Neurobiology of Stress, Depression, and Antidepressants: Remodeling Synaptic Connections

Ronald S. Duman, PhD
Department of Psychiatry
Yale University School of Medicine
Mood Disorders

• Depression affects ~17% of the population: higher risk for women (2:1).
• Economic cost is over $100 billion annually.
• Available treatments require weeks to months.
• Causes of depression and mechanisms of treatment response have not been identified.
• Studies demonstrate a role for neuronal atrophy and loss of neurotrophic factor support.
Evidence of Atrophy of Limbic and Cortical Regions In Major Depressive Disorder (MDD)

- **Decreased hippocampal** volume in MDD patients; reduction in volume is related to the duration of depression, and is blocked or reversed by antidepressant treatment.

- **Decreased prefrontal cortex** volume and hypofunction, correlates with disease severity in both MDD and BD.
Evidence of Neuronal Atrophy and Loss in Response to Stress: Preclinical Studies

• Chronic stress, which can lead to depression, decreases synaptic connections in the prefrontal cortex and hippocampus.

PFC layer V pyramidal neurons; Liu and Aghajanian, 2008
Chronic stress, which can lead to depression, decreases synaptic connections in the PFC and hippocampus; decreased synapses also reported in postmortem PFC of depressed subjects.

Loss of connections decreases circuit control of emotion, mood, and cognition, contributing to depressive symptoms.
Typical Antidepressants: Limitations

• Act on serotonin and/or norepinephrine monoamines (e.g., block reuptake transporter).

• Do not directly influence spine number and function.

• Delayed response of weeks to months.

• Low rate of efficacy: ~1/3 of patients respond to 1st drug, up to 2/3’s with multiple trials.

• Treatment resistant depression (TRD) of ~1/3 of patients.
Delayed and Low Response to Typical Antidepressants

5-HT neurotransmitter system: Slow Modulation

SSRI — 5-HT Transporter

Second Messengers (e.g. cAMP)

Delayed Adaptive Responses

PKA → Multiple Physiological Effects

Antidepressant Responses: Neuroprotection, Neuroplasticity, Neurogenesis

Nucleus

Regulation of BDNF Gene Expression
Drugs Acting on the Glutamate Neurotransmitter System

Ketamine

Glutamate Fast Excitation

NMDA

AMPA

Na^+,Ca^{2+}

Na^+

CAMK

Rapid Response

Activity-dependent Release of BDNF

5-HT neurotransmitter system: Slow Modulation
SSRI → 5-HT Transporter

Second Messengers (e.g. cAMP)

Delayed Response

PKA

Multiple Physiological Effects

Nucleus

Regulation of BDNF Gene Expression

1

2
Ketamine Produces Rapid Antidepressant Effects

- NMDA receptor antagonist and dissociative anesthetic at hi doses.
- At low doses, ketamine produces a rapid response in treatment resistant depressed patients
Larger Replication Study Demonstrating Rapid Antidepressant Actions of Ketamine

Zarate, Charney, et al., at NIMH et al., 2006
Therapeutic actions of ketamine in bipolar depressed patients

Zarate et al., 2012; Biological Psychiatry
Ketamine and Suicide Ideation


Ketamine and Suicide Ideation


These effects are particularly relevant given that:
- 36,000 individuals die from suicide/yr, twice as many as by homicide (Center for Disease Control).
- 23% of suicide victims were on antidepressant treatments at the time of death.
Multiple Replication Studies

Percent of Patients classified as Responders

- MDD, open-label, 6 doses, $N=9$ (33)
- MDD, open-label, 1 dose, $N=26$ (32)
- MDD, controlled, 1 dose, $N=8$ (1)
- MDD, controlled, 1 dose, $N=17$ (4)
- MDD, controlled, 1 dose, $N=10$ (31)
- BD, controlled, 1 dose, $N=17$ (14)
- BD, controlled, 1 dose, $N=14$ (15)

aan het Rot et al. Biol Psychiatry 2012
The discovery that ketamine produces rapid antidepressant effects in treatment resistant depressed patients, by a novel mechanism (NMDA receptor blockade), is arguably the most significant advance in the field in over 50 years.
The discovery that ketamine produces rapid antidepressant effects in treatment resistant depressed patients, by a novel mechanism (NMDA receptor blockade), is arguably the most significant advance in the field in over 50 years.

