Mood and anxiety disorders

Mood disorders	**10%**
Anxiety disorders	**15-20%**
Comorbidity	**30-60%**

Anxiety Disorders

Generalized anxiety disorder (GAD)
Panic Disorder
Phobia — Social / Specific
Post-traumatic Stress Disorder
Obsessive Compulsive Disorder

Lifetime prevalence = 10-20%

Pharmacotherapy

Mood disorders

- MAOI
- TCA
- SSRI
- Lithium
- CRF, SP antagonists

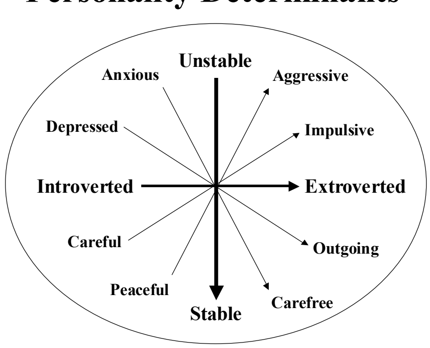
Anxiety disorders

- Benzodiazepines
- TCA
- SSRI
- CRF, SP antagonist

Anxiety and Depression

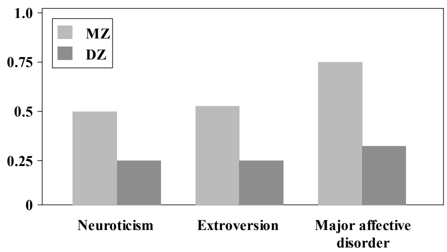
Stimulus	Adaptative response	Pathology
Danger →	Fearful/anxious ⇢	Anxiety disorder
Loss →	Sad/unhappy ⇢	Depression

Personality Determinants



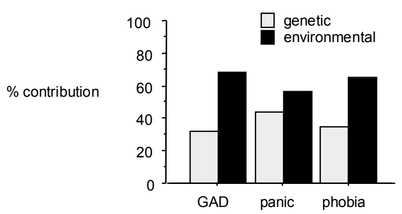
Eysenk (1947)

Twin Studies



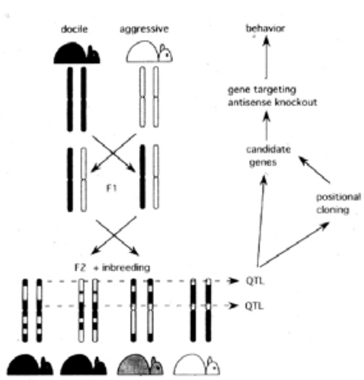
Genetic and Environmental Factors in Anxiety Disorders

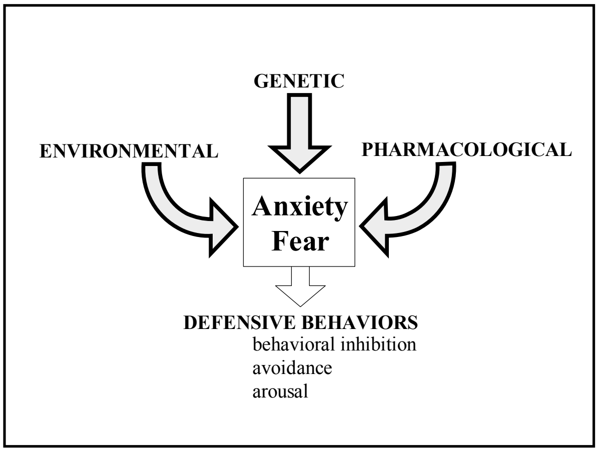
Twin studies

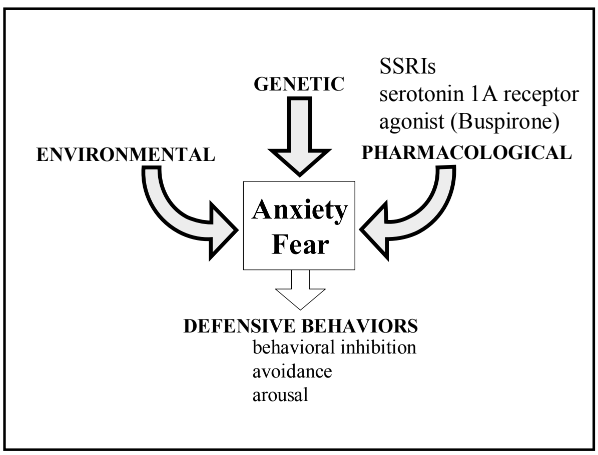


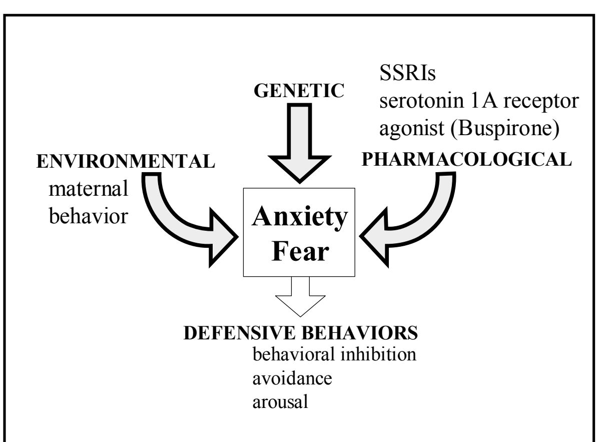
from Kendler et al., 1995

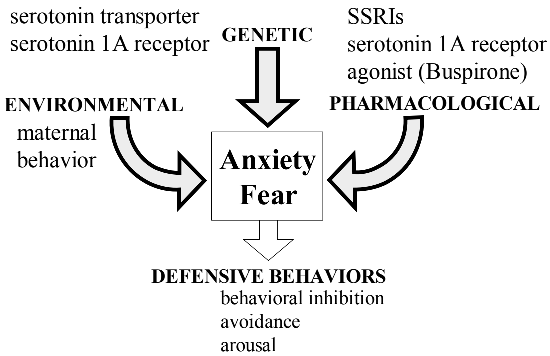
Mouse Genetic Strategies




GENETIC
ENVIRONMENTAL
PHARMACOLOGICAL
Anxiety
Fear
DEFENSIVE BEHAVIORS
behavioral inhibition
avoidance
arousal


SSRIs
serotonin 1A receptor
agonist (Buspirone)
GENETIC
ENVIRONMENTAL
PHARMACOLOGICAL
Anxiety
Fear
DEFENSIVE BEHAVIORS
behavioral inhibition
avoidance
arousal


SSRIs
serotonin 1A receptor
agonist (Buspirone)
GENETIC
ENVIRONMENTAL
maternal
behavior
PHARMACOLOGICAL
Anxiety
Fear
DEFENSIVE BEHAVIORS
behavioral inhibition
avoidance
arousal



