CONCLUSIONS

• Selective changes in NMDAR subunits were found in corticolimbic brain regions of FSL rats, an animal model of depression.

• Chronic antidepressant administration of nortriptyline and escitalopram both increased BDNF mRNA expression, but induced phosphorylation-site specific regulation of TrkB (and not TrkA) in the hippocampus of FSL rats.

• The distinct regulation of BDNF/TrkB signaling illustrate differential activation of downstream signaling cascades by the TCA and SSRI antidepressants.

BACKGROUND

The variable efficacy of current antidepressant medications on neurovegetative symptoms of depression is clinically recognized [1]. Besides elevation of mood, antidepressants also differ in efficacy to alleviate additional symptoms, such as relieving cognitive impairments commonly observed in depressed patients [1].

Limited preclinical data is available on signaling pathways activated by different types of antidepressant agents in brain regions derived from healthy rodents and disease models [2,3].

Brain-derived neurotrophic factor (BDNF) is implicated in antidepressant actions, via TrkB receptor-mediated modulation of synaptic plasticity and responses to environmental stimuli in brain circuits regulating emotionality and cognition [2]. Glutamatergic neurotransmission regulates BDNF signaling and particularly alterations of the NMDAR has been reported in clinically depressed patients [1].

Aims

• Examination of glutamatergic AMPAR and NMDAR subunits and their phosphorylation state in the Flinders Sensitive Line (FSL) rat model of depression.

• Comparison of two principal antidepressant therapies, the tricyclic agent (TCA) nortriptyline and the selective serotonin reuptake inhibitor (SSRI) escitalopram.

• To examine regulation of BDNF signaling and separate phosphorylation effects on TrkB from effects on TrkA.

METHODS

THE FLINDER SENSITIVE LINE (FSL)

Male FSL and control FRL rats were used, derived from selective breeding of Sprague-Dawley rats. FSL rats are utilized as a translational model for validation of pharmacological targets with resemblance for behavioral and neurochemical features of depression [3].

Antidepressant Food Delivery

Escalator (340 mg/kg food pellets for 3 weeks followed by 410 mg/kg), nortriptyline (330 mg/kg) or vehicle was administered to adult FSL and FRL rats. Behavioral antidepressant-like actions were verified after 6 weeks of chronic antidepressant treatment.

Immunoprecipitation

Anti-sera against phospho-TrkB may cross-react with phospho-TRKA, but most immunoprecipitation studies have used unselective antibodies for TrkB/TrkA in brain regions containing both receptor types or reported low efficacy for immunoprecipitation with selective antibodies [2].

To distinguish between phosphorylation events at TrkA vs. TrkB, we used antibodies towards total TrkA, TrkB or phospho-TrkB for immunoprecipitation with Protein-G linked Magnetic Beads (Dynabeads®) to increase sensitivity.

Immunoblotting

Blast differences between FSL and FRL rats and following antidepressants were investigated with Western blot using antibodies for total protein or specific phosphorylation-sites.

In situ Hybridization

Histological measurements were performed on sections using 35S-radiolabeled antisense riboprobes against BDNF.

RESULTS

1. Corticolimbic reductions of NMDAR subunits

2. Up-regulated phosphorylation at NR1 and NR2A subunits

3. Increased BDNF mRNA by both antidepressants

4. Phosphorylation-site specific regulation of TrkB

5. Possible involvement of TrkA in TrkB-attributed effects

6. Selective phosphorylation of TrkB and not TrkA

REFERENCES


5. Imbalance in the regulation of NMDAR subunits in corticolimbic brain regions of FSL rats, an animal model of depression.

6. Increased phosphorylation of TrkB in the hippocampus of FSL rats, although no changes in phospho-TrkA were observed.

7. The distinct regulation of BDNF/TrkB signaling illustrate differential activation of downstream signaling cascades by the TCA and SSRI antidepressants.

8. Examination of glutamatergic AMPAR and NMDAR subunits and their phosphorylation state in the Flinders Sensitive Line (FSL) rat model of depression.

9. Comparison of two principal antidepressant therapies, the tricyclic agent (TCA) nortriptyline and the selective serotonin reuptake inhibitor (SSRI) escitalopram.

10. To examine regulation of BDNF signaling and separate phosphorylation effects on TrkB from effects on TrkA.

11. The variable efficacy of current antidepressant medications on neurovegetative symptoms of depression is clinically recognized [1].

12. The difference between FSL and FRL rats and following antidepressants were investigated with Western blot using antibodies for total protein or specific phosphorylation-sites.

13. Histological measurements were performed on sections using 35S-radiolabeled antisense riboprobes against BDNF.