What is the mechanism for the rapid actions of ketamine?
Synaptogenesis and rapid actions of ketamine?

- Might ketamine, thru effects on glutamate act via regulation of the number and function of spine-synapses?
- Synapses undergo rapid remodeling in response to glutamate activity.
- Typical antidepressants do not directly effect synapses.
- What is the effect of low dose ketamine on spine synapses in the PFC?
What are Synaptic Connections?

Single neuron

Branch

Connection/Synapse
Chronic stress, which can lead to depression, decreases synaptic connections in the PFC and hippocampus; decreased synapses also reported in postmortem PFC of depressed subjects.

Loss of connections decreases circuit control of emotion, mood, and cognition, contributing to depressive symptoms.
Ketamine Rapidly Increases Neuronal Connections
Ketamine Rapidly Increases Synaptic Proteins in PFC

- Increased spine number, including
- Increased number of “mushroom” or mature spines

Li et al., Science, 2010
Ketamine Rapidly Increases Synaptic Proteins in PFC

Ketamine Time Course

0 1 2 6 24 72 hr

Synaptoneurosome Preparation

Western Blot Synaptic Proteins
Time Course for the Induction of Synaptic Proteins Corresponds to the Time Course for the Clinical Response

Zarate et al., 2006

![Graph showing the time course of HDRS scores and fold change in synaptic proteins.]
Ketamine, Synapses, and Behavior

• Ketamine has rapid actions in forced swim and learned helplessness models of depression.

• Chronic unpredictable stress (CUS) causes depressive behaviors (e.g., anhedonia) & decreases synapses.

• Antidepressants (e.g., fluoxetine) take weeks.

• Rigorous rodent test of the rapid actions of ketamine to reverse the spine and behavioral deficits caused by stress.
Ketamine rapidly reverses the spine and behavioral deficits caused by chronic stress (3 weeks).

Pathophysiology and treatment of depressive behaviors are associated with the number and function of synaptic connections.
What is the mechanism by which ketamine increases spine number and function?

Control     Ketamine

How does administration of an NMDA receptor antagonist cause an increase in synaptogenesis?
Ketamine Blocks the Firing of GABAergic Interneurons that Inhibit Glutamatergic Transmission

Houman and Moghaddam, 2007: “NMDA receptors preferentially drive the activity of cortical inhibitory interneurons suggesting that NMDA receptor inhibition causes cortical excitation by disinhibition of pyramidal neurons.”
mTOR mediates long-term, protein synthesis dependent learning and memory.

mTOR regulates translation initiation.

Present in dendrites, as well as cell bodies.

Regulated by phosphorylation: phospho-mTOR.
Rapamycin, a Selective inhibitor of mTOR, Blocks the Antidepressant Actions of Ketamine

- **Glutamate Burst**
  - Increased translation of synaptic proteins: e.g., GluA1 and PSD95
  - Rapamycin Blocks the induction of:
    - Synaptic proteins
    - Spine number and function
    - Antidepressant behavior

- **Ketamine**
  - mGluR2/3
  - Glutamate
  - GABA
  - NMDA
  - AMPA
  - NBQX

- **Spine Synapse Number & Function**
  - Glutamate
  - BDNF
  - TrkB
  - mTOR
  - Akt
  - ERK
  - Rapamycin

- **Rapamycin**
  - A Selective inhibitor of mTOR
  - Blocks the Antidepressant Actions of Ketamine
Mechanisms for the rapid actions of ketamine: Role for Brain Derived Neurotrophic Factor

- BDNF is required for the survival of neurons in adult animals.
- BDNF is also required for activity-dependent induction of synaptic function.
Neurotrophic Factors

- Developing brain: growth, guidance, and survival of neurons.