Animal Models

	Human	Animal
Emotional stimulus	• **Danger**	**Y**
	• **Loss**	
↓	↓	
Emotional response	• **Behavioral response**	**Y**
	• **Physiological response**	**Y**
	• **Feeling (conscious experience)**	**?**

Validation of Animal Models

Behavioral validation

- Multiple behavioral and physiological tests
- Factor analysis (covariance analysis)

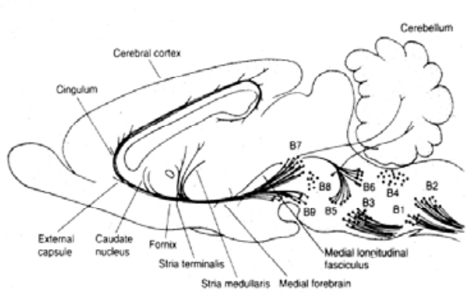
Pharmacological validation

- Response to therapeutically active drugs but not to inactive drugs
- Comparable pharmacokinetic profiles

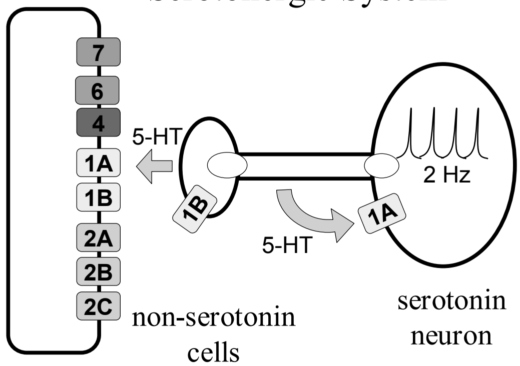
Candidate Systems for Anxiety and Depression

Lesion and stimulation studies	Pharmacological studies	
Amygdala	GABA	Benzodiazepine
Hippocampus	5-HT	5-HT1A Agonist
Hypothalamus		SSRI
Locus coeruleus	CRF, SP, NE	Antagonist
Raphe		

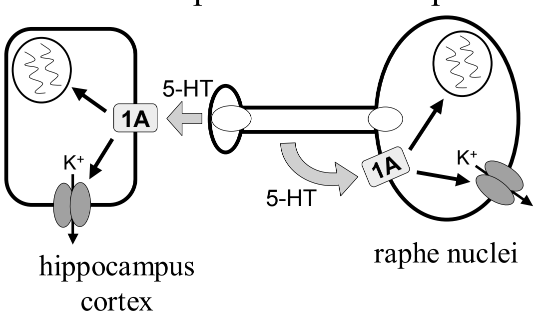
Anatomy of the serotonergic system



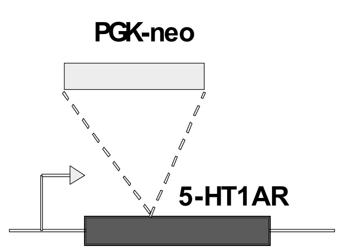
Serotonergic System



Serotonin 1A Receptor heteroreceptor vs. autoreceptor



Construction of 5-HT1AR KO mice



5-HT1A KNOCKOUT

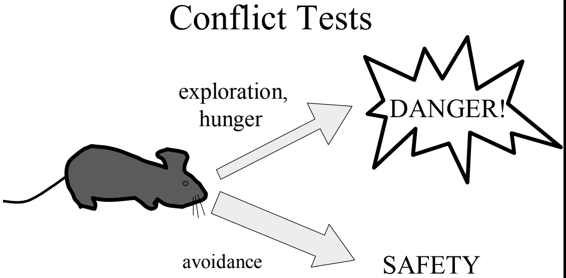
5-HT1A knockout mice

Anxiety-like phenotype

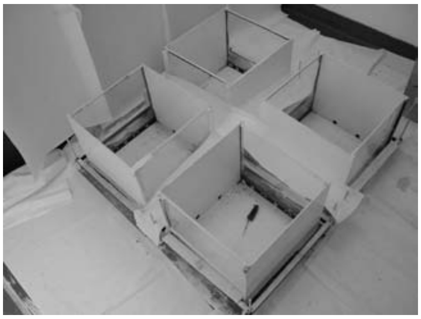
Effects of antidepressants on behavior and neurogenesis

Tissue-specific and developmental rescue

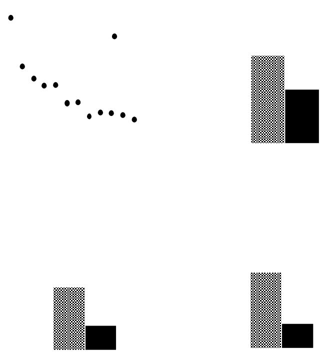
Conflict Tests



Open Field

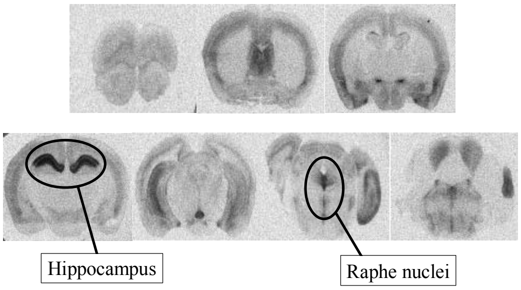


Open Field

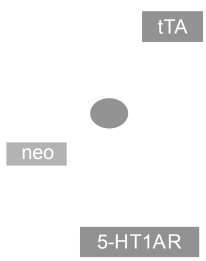


Elevated-Plus Maze

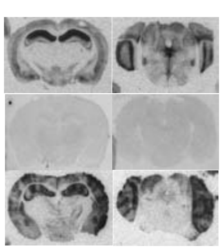
Mouse Serotonin 1A Receptor (5-HT1AR)



Forebrain Rescue Strategy

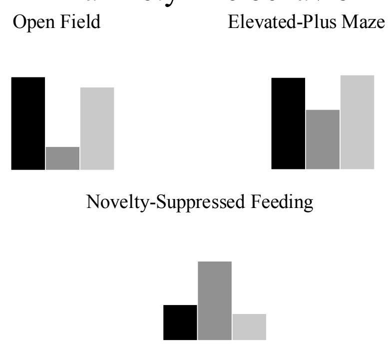


Forebrain Rescue of 5-HT1AR

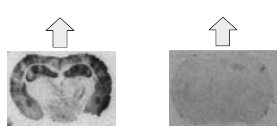


Does forebrain receptor reverse the phenotype of the 5-HT1A KO mice?

