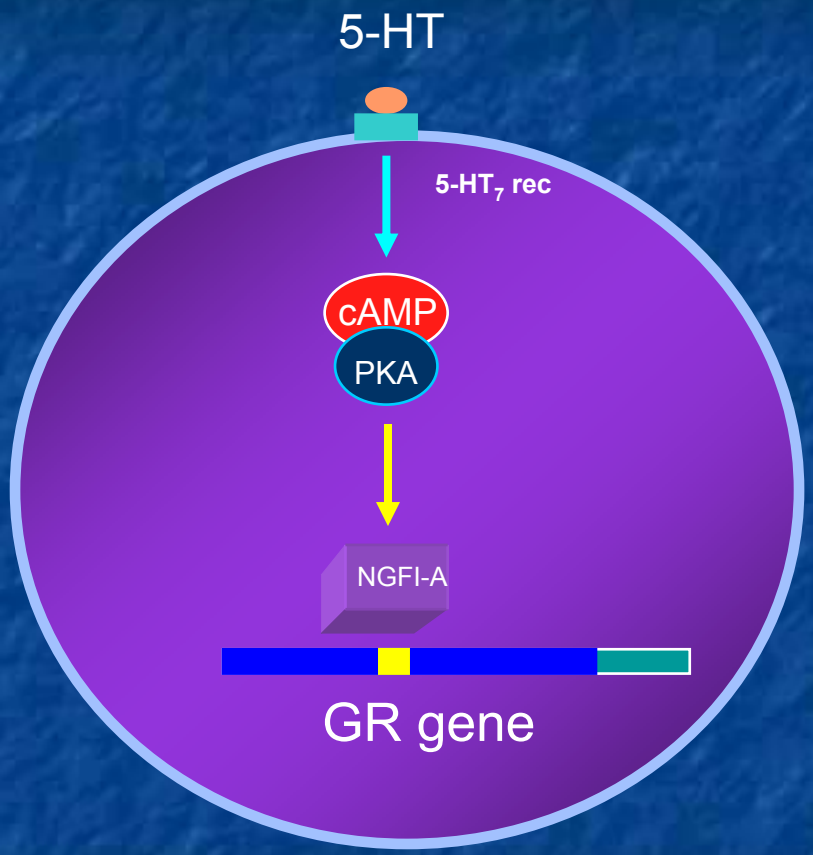
NGFI-A mRNA levels

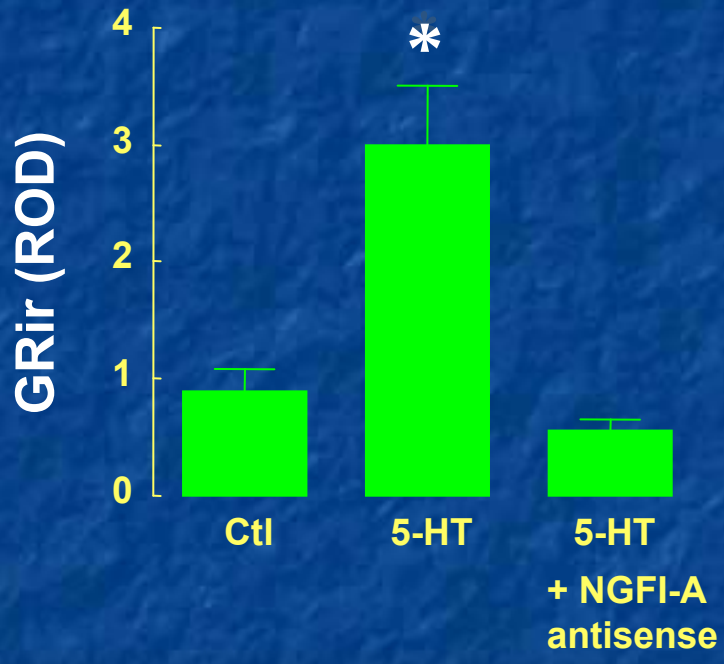


NGFI-A mRNA (stroking)

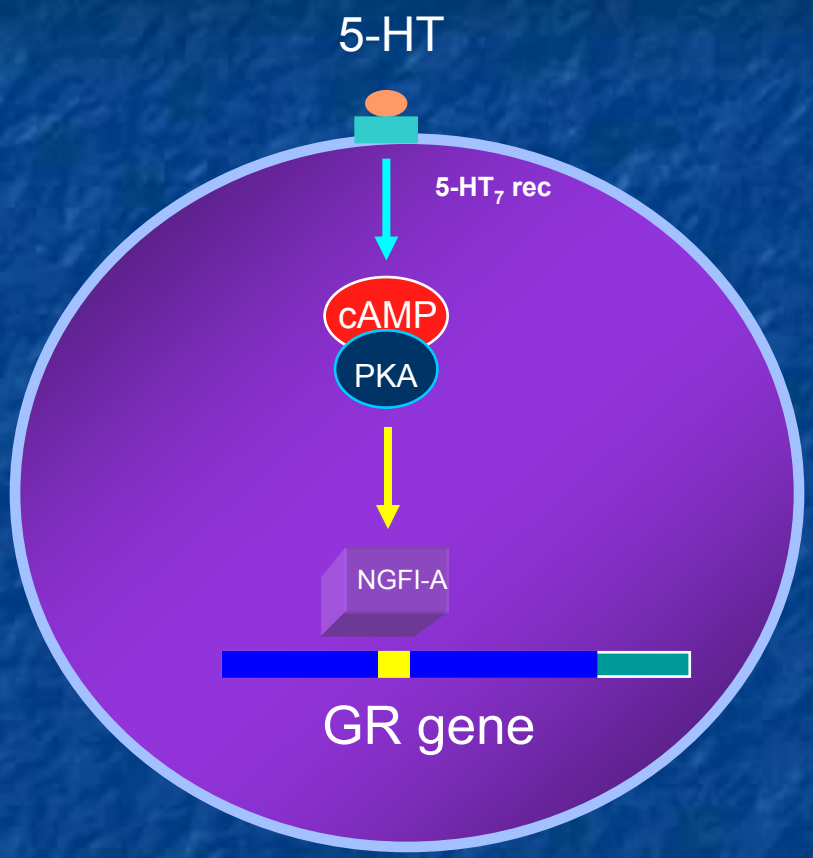


In vitro (primary hippocampal neuronal cultures) studies





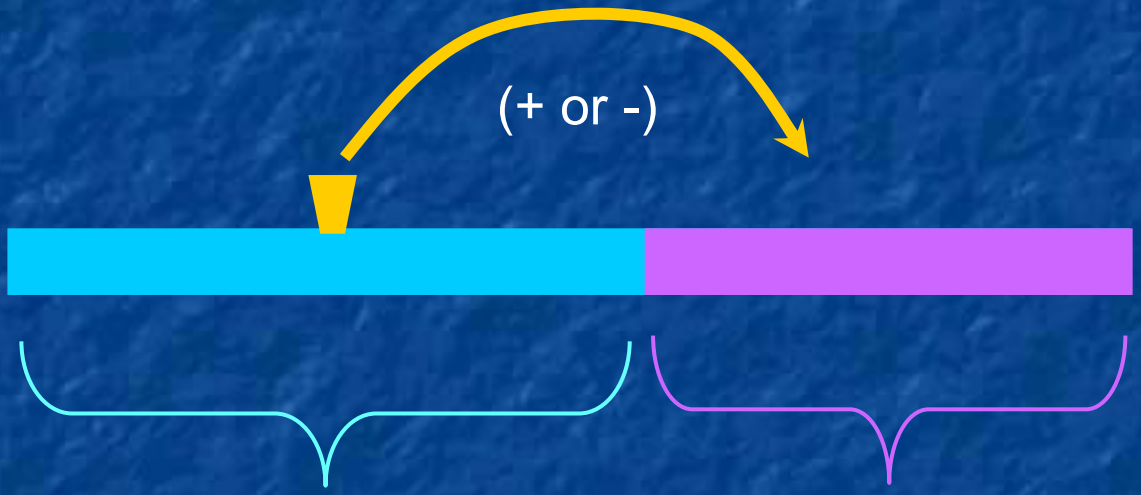
Weaver et al., 2006



What are the relevant genomic targets?