- Adult brain: regulate neuronal function, and growth/survival.

- Regulated by neuronal activity (e.g., learning and memory).

Activity-dependent Release of BDNF

Regulation of BDNF Gene Expression

Transcription
# Neurotrophic/Growth Factor Families Implicated in Stress and Depression

<table>
<thead>
<tr>
<th>Family</th>
<th>Stress/Depression</th>
<th>Antidepressant</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brain Derived Neurotrophic Factor</td>
<td>Decrease</td>
<td>Increase</td>
</tr>
<tr>
<td>Vascular Endothelial Growth Factor</td>
<td>Decrease</td>
<td>Increase</td>
</tr>
<tr>
<td>Fibroblast Growth Factor</td>
<td>Decrease</td>
<td>Increase</td>
</tr>
<tr>
<td>Insulin-Like Growth Factor</td>
<td>no change</td>
<td>Increase</td>
</tr>
</tbody>
</table>
BDNF Val\textsuperscript{66}/Met Polymorphism

BDNF Met allele

- Decreases processing and activity-dependent BDNF release.
- Incidence: ~25% of the population.

**Associated with:**

- Reduced episodic memory performance and executive function.
- Decreased hippocampal volume in normal subjects, MDD subjects, and bipolar patients.
- Increases vulnerability for depression: gene x stress interaction, (Kaufman et al., 2006; Kim et al., 2007; Gatt et al., 2009).
Ketamine Induction of spines and antidepressant behavior is blocked in BDNF Met mice

Rong-Jian Liu et al., 2012, Biol Psych
Consistent with Autry et al., 2011.

Met/Met blocks
Blocks BDNF release

Xiao-Yuan Li and Rong-Jian Liu
BDNF Val66Met Polymorphism and Antidepressant Efficacy of Ketamine in Depressed Patients


• Based on the findings in BDNF Met/Met mice, hypothesized that patients carrying a BDNF Met substitution would show an attenuated antidepressant response to ketamine infusion compared with Val/Val patients.

• Met carriers showed a significantly reduced (~50%) response to ketamine compared to Val carriers. \( F = 5.59, \) \( df = 4, \) \( p = .0007 \).
• Val66Met allele can be used as a marker to identify patients who are responders or nonresponders to ketamine.
Influence of ketamine vs. typical antidepressants on BDNF: release vs. expression

• Ketamine, via glutamate-synaptic activity, increases BDNF release.
• Typical ADTs increase BDNF expression, but no evidence of release.
• This could account for the rapid and efficacious actions of ketamine.
Stress decreases synaptic connections: Rapid reversal by ketamine

Ketamine produces nascent spines on pyramidal neurons in the PFC: stabilization?

Control of mood/emotion requires synaptic integrity of PFC neurons
What connections/circuits underlie the antidepressant actions of ketamine as well as stress and depression?

Control of mood/emotion requires synaptic integrity of PFC and inhibitory connections with the amygdala and other brain regions.
Ketamine is a drug of abuse with side effects:
Need for safer rapid acting, ketamine-like antidepressants

Hamilton Depression Scale: $p = 0.0001$

VAS, “High” $P = 0.0001$

BPRS, Positive Symptoms of Schizophrenia $P = 0.007$

Berman et al., Biol Psychiatry 2000
Development of Safer Rapid Acting Agents With Fewer Side Effects

- **Ketamine, repeated dosing and nasal administration**: both have shown efficacy and provide evidence for approval of ketamine.

- **Nonselective NMDA antagonists**: drugs with fewer side effects; AZD6367, reported to be effective with repeated IV dosing.