Forebrain rescue mice have normal anxiety-like behavior



Receptor OFF in adulthood



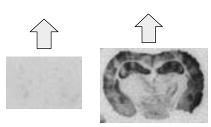
Shutting off receptor expression in the adult does not reverse the rescue phenotype

Open Field

Elevated-Plus Maze

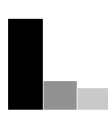
Novelty-Suppressed Feeding

Receptor ON at P21

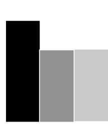


Shutting off receptor expression before P21 reverses the rescue phenotype

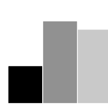
Open Field



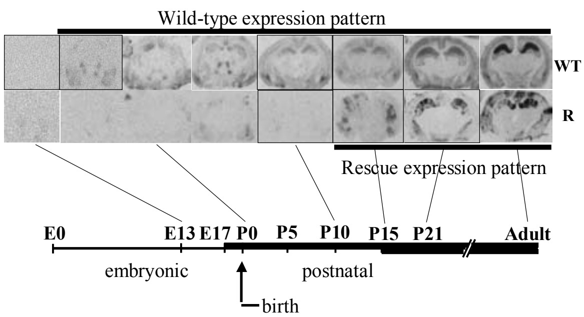
Elevated-Plus Maze



Novelty-Suppressed Feeding



5-HT1AR expression during development



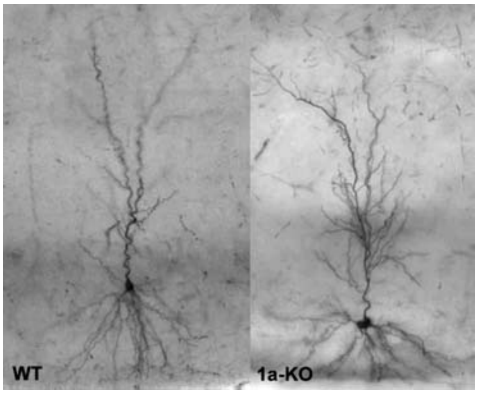
Summary
Forebrain Rescue

- Forebrain receptor sufficient to reverse knockout phenotype
- Adult receptor not required to maintain rescue
- Expression during development critical to rescue

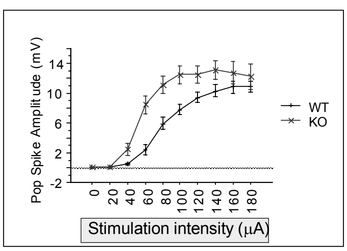
Evidence for developmental role of 5-HT

- 5-HT mediates lifelong effects of altered maternal care (M. Meaney)

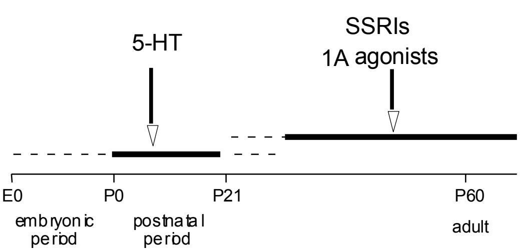
- 5-HT depletion or excess during postnatal period:
  - disrupted barrel and visual cortices
  - reduced hippocampal dendritic spine density (J. Haring)
  - disrupted calretinin-IR neuron morphology (J.-P. Hornung)

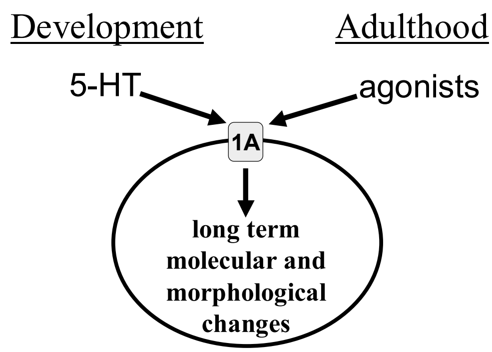


Increased excitability in CA1 pyramidal neurons of knockout mice



5-HT1AR function: development vs. adulthood

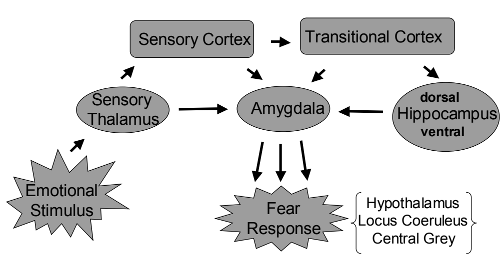




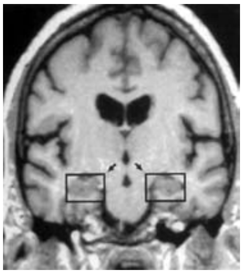
Mechanism of action of antidepressants

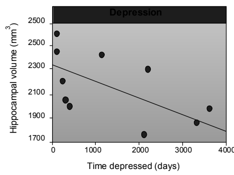
- Change of the setpoint of monoamine transmission
- Plastic changes occurring in the limbic target areas of monoaminergic projections (Hipp., Amy., Ctx)
  - Halt hippocampal atrophy
  - Prevent stress-induced dendritic shrinkage
  - Increase hippocampal neurogenesis

Mood and Anxiety Circuits

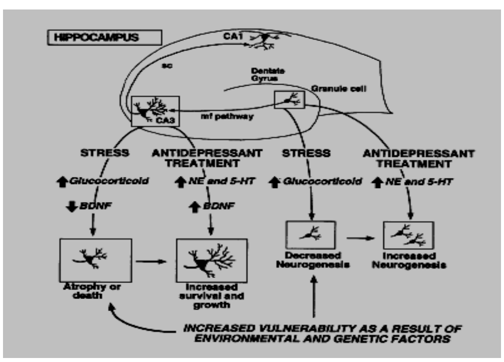


Hippocampal Atrophy In Recurrent Major Depression





Effects of stress and antidepressants on hippocampal plasticity



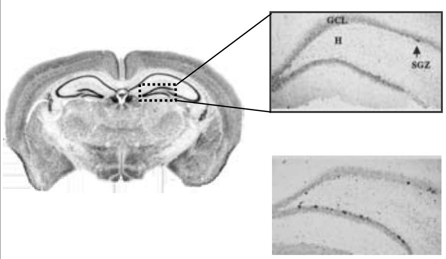
Adult neurogenesis: overturning the dogma

Species	Year	Scientist
Rodent	1965	Altman and Das
Song bird	1980	Nottebhom
Primate	1990s	Gould and McEwen
Human	1998	Eriksson and Gage

Factors that affect adult neurogenesis

	Increase	Decrease
Environment	Enrichment	Stress
Learning	Task acquisition	--
Drugs	Antidepressants	Opiates

Fluoxetine (Prozac) increases neurogenesis in the hippocampus



Control

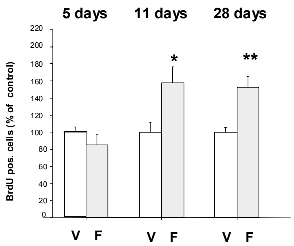
fluoxetine (Prozac)

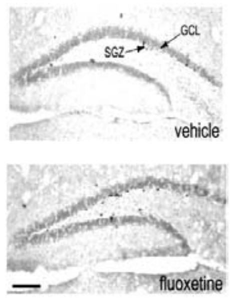
All Antidepressants Increase Hippocampal Neurogenesis