Clone the 5’ untranslated region of the rat hippocampal glucocorticoid receptor gene

Gene organization

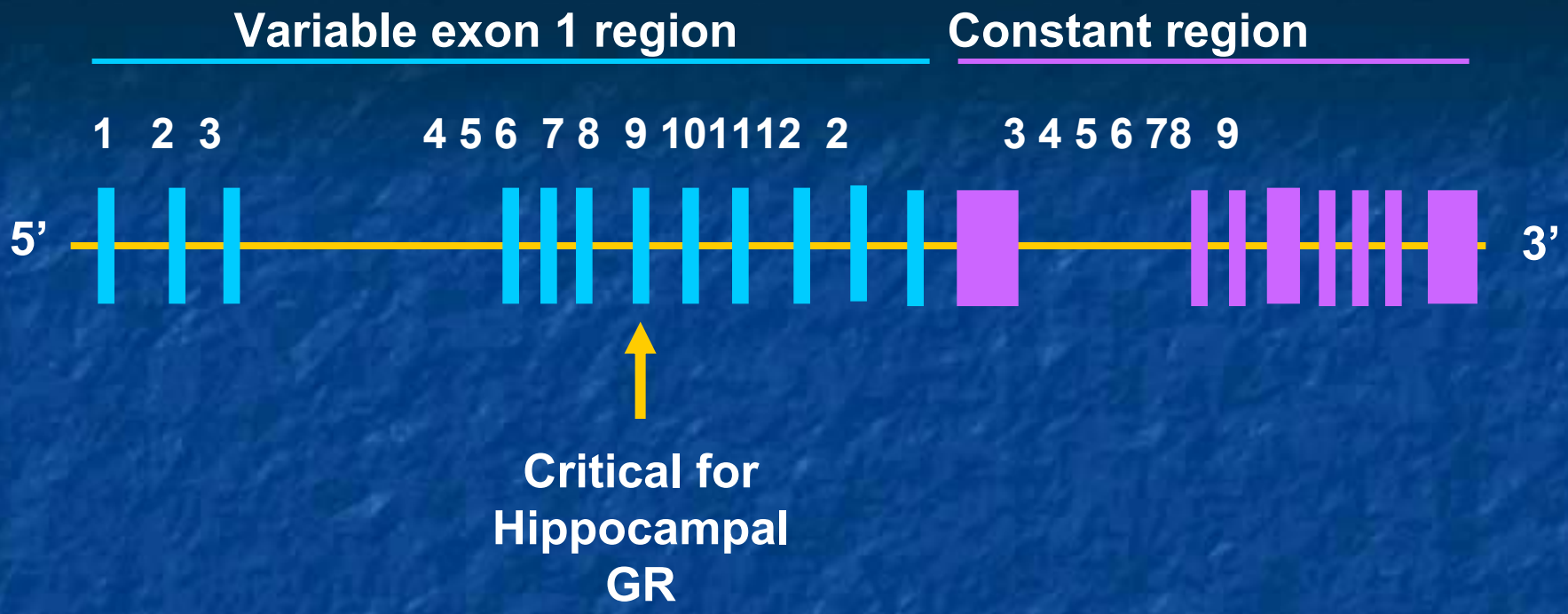


Non-coding, regulatory region (contains enhancers, repressors, etc.).

Coding region responsible for protein synthesis.

Glucocorticoid receptor gene

Clone the 5’ untranslated region of the rat hippocampal glucocorticoid receptor gene



Transfection studies with promoter-reporter constructs reveal exon 1_7 sequence has considerable transactivational capacity.

(McCormick et al. *Mol Endo* 2000)

DNA sites that regulate glucocorticoid receptor gene

GR Promoter 1_7 Sequence

1681 ccc

1741 ctctgctagt gtgacacact t[1]**cg**[2]**cg**caact c[3]**cg**cagttgg [4]**cg**ggg[5]**cg**[6]**cg**ga ccacccctg[7]**c**

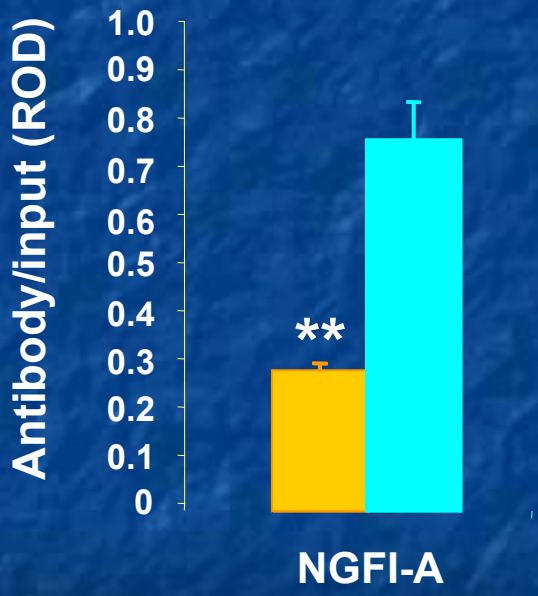
1801 **g**gctctgc[8]**cg** gctggctgtc accct[9]**cg**ggg gctctggctg c[10]**cg**accca[11]**cg** ggg[12]**cg**ggct

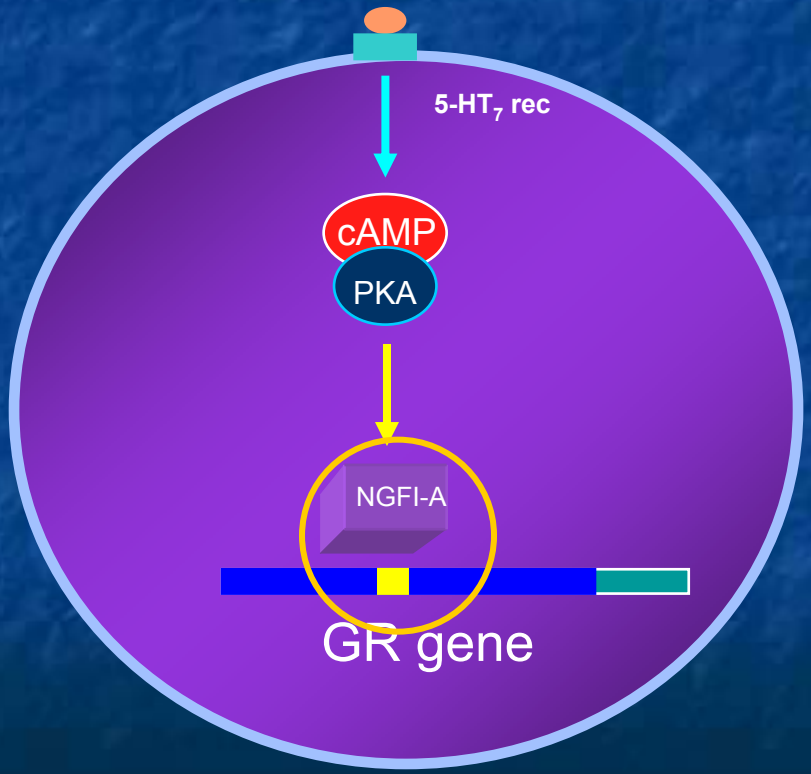
1861 c[13]**cg**ag[14]**cg**gtt ccaagcct[15]**cg** gagtggg[16]**cg** gggg[17]**cg**ggag ggagcctggg agaa