- **Selective NR2B receptor antagonists**: CP-101,606 reported to have antidepressant effects (Preskorn et al., 2007); Ro 25-6981, basic studies.

- **mGlu2/3 receptor antagonists**: basic studies of LY341495, MGS0039 increase glutamate by blocking autoreceptors.

- **NMDA-glycine receptor agents** (GLYX-13 and D-cycloserine) reported to have clinical efficacy after single dose or chronic dosing.

- **Muscarinic receptor antagonists**: Scopolamine reported to produce rapid antidepressant actions in depressed patients.
Development of Safer Rapid Acting Antidepressants

**NMDA antagonists:** Ketamine, CP-101,606, AZD6765, Ro 25,6981

**mGluR2/3 antagonists:** LY341495, MGS0039

**Muscarinic antagonists:** Scopolamine, Telenzapine

**AMPA Receptor Potentiating drugs**

**GSK-3 antagonists:** Lithium, SB216763

**Rapid reversal of the Synaptic Loss Caused by Stress and Depression**
What are the signaling mechanisms underlying neuronal atrophy?

<table>
<thead>
<tr>
<th>Control</th>
<th>Stress</th>
</tr>
</thead>
</table>

- Neuronal atrophy caused by relatively mild stress (1 wk)
- Could have wide spread consequences: MDD, PTSD, schizophrenia, cognitive deficits, other.
- Does stress decrease mTOR signaling and synaptic protein synthesis?
Does stress decrease spine synapses via inhibition of mTOR signaling: Mechanisms?

- REDD1 (Regulated in Development and DNA damage)
- REDD1 is induced by glucocorticoids in muscle and brain
- REDD1 inhibits cell growth and protein synthesis directed by mTOR via stabilization of TSC1/2 (tuberin)
REDD1 Expression is increased in by chronic stress in rat PFC

![Graph showing increased REDD1 expression in CUS compared to control](image)

- mRNA: Control = 1.0, CUS = 1.5
- Protein: Control = 1.0, CUS = 1.5

Figure 1 (Ota et al.)

21 Days CUS → Sacrifice

REDD1
- Control
- CUS

Fold Change Relative to Control (Normalized to GAPDH)

Ota et al., Nature Medicine, 2014
REDD1 mRNA Expression is increased in postmortem dIPFC of depressed subjects

Two independent cohorts
Total of 38 controls and 38 MDD

~65% increase in MDD

Ota et al., Nature Medicine, 2014
REDD1 knock out mice are resilient to the synaptic and behavioral deficits (anhedonia) caused by chronic stress.
Stress and Depression decrease mTOR signaling via induction of REDD1

Control  Stress/MDD

BDNF  TrkB  GluR1

PSD95  GluR1  Insertion

S6K  eEF2K  4E-BP

mTORC1  mTOR

Tsc2  Tsc1  Rheb  Akt  PI3K  MEK  ERK

Translation

Stress/MDD  REDD1
Model of Depression and Rapid Antidepressant Response: Remodeling of Synaptic Connections

- Develop novel synaptogenic treatments with fewer side effects.
- What causes relapse after 7-10 d? Loss of spines coincides with relapse?
- What treatments sustain the increase in spine number and function?
Acknowledgements

• Nick Li
• Boyoung Lee
• Maha Elsayed
• Bhavya Voleti
• Andrea Navarria
• Mounira Banasr
• Xiao-Yuan Li
• Neil Fournier
• Manabu Fuchikami
• Ashley Lepack
• Jason Dwyer
• Kristie Ota
• Sophie Dutheil
• Astrid Becker
• Masaaki Iwata
• Pawel Licznerski
• Rose Terwilliger
• Alexandra Thomas
• George Aghajanian
• Rong-Jian Liu
• Samuel Sathyanesan
• Cathy Duman
• Gerard Sanacora

Funding Sources
• NIMH
• NARSAD
• State of CT
• Yale