Tricyclics
SSRIs
Lithium
ECT
NK1 antagonist
Vasopressin V1B antagonist

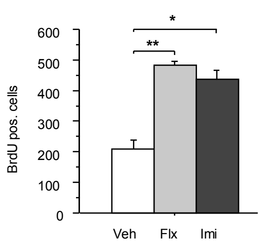
Running, Enriched Environment

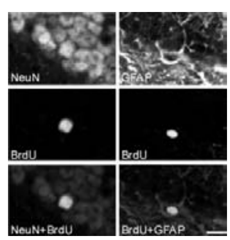
Chronic fluoxetine stimulates neurogenesis in the dentate gyrus





Survival and Differentiation of BrdU positive cells





NeuN: 70%
GFAP: 15%
ND: 15%

Strategies to block hippocampal neurogenesis

Pharmacological: drugs that block cell division (MAM)

Genetic:
1. Mutation in serotonin pathway: 5-HT1A KO mice
2. Selective and regulatable ablation of adult DG progenitor cells

Radiological: selective X-irradiation of a portion of the brain containing the hippocampus

Behavioral tests to detect antidepressant action

Models of Acute response

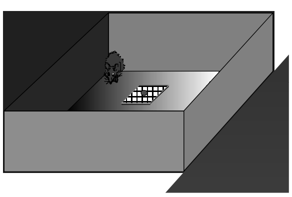
- Tail suspension test
- Forced swimming test
- Ultrasonic vocalization

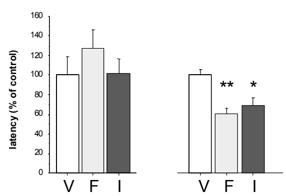
Models of Chronic response

- Learned helplessness
- Chronic unpredictable stress
- Novelty-suppressed feeding

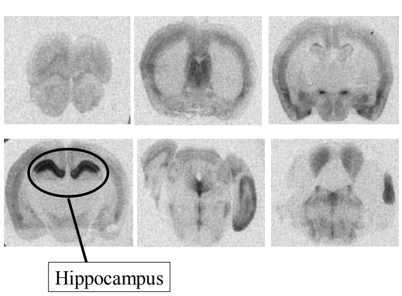
Behavioral Test to Detect Chronic Antidepressant Action

Novelty-Suppressed Feeding (NSF)

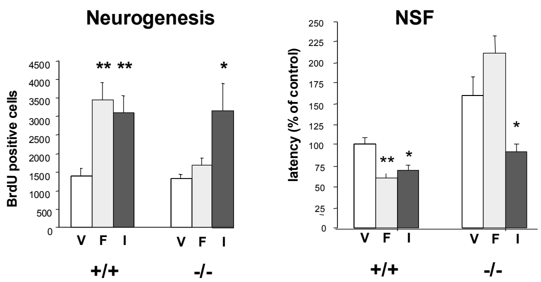




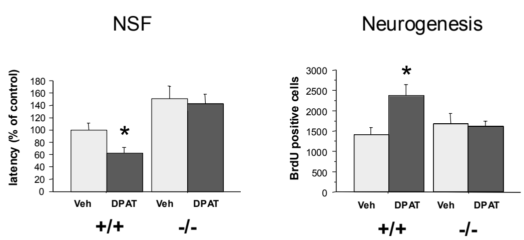
Mouse Serotonin 1A Receptor (5-HT1AR)



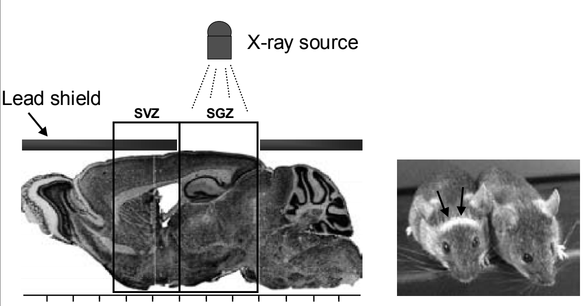
5-HT1A KO mice are insensitive to the neurogenic and behavioral effects of chronic fluoxetine



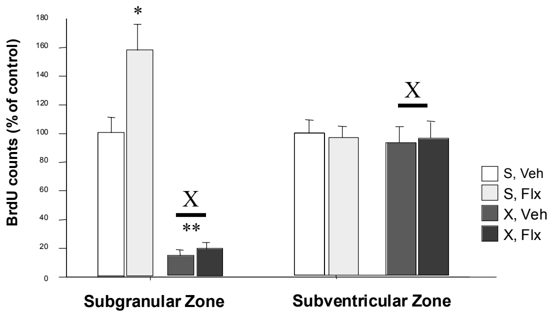
Behavioral and neurogenic effect of chronic 5-HT1A agonist



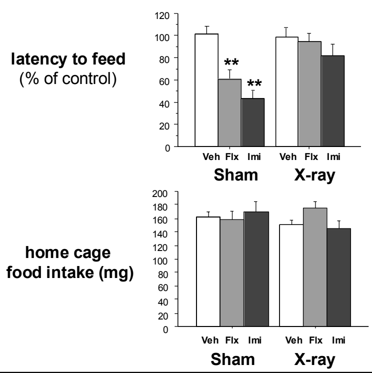
Hippocampus-specific X-ray delivery



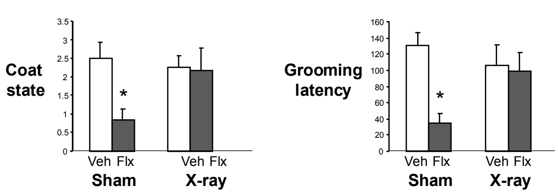
Hippocampus-specific suppression of neurogenesis



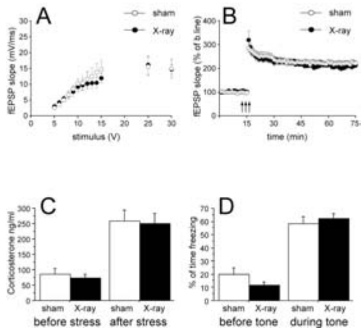
Irradiation prevents the effects of antidepressants in the Novelty-suppressed feeding test



Irradiation prevents the effects of antidepressants in the Chronic Unpredictable Stress paradigm



X-Ray treatment does not affect CA3/CA1 physiology, stress response and cued fear conditionning



Summary

All antidepressants stimulate hippocampal neurogenesis : marker or cause?