NGFI-A

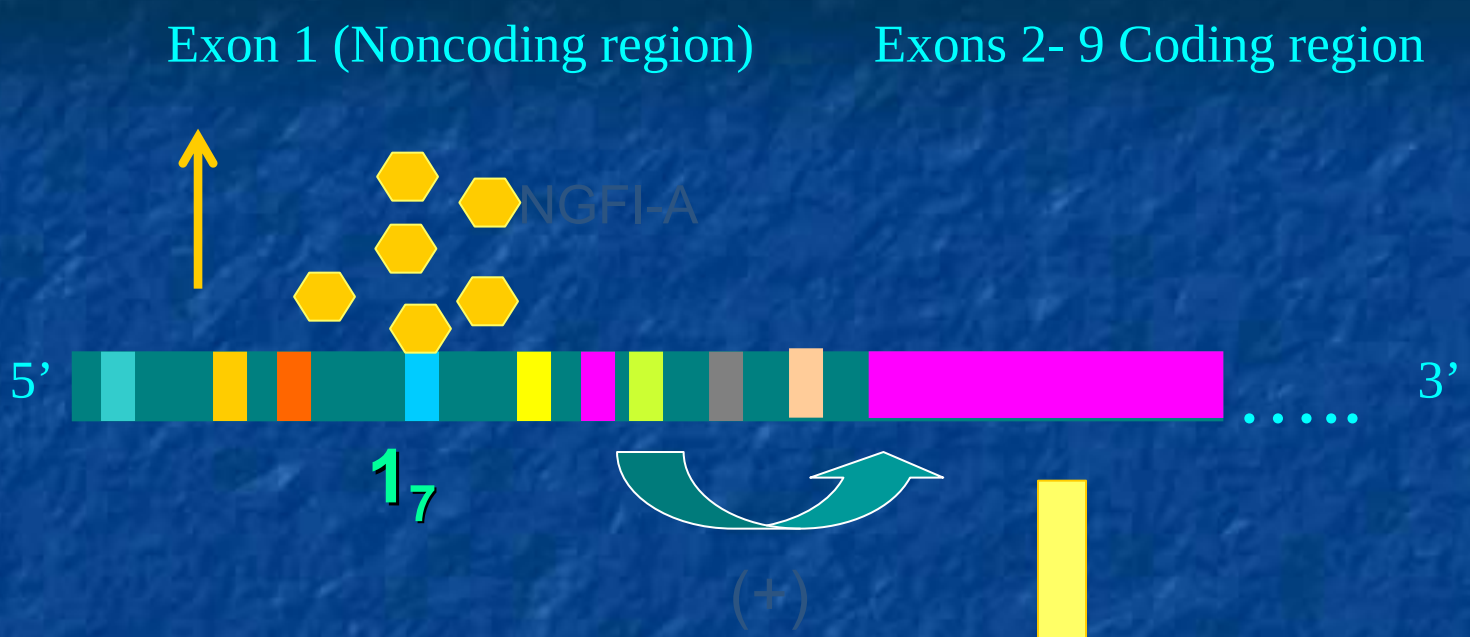
NGFI-A binding to the GR(1_7) promoter in neonates