Stimulation of hippocampal neurogenesis is necessary for antidepressant effects

Targeting directly the mechanisms underlying hippocampal neurogenesis may lead to novel antidepressants

Motor System Disease

Degeneration

Dysfunction

Motor System Disease

Degeneration | Dysfunction

- Parkinson’s Disease
- Huntington’s Disease
- A.L.S. (Lou Gherig’s Disease)

Motor System Disease

Degeneration

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- Huntington’s Disease
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Dysfunction

- Primary Dystonia
- Tourette’s Syndrome
- Essential Tremor

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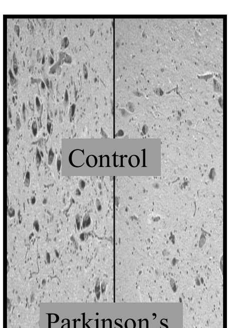
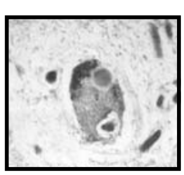
Dysfunction
- Primary Dystonia
- Tourette’s Syndrome
- Essential Tremor

Parkinson’s Disease: Clinical Characteristics

- Disease of aging:
  - -Incidence: Overall 20/100,000
    Age 70 120/100,000
- Clinical Features: tremor, rigidity, slowness of movement, postural instability
- Pathology: substantia nigra pars compacta, locus coeruleus, nucleus basalis of Meynert, olfactory bulb



Substantia Nigra


Lewy Body

Parkinson's Disease: Environment

- 1918: influenza pandemic

- 1983: MPTP

MPTP Induced Parkinsonism

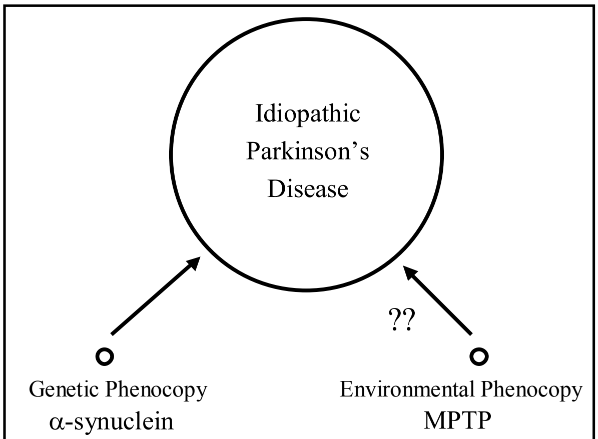
- Clinical: All cardinal manifestations of idiopathic PD.
- Pharmacology: Respond to dopaminergic drugs
- Pathology: Ventral substantia nigra, locus coeruleus, hypothalamus
- Pathogenesis: Mitochondrial dysfunction, oxidative stress

Parkinson's Disease: Genetics

Locus	Inheritance	Onset	Gene
PARK1	Dominant	40's	α-Synuclein
PARK2	Recessive	20's	Parkin
PARK3	Dominant	60's	??
PARK4	Dominant	30's	??
PARK5	Dominant	50's	Ubiquitin C-terminal hydrolase L1
PARK6	Recessive	20's	??
PARK7	Recessive	20's	??

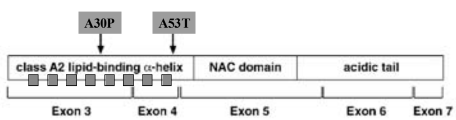
Parkinson's Disease and α-Synuclein

- Mutations produce autosomal dominant PD
- Abundant in Lewy bodies
- Present in Glial Cytoplasmic Inclusions (MSA)
- Accumulates in Hallevordan-Spatz disease

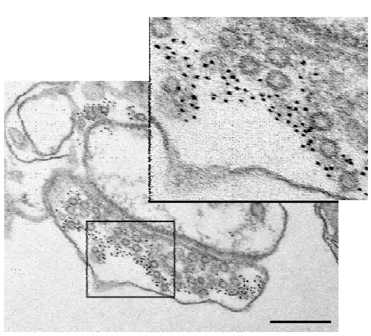


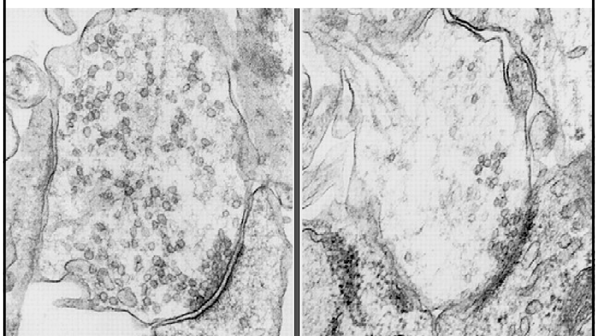
α-Synuclein Biology

- Highly abundant in CNS
- Concentrated in presynaptic terminals near vesicles
- Lipid and vesicle binding
- Conformational change upon lipid binding
- Modulation of rate of recycling of the readily releasable pool



α-Synuclein Function: Vesicles





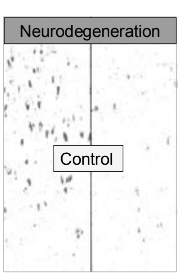
Control α-Synuclein Anti-Sense

What is the nature of α-synuclein dysfunction that leads to dopamine neuron degeneration ?

α-Synuclein

Altered Normal Function ??

?? Novel Toxic Function



α-Synuclein

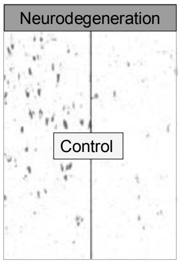
Altered Normal Function

\+ -

??

??

Novel Toxic Function



α-Synuclein

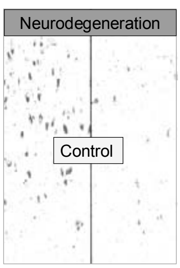
Altered Normal Function

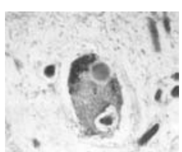
\+ -

??

??