5-HT





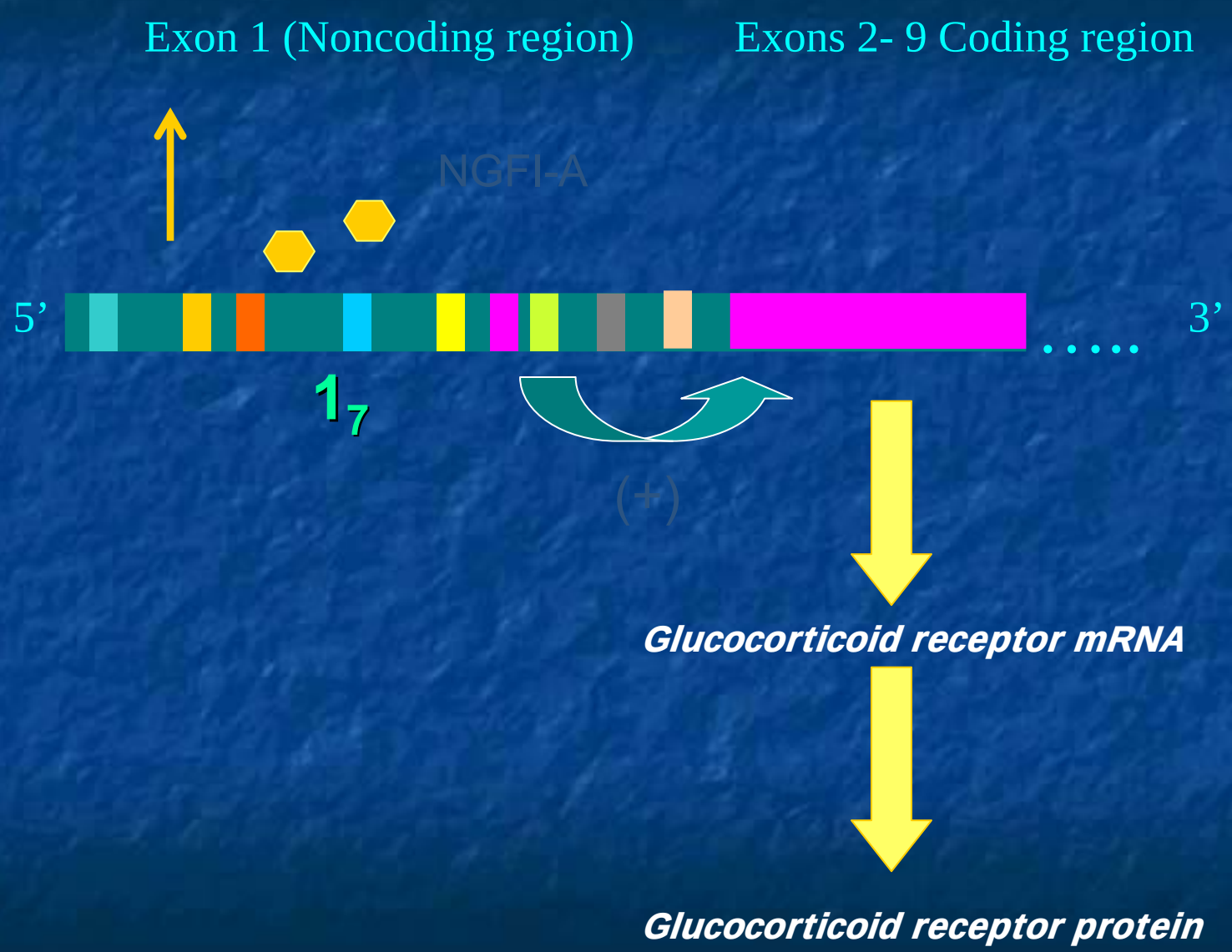
Offspring of High LG mothers



Glucocorticoid receptor mRNA

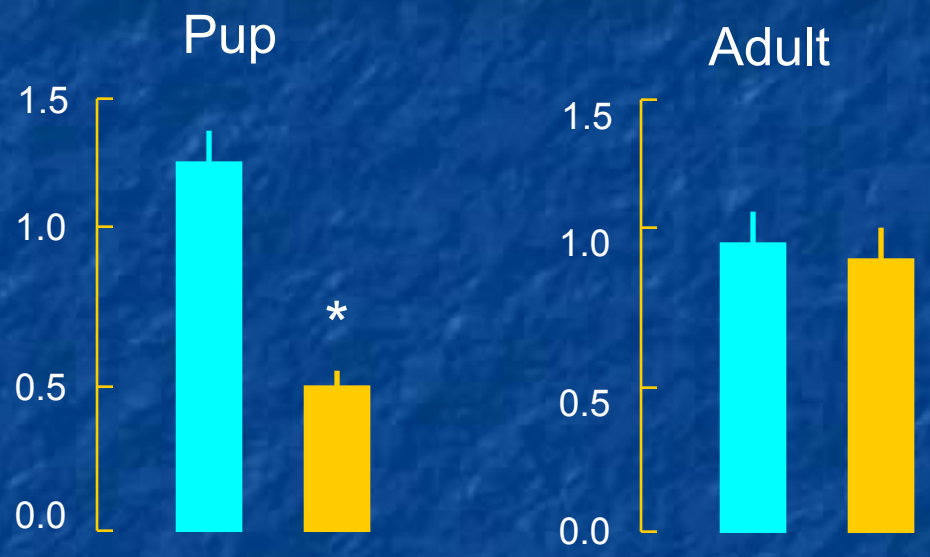
Glucocorticoid receptor protein

Offspring of High LG mothers



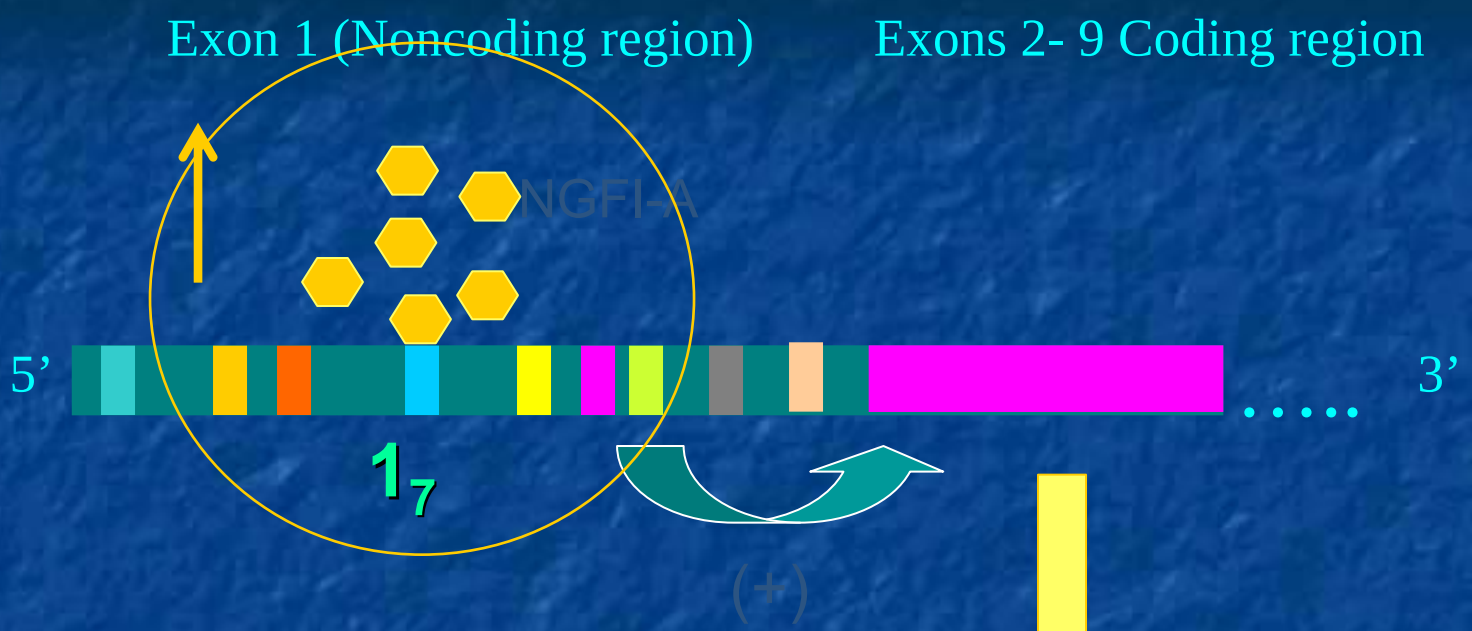
But…..

NGFI-A levels



So, while increased levels of NGFI-A can explain the increased activity of the glucocorticoid receptor gene in the pup, it does not explain why the difference is still observed in adult animals?

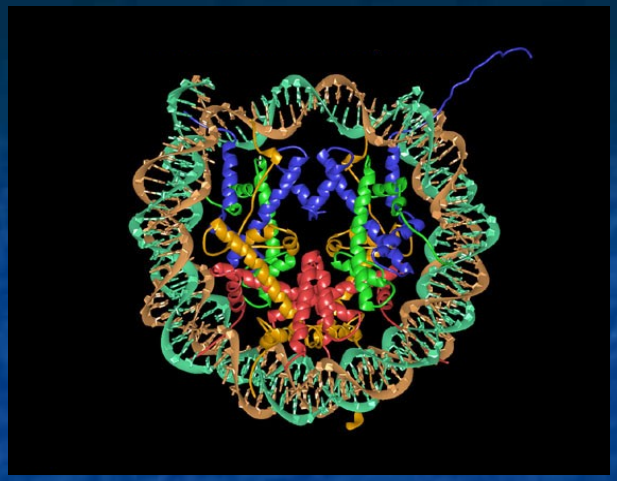
Offspring of High LG mothers



Stable modification of the DNA????

Glucocorticoid receptor mRNA

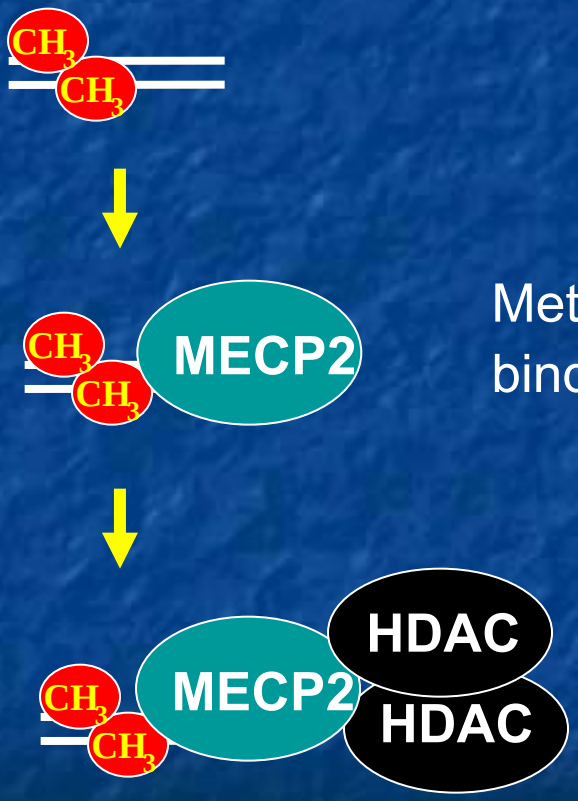
Glucocorticoid receptor protein



GR expression & HPA function

- DNA methylation occurs at cytosines.
- DNA methylation is chemically very stable.

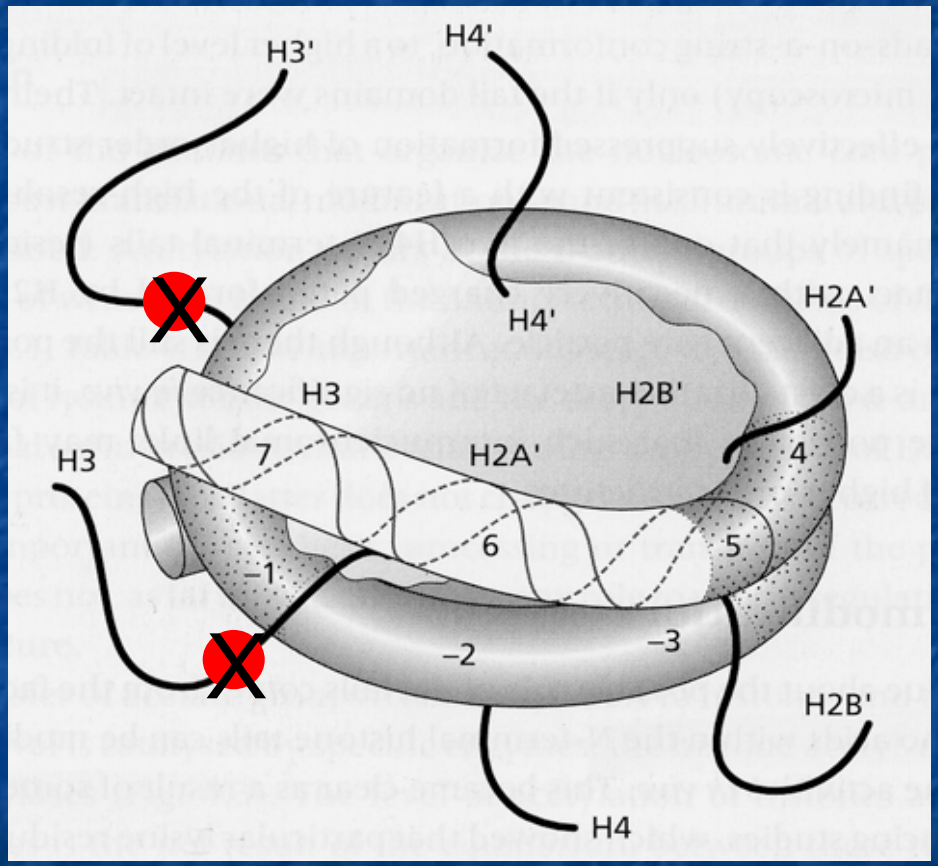
DNA Methylation can inhibit gene expression by blocking transcription factors binding



Methylated DNA binding protein

HDAC: Histone deacetylase

DNA methylation silences gene expression



DNA sites that regulate glucocorticoid receptor gene

GR Promoter 1_7 Sequence

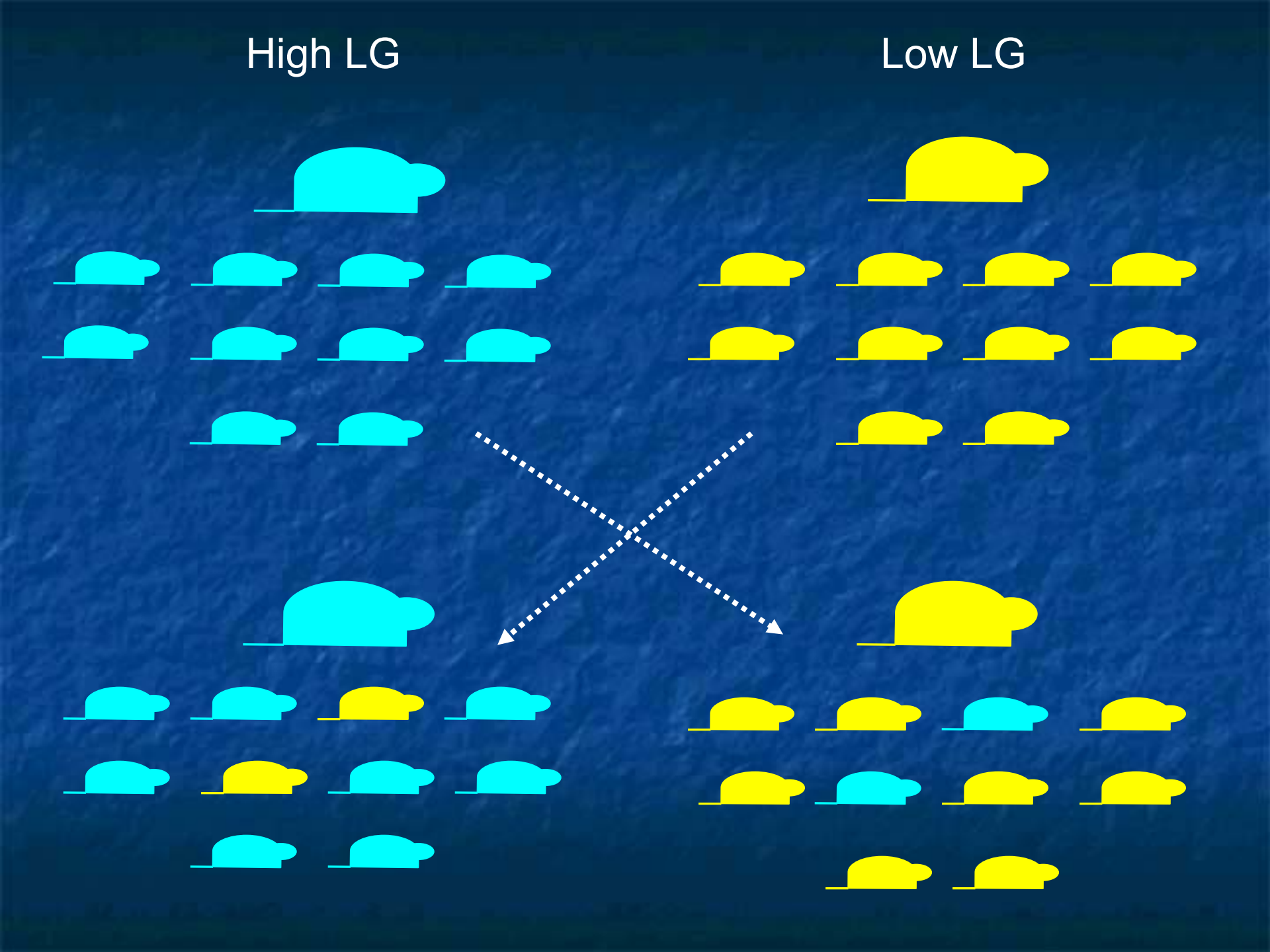
1681 ccc

1741 ctctgctagt gtgacacact t[1]**cg**[2]**cg**caact c[3]**cg**cagttgg [4]**cg**ggg[5]**cg**[6]**cg**ga ccacccctg[7]**c**

1801 **g**gctctgc[8]**cg** gctggctgtc accct[9]**cg**ggg gctctggctg c[10]**cg**accca[11]**cg** ggg[12]**cg**ggct

1861 c[13]**cg**ag[14]**cg**gtt ccaagcct[15]**cg** gagtggg[16]**cg** gggg[17]**cg**ggag ggagcctggg agaa

NGFI-A


High LG
Low LG

NGFI-A binding to ….

5’…T G C G G G G G C G G G G …… 3’

(no methylation)

(High LG)

5’…T G C G G G G G C G G G G …….3’

(CH_3 on C)

(methylated only at 3’; High LG)

5’…T G C G G G G G C G G G G …….3’

(CH_3 on C)

(methylated only at 5’)

(Low LG)

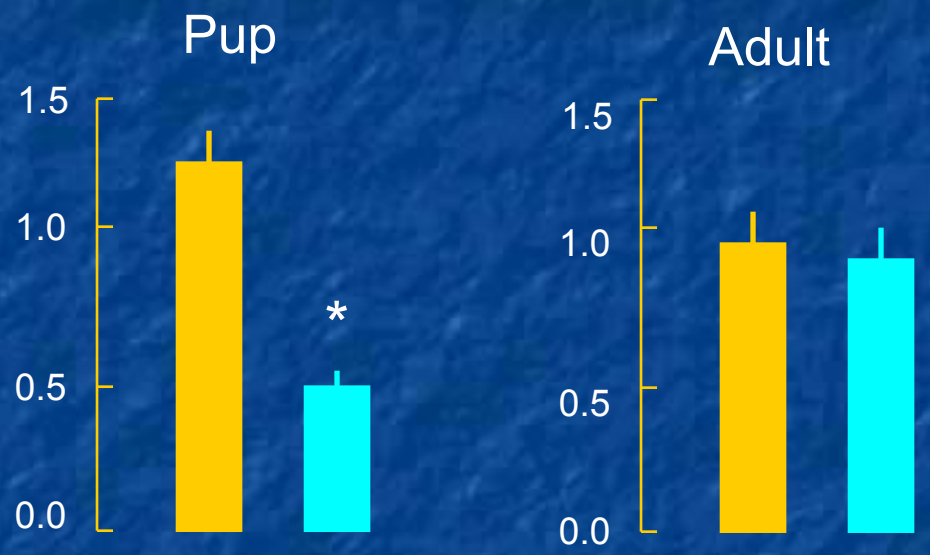
5’…T G C G G G G G C G G G G …… 3’

(CH_3 on C, CH_3 on C)