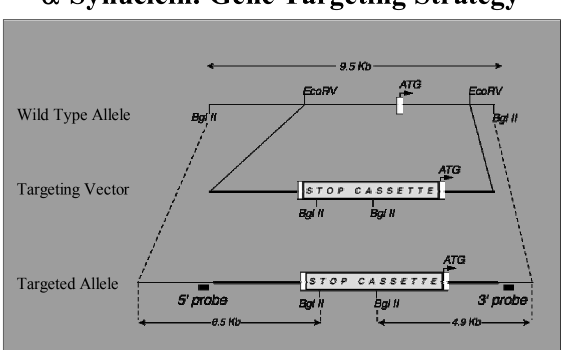
Novel Toxic Function



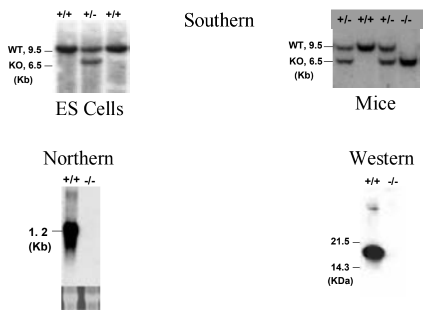


Lewy Body

α-Synuclein: Gene Targeting Strategy



α-Synuclein: Knock Out Characterization



MPTP Regimens

Acute — *NECROSIS* → Sacrifice After 7 Days

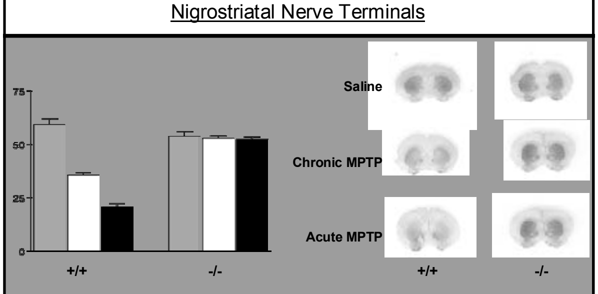
80 mg/kg

Chronic — *APOPTOSIS* → Sacrifice After 21 Days

30 mg/kg x 5 days

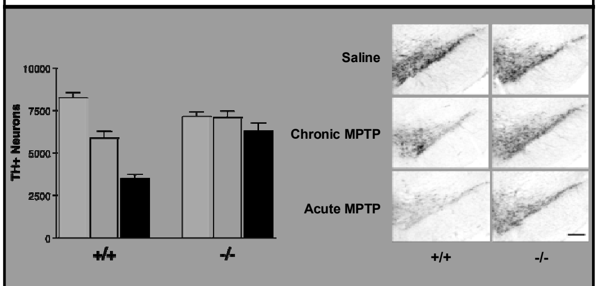
α-Synuclein Null Mice: Resistance to MPTP-Induced Neurodegeneration

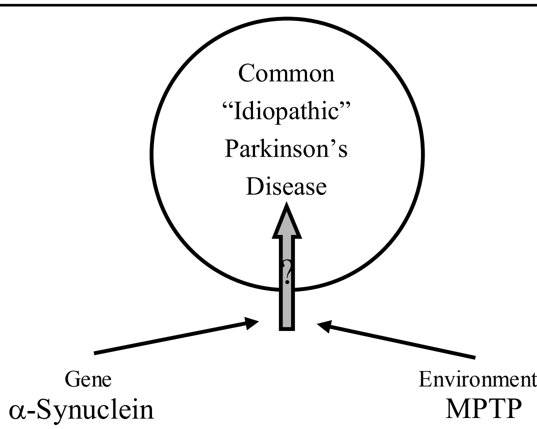
Nigrostriatal Nerve Terminals

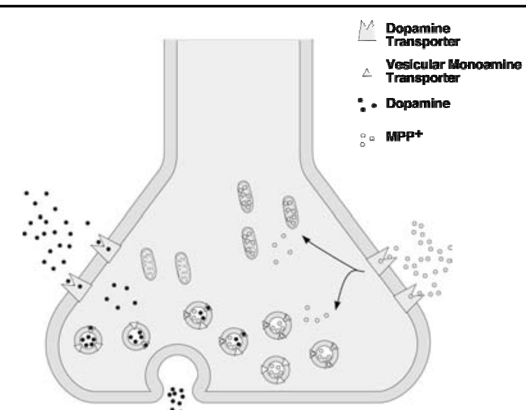


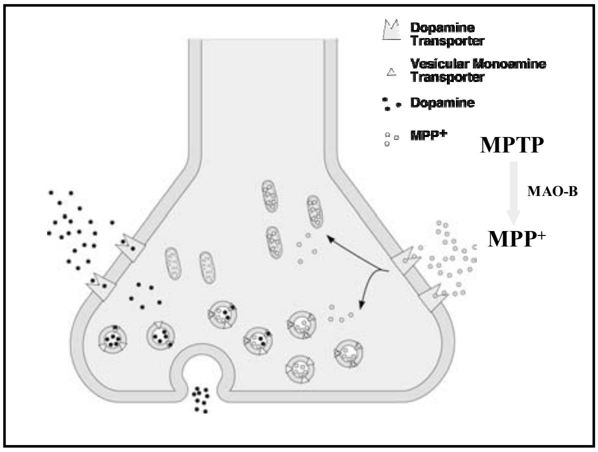
α-Synuclein Null Mice: Resistance to MPTP-Induced Neurodegeneration

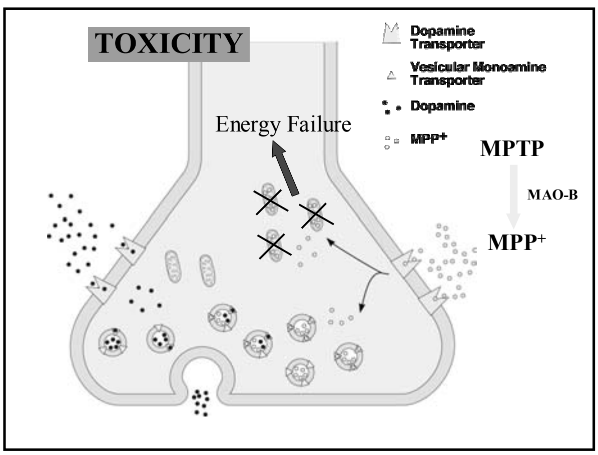
Nigrostriatal Cell Bodies

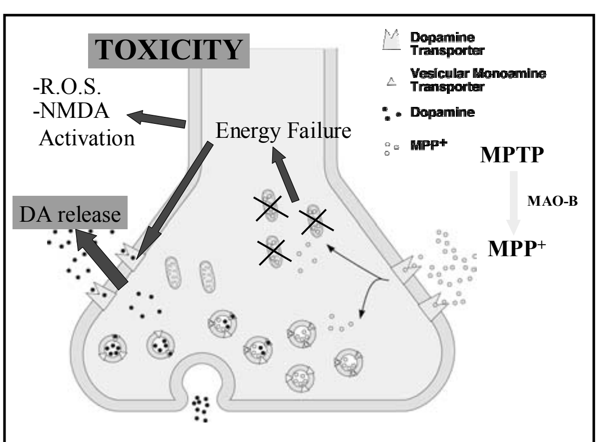







Dopamine Transporter
Vesicular Monoamine Transporter
Dopamine
MPP+
MPTP
MAO-B
MPP+


TOXICITY
Dopamine Transporter
Vesicular Monoamine Transporter
Dopamine
MPP+
Energy Failure
MPTP
MAO-B
MPP+


TOXICITY
Dopamine Transporter
Vesicular Monoamine Transporter
Dopamine
-R.O.S.
-NMDA
Activation
Energy Failure
MPP+
MPTP
DA release
MAO-B
MPP+

An Inducible Model of HD

A brief introduction to HD (1):