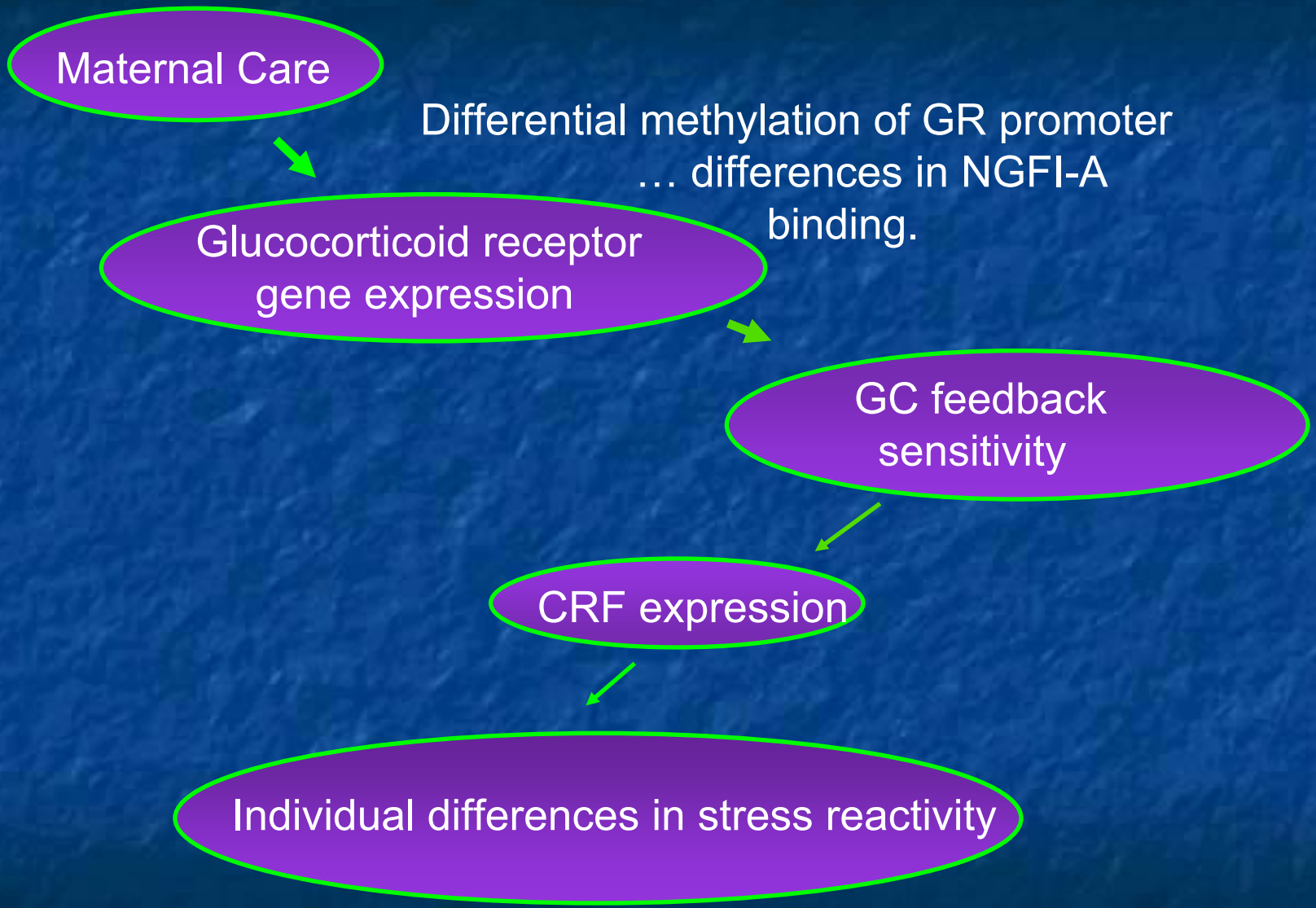
(methylated at both 5’ and 3’; Low LG)

But….

NGFI-A levels



So while levels of NGFI-A are similar in animals reared by High or Low licking/grooming mothers, the NGFI-A site in the adult offspring of Low LG mothers is methylated and therefore cannot interact with NGFI-A.


Maternal Care
Differential methylation of GR promoter … differences in NGFI-A binding.
Glucocorticoid receptor gene expression
GC feedback sensitivity
CRF expression
Individual differences in stress reactivity

Do these ‘maternal’ signals alter the methylation of the exon 17 GR promoter?

Tactile stimulation
(maternal LG)

(H-P-T)

(T3)

5-HT

5-HT7 rec

cAMP

PKA

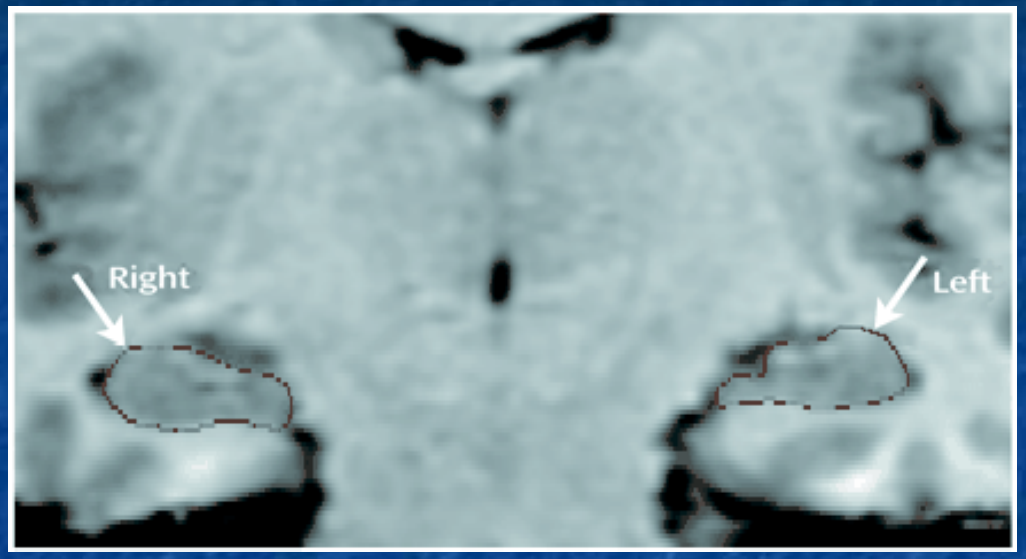
NGFI-A

GR gene

Do comparable processes occur in humans?

- Post-mortem studies of hippocampus.
- Samples from suicide victims/controls.
- QSBB (Gustavo Turecki) - forensic phenotyping.
- Human exon 1F promoter (Turner & Muller, J Molec Endo, 2005)

Hippocampal samples from humans

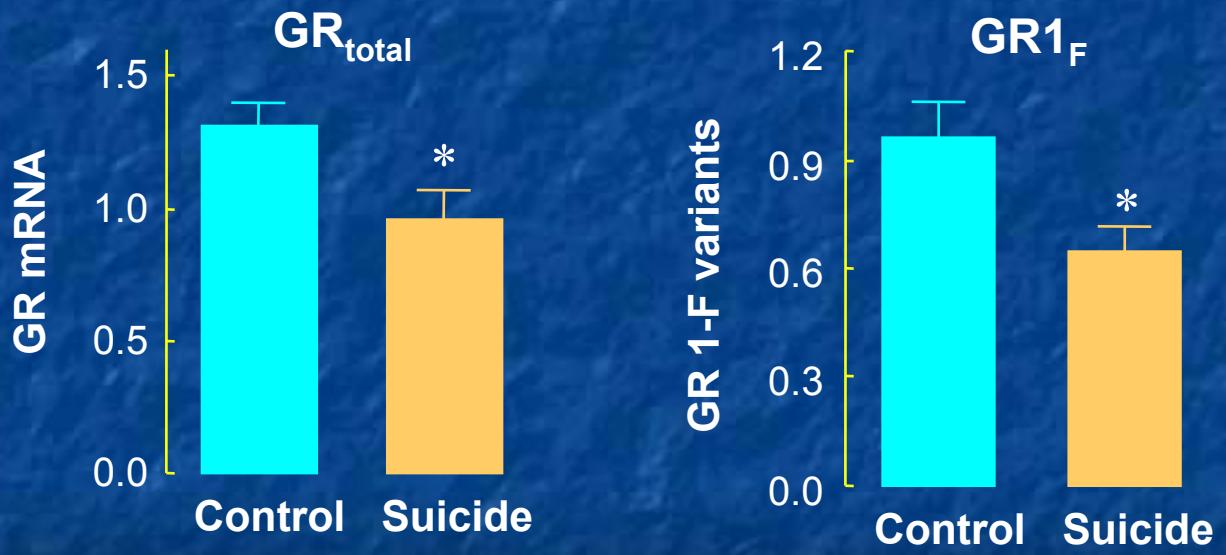


- Human brain bank (suicide victims vs controls).

- All suicide victims (and none of the controls) experienced verified abuse in childhood.

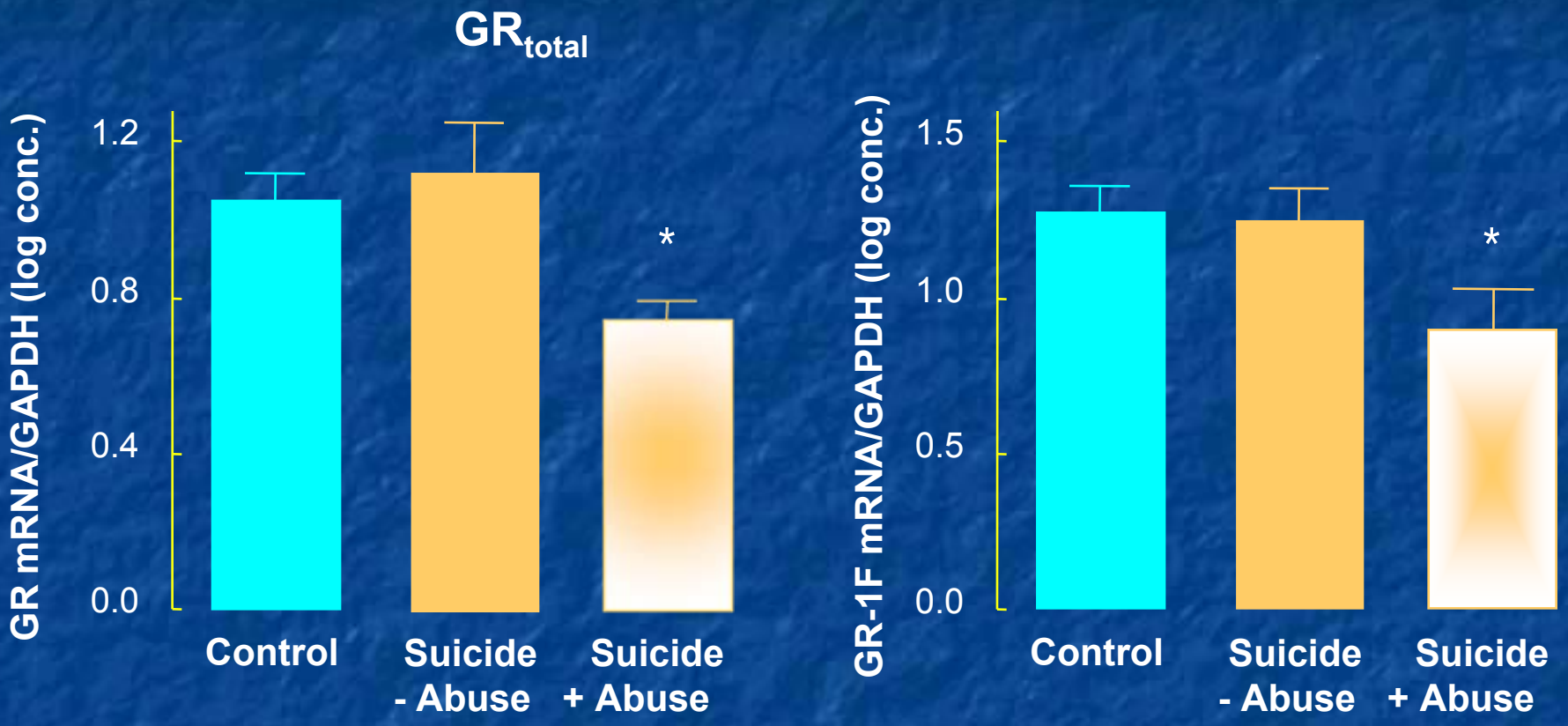
Human glucocorticoid receptor gene

Human glucocorticoid receptor gene



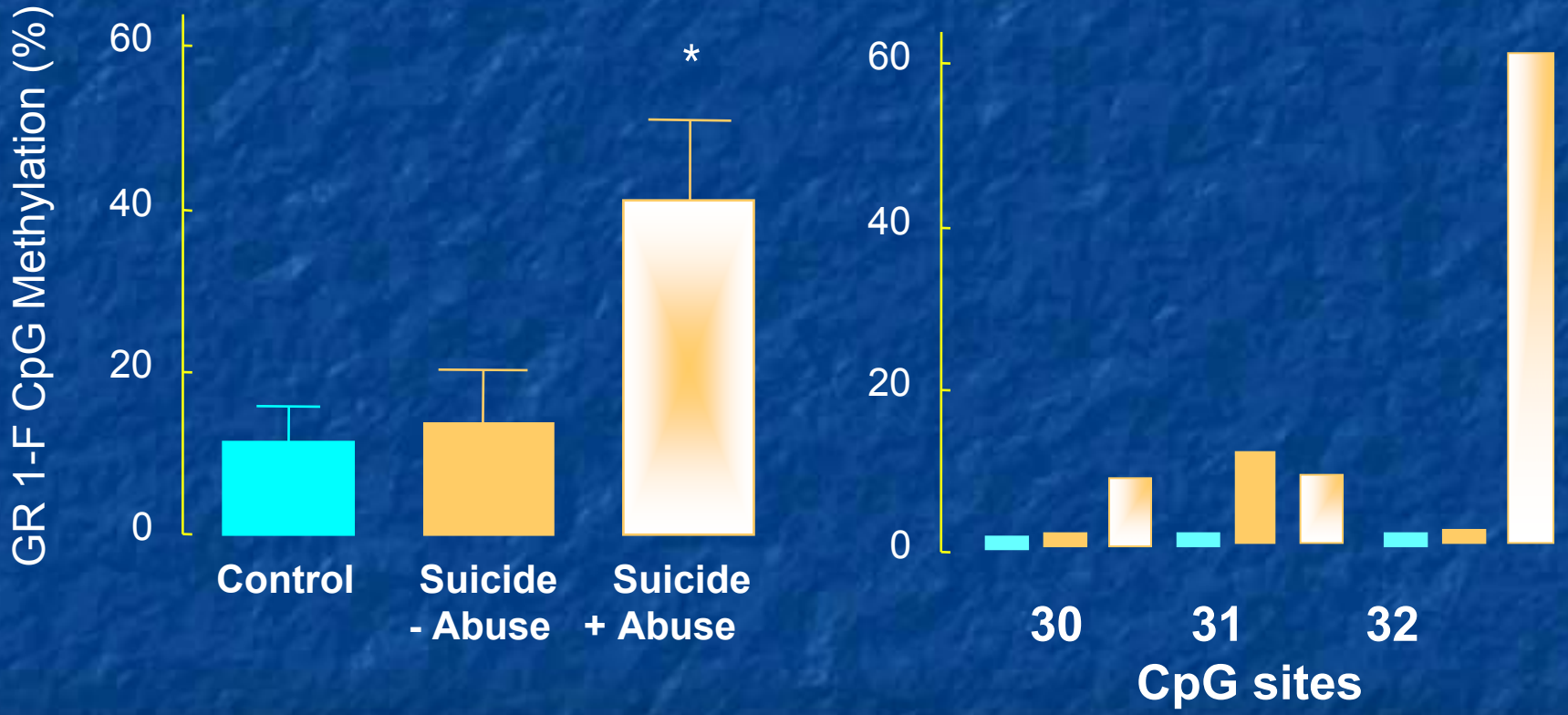
McGowan et al. PlosOne 2008, Nature Neuroscience 2009