- George Huntington, 1872
  - Age of onset: 40 to 50 yrs
  - Progressive
    - Motor: chorea, dystonia
    - Psychiatric: depression, anxiety, suicide
    - Cognitive changes: declarative memory, dementia
  - Autosomal dominant inheritance

Striatal specific atrophy of HD

A brief introduction to HD (2):

- The *HD* gene, IT15 (HDCRG, 1993)
  - Promoter: house keeping gene?
  - Conserved to *Fugu* puffer fish
  - Gene KO studies
    - Nasir et al., Zeitlin et al., Duyao et al.
- Gene product, huntingtin (htt)
  - Large protein in complex
  - Function: unknown
  - Ubiqutious expression
  - Cleavage by calpain and caspase-3

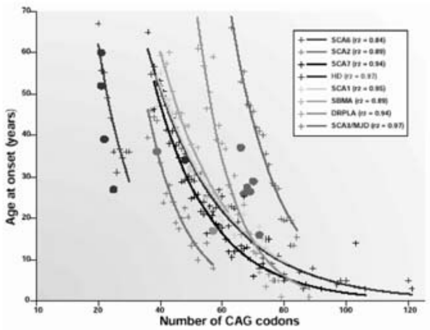
A brief introduction to HD (3):

- Triplet repeat expansion (C-A-G)
  - Unaffected, n= 4 to 35; Pathogenic, n > 37
  - Triplet repeat disorders (17)

Trinucleotide repeat disorders. Noncoding repeats (4) and coding repeats (11).

Disease	Gene locus	Gene product	Inheritance pattern	Repeat	Normal length	Expanded length	Affected region	Citation
Fragile X	Xq27.3	FMR1	X-linked dominant	CGG	6-52	230- 1000	Brain enlargement, connective tissue	Oberle et al., 1991.
Fragile XE	Xq28	FMR2	X-linked sominant(?)	GCC	7-35	230-750	Brain enlargement, connective tissue	Knight et al., 1994; Parrish et al., 1994.
Myotonic Dystrophy (DM)	19q13	DMPK	Dominant	CTG	5-37	50-3000	Wide range of organ systems.	Harley et al., 1992.
Friedrich's ataxia	9q13 - 21.1	Frataxin	Recessive	GAA	7-22	200-900	DRG (sensory neurons), spimocerebellar tract	Campuzano et al., 1996.
SBMA	Xq11-12	Androgen receptor	X-linked recessive	CAG	9-36	38-62	Spinal chord- motoneurons	La Spada et al., 1991.
HD	4p16.3	Huntingtin	Dominant	CAG	6-34	36-121	Striatum and cortex	HDCRG, 1993.
SCA1	6p22-23	Ataxin-1	Dominant	CAG	6-44	39-82	Cerebellar purkinje cells, fronto-temporal lobes	Orr et al., 1993.
SCA2	12q23-24	Ataxin-2	Dominant	CAG	15-31	36-63	Cerebellar dentate neurons, brain stem	Trottier et al., 1995.
SCA3	14q24.31	Ataxin-3	Dominant	CAG	12-41	62-84	Cerebellar dentate cells, striatum, brain stem	Kawaguchi et al., 1994.
SCA6	19p13	CaCNA1a	Dominant	CAG	4-18	21-33	Cerebellar purkinje cells, inferior olive, dentate	Zhuchenko et al., 1997.
SCA7	3p12 –13	Ataxin-7	Dominant	CAG	4-35	37-306	Macula, cerebellum, visual cortex	David et al., 1997.
SCA12	5q	PP2A-B?	Dominant	CAG	7-28	65-78	Cortex and cerebellum	Holmes et al., 1999.
DRPLA	12q	Atrophin-1	Dominant	CAG	6-36	49-84	Cortex, striatum, cerebellum	Burke et al., 1994; Koide et al., 1994.
SCA17		TBP	Dominant	CAG	29-42	47-55	Cerebellum, striatum	Nakamura et al., 2001.
SCA8	13q21	Unknown	Dominant	CTG	16-91	107-127	Cortex and cerebellum	Koob et al., 1999.

CAG repeat length influences age of onset



Effect of CAG expansion on htt

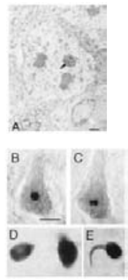
- CAG expansion is translated
  - CAG encodes for glutamine (Q)
- No loss of function
  - Patients homozygote for HD mutation
- Dominant gain of function

Mouse models of HD:

- Bates' R6 transgenic (R6/2)
  - Mangiarini et al. (1995) Cell
- cDNA transgenic
  - Reddy et al. (1998) Nat Genet *20*, 198-202
- N171 transgenic
  - Schilling et al. (1999) Hum Mol Genet *8*, 397-407
- YAC transgenic
  - Hodgson et al. (1999) Cell *23*, 181-92
- (CAG)n knock-in
  - Wheeler et al. (1999) Hum Mol Genet *8*, 115-22
  - Shelbourne et al. (1999) Hum Mol Genet *8*, 763-74
  - Lin et al. (2001) Hum Mol Genet *10*, 137-44

Intracellular aggregates

- Davies et al., 1997
  - Intranuclear aggregates
  - "Dark bodies" (Roizin, 1977)
- DiFiglia et al., 1997
  - Intranuclear aggregates
  - cytoplasmic aggregates
- Scherzinger et al., 1997
  - Ribbon-like morphology

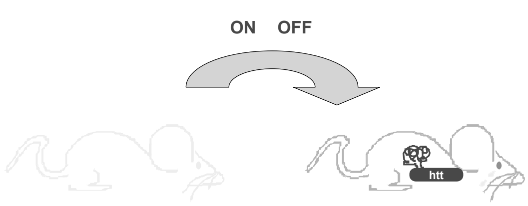


Question:

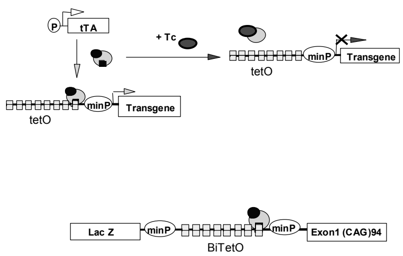
- Is continuous expression of the transgene required for the HD-like phenotype?
- Is the HD-like phenotype reversible?

Inducible mouse model

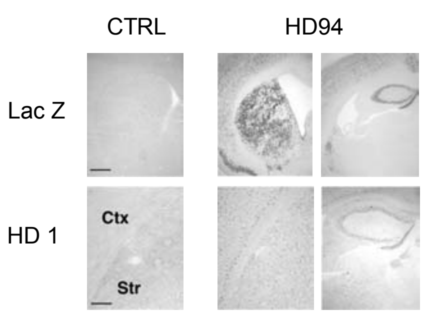
A model with a switch



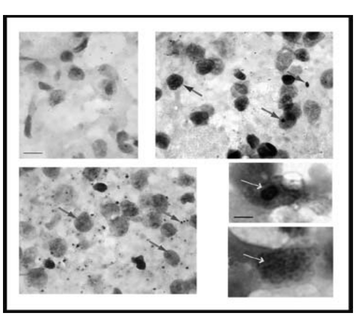
Tet-regulatable system



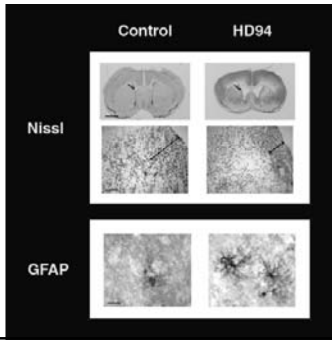
Expression pattern of polyQ-htt



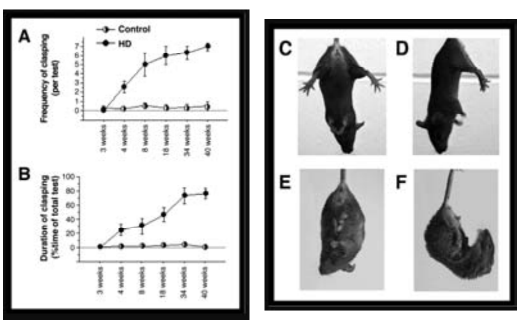
Intranuclear & cytoplasic aggregates



Neuropathology in HD94 mice

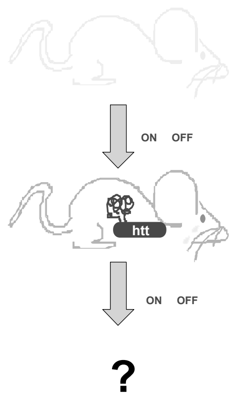


HD mice demonstrate a progressive clasping phenotype



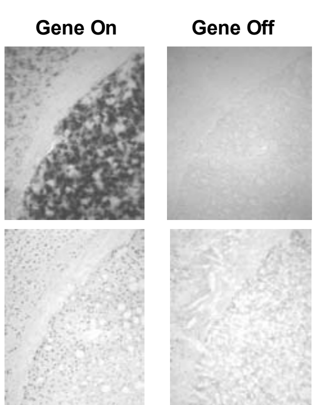
Summary

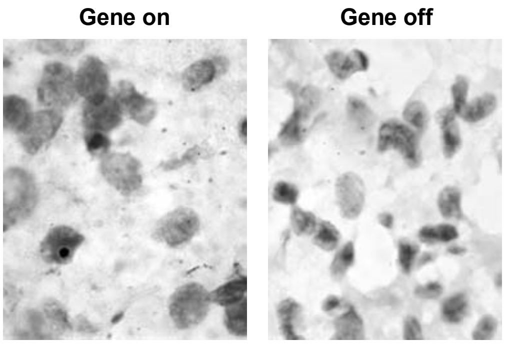
- Expression of exon1 CAG94 leads to an HD-like phenotype

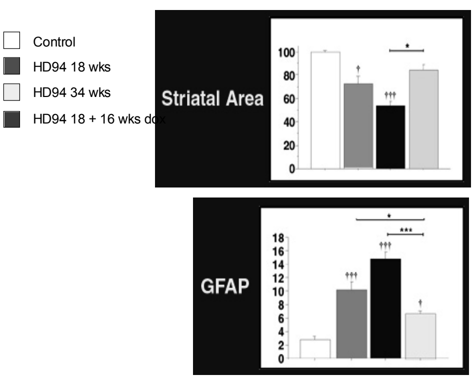


Protocol

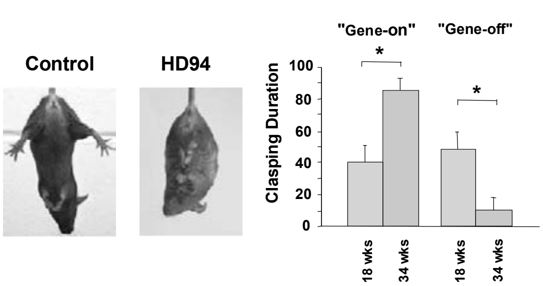
- 2 mg dox/ ml 5% sucrose
- Duration: 16 weeks
  - Halt progression
  - Amelioration







Behavioral Reversal in HD94 mice



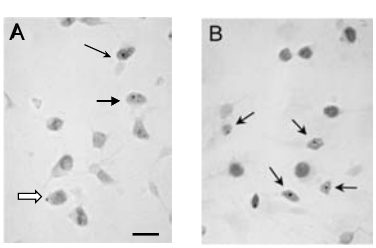
Summary

- Expression of exon1 CAG94 leads to an HD-like phenotype
- Abolishing gene expression for 16 weeks leads to a reversal of aggregate formation and motor phenotype

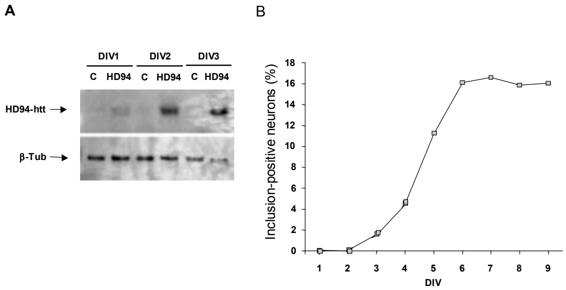
Primary striatal cultures from HD94 mice

- Kinetics of aggregate formation and reversion
- Possible mechanisms for aggregate reversion

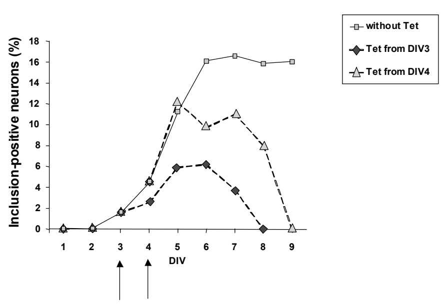
Aggregate formation *in vitro*



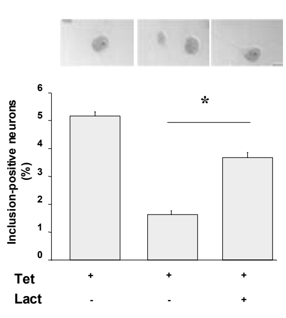
Aggregate Formation *in vitro*



Aggregate Formation and Reversion *in vitro*



Lactacystin inhibits aggregate reversion



Summary (2)

- *In vitro*
  - "Soluble" exon1CAG94 clears within 2 to 3 days of shutting down gene expression.
  - Aggregates clear within 5 days of shutting down gene expression (3d + 2d).
  - Aggregate clearance is inhibited in the presence of lactacystin (proteasome).

Tissue specificity of the pathology

Tissue-specific expression of mutant htt

HD94

Str-HD94

Ctx-HD94