Suicide vs abuse - GR expression



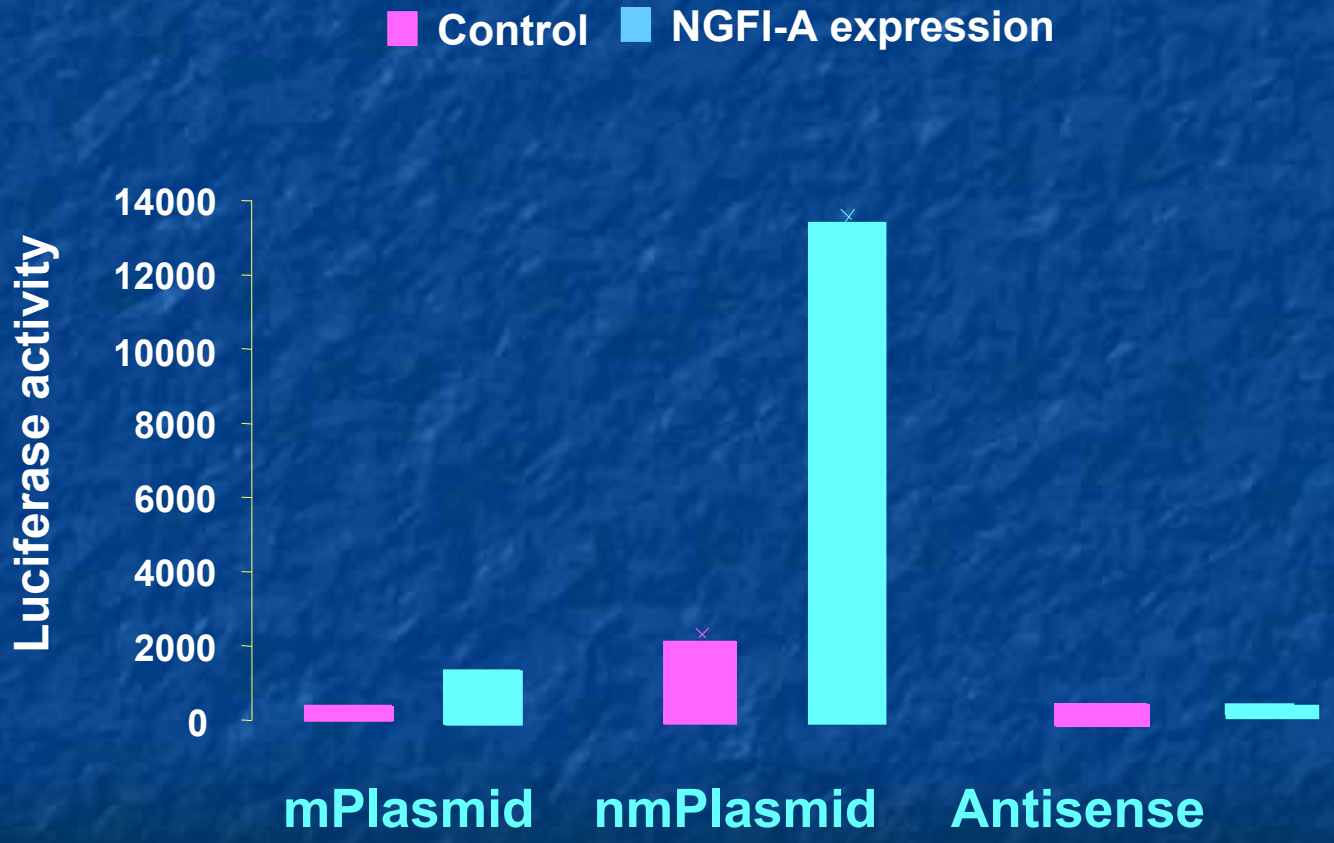
McGowan et al. PlosOne 2008, Nature Neuroscience 2009

Suicide vs abuse - CpG methylation



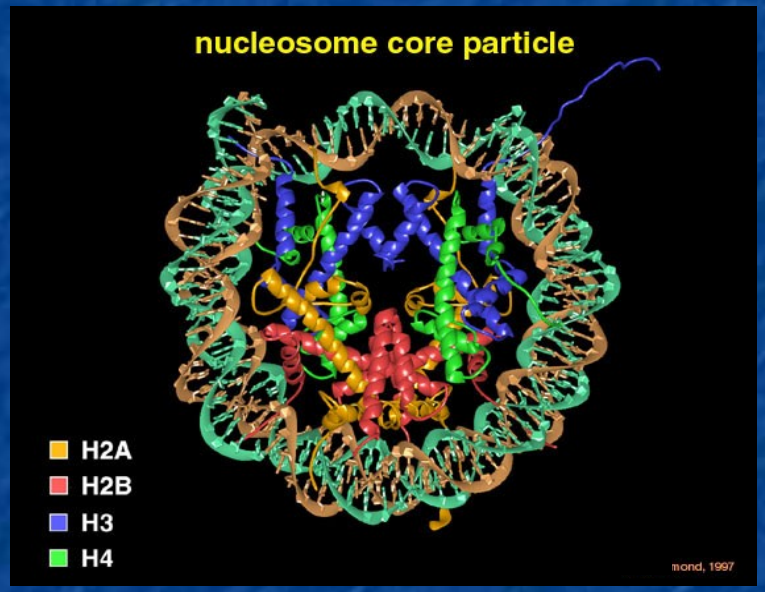
McGowan et al. PlosOne 2008, Nature Neuroscience 2009

Co-transfection studies (NGFI-A vector w/ human GR exon 1F-luciferase construct)



McGowan et al. PlosOne 2008, Nature Neuroscience 2009

DNA methylation serves to imprint social factors,
such as maternal behavior, upon the offspring's genome.



DNA methylation serves as an interface between
the dynamic environment and the fixed genome

Tactile stimulation
(maternal LG)

5-HT

5-HT7 receptor

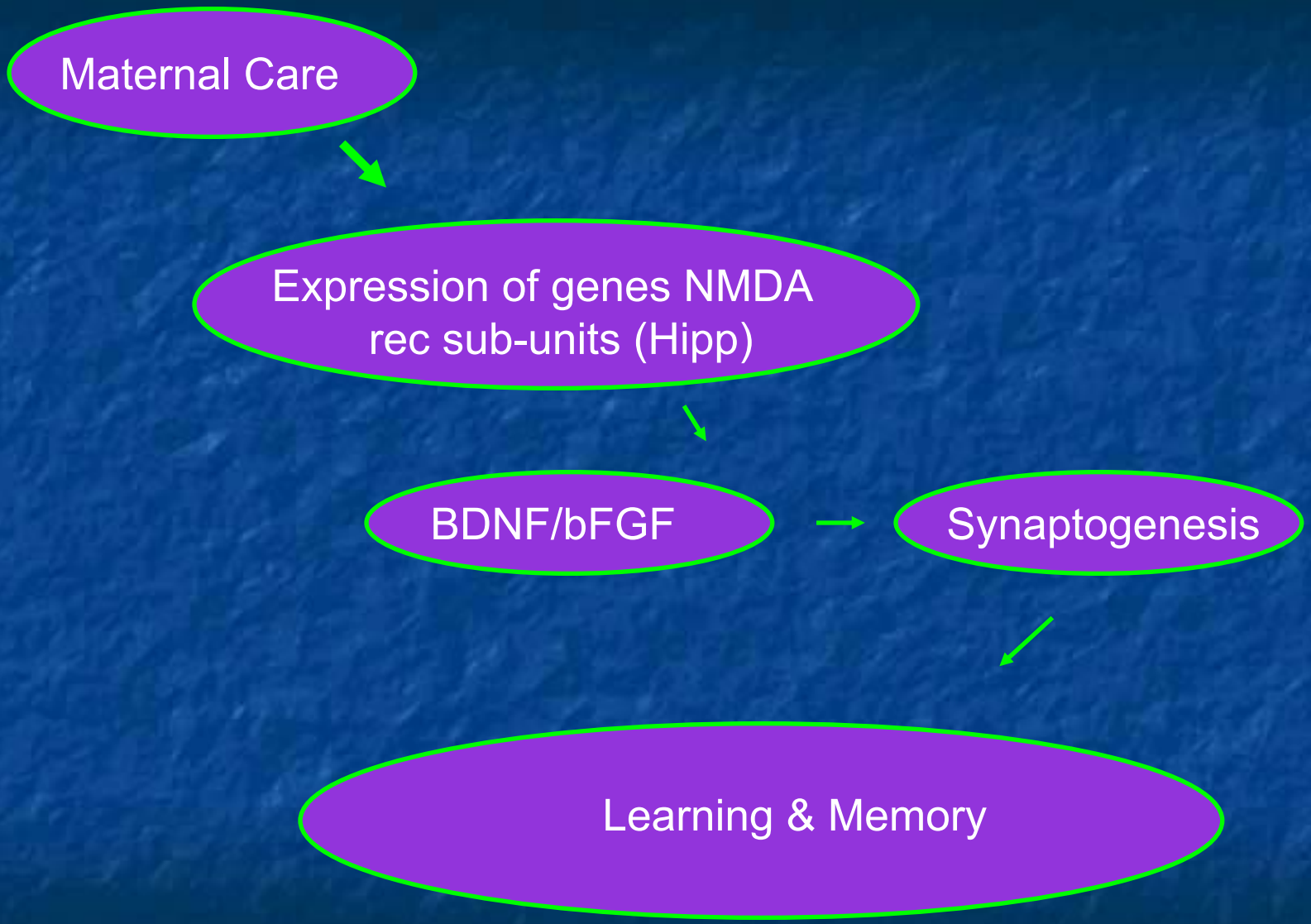
cAMP

PKA

CBP

NGFI-A

Synaptic plasticity


Maternal Care
Expression of genes NMDA rec sub-units (Hipp)
BDNF/bFGF
Synaptogenesis
Learning & Memory