Neuroscience Letters 598 (2015) 23–28

Contents lists available at ScienceDirect

Neuroscience Letters

journal homepage: www.elsevier.com/locate/neulet

Research article

# Dishevelled-2 regulates cocaine-induced structural plasticity and Rac1 activity in the nucleus accumbens

Caroline Dias[a], David Dietz[a,1], Michelle Mazei-Robison[a,2], Haosheng Sun[a], Diane Damez-Werno[a], Deveroux Ferguson[a,3], Matthew Wilkinson[a], Jane Magida[a], Virginia Gao[a], Rachael Neve[b], Eric J. Nestler[a,*]

[a] Fishberg Department of Neuroscience and Friedman Brain Institute, Icahn School of Medicine at Mount Sinai, NY, USA
[b] Department of Brain and Cognitive Sciences, Massachusetts Institute of Technology, Cambridge, MA, USA

## HIGHLIGHTS

- Down-regulation of Dishevelled-2 occurs in the nucleus accumbens with cocaine.
- Dishevelled-2 activates Rac1 *in vivo* in the mature adult brain.
- Dishevelled-2 is important in controlling spine dynamics in response to cocaine.

## ARTICLE INFO

*Article history:*
Received 19 March 2015
Received in revised form 29 April 2015
Accepted 1 May 2015
Available online 6 May 2015

*Keywords:*
Dendritic spines
Morphological plasticity
Medium spiny neurons
Wnt signaling

## ABSTRACT

Chronic cocaine exposure increases the density of dendritic spines on medium spiny neurons (MSNs), the predominant neuronal cell type of the nucleus accumbens (NAc), a key brain reward region. We recently showed that suppression of Rac1, a small GTPase, is a critical mediator of this structural plasticity, but the upstream determinants of Rac1 activity in this context remain to be elucidated. In this study we examined whether isoforms of Dishevelled, a key hub protein of multiple branches of Wnt signaling, including Rac1, are regulated in the NAc by chronic cocaine, and whether these Dishevelled isoforms control Rac1 activity in this brain region *in vivo*. We found that chronic cocaine administration decreased expression of Dishevelled-2, and several other Wnt signaling components, in the NAc, and that overexpression of Dishevelled-2, but not Dishevelled-1, conversely upregulated Rac1 activity and prevented the cocaine induction of dendritic spines on NAc MSNs. We posit that the cocaine-induced downregulation of Dishevelled-2 in the NAc is an upstream regulator of Rac1 activity and plays an important role in the dynamic structural plasticity of NAc MSNs seen in response to chronic cocaine exposure.



## 1. Introduction

Although initially studied in the context of development, recent findings have demonstrated an important role for Wnt signaling in the mature vertebrate central nervous system [1]. For example, we recently demonstrated a critical role for this signaling pathway in the nucleus accumbens (NAc), a key brain reward region, in a mouse model of depression [2,3]. Because the NAc is also a brain region critical to the development of addiction pathophysiology, we were interested in studying Wnt signaling in the context of chronic cocaine exposure. Specifically, we focused on a protein that acts as a central hub of Wnt signaling: Dishevelled.

Dishevelled, an intracellular protein named initially based on the *Drosophila* phenotype of unoriented body and wing hairs seen upon knockout of the gene [4], has since been determined to represent a key hub in signal transduction through several different pathways of Wnt signaling [5]. In "canonical" Wnt signaling, activation of Dishevelled through a transmembrane Wnt-receptor complex eventually leads to stabilization of β-catenin, allowing β-catenin to translocate to the nucleus where it acts as a transcriptional activator at Wnt target genes. In the NAc, it is this

* Corresponding author.
*E-mail address:* eric.nestler@mssm.edu (E.J. Nestler).
[1] Department of Pharmacology and Toxicology, University at Buffalo, Buffalo, New York, USA.
[2] Department of Physiology, Michigan State University, East Lansing, Michigan, USA.
[3] Department of Basic Medical Sciences, The University of Arizona College of Medicine, Phoenix, Arizona, USA.

http://dx.doi.org/10.1016/j.neulet.2015.05.003
0304-3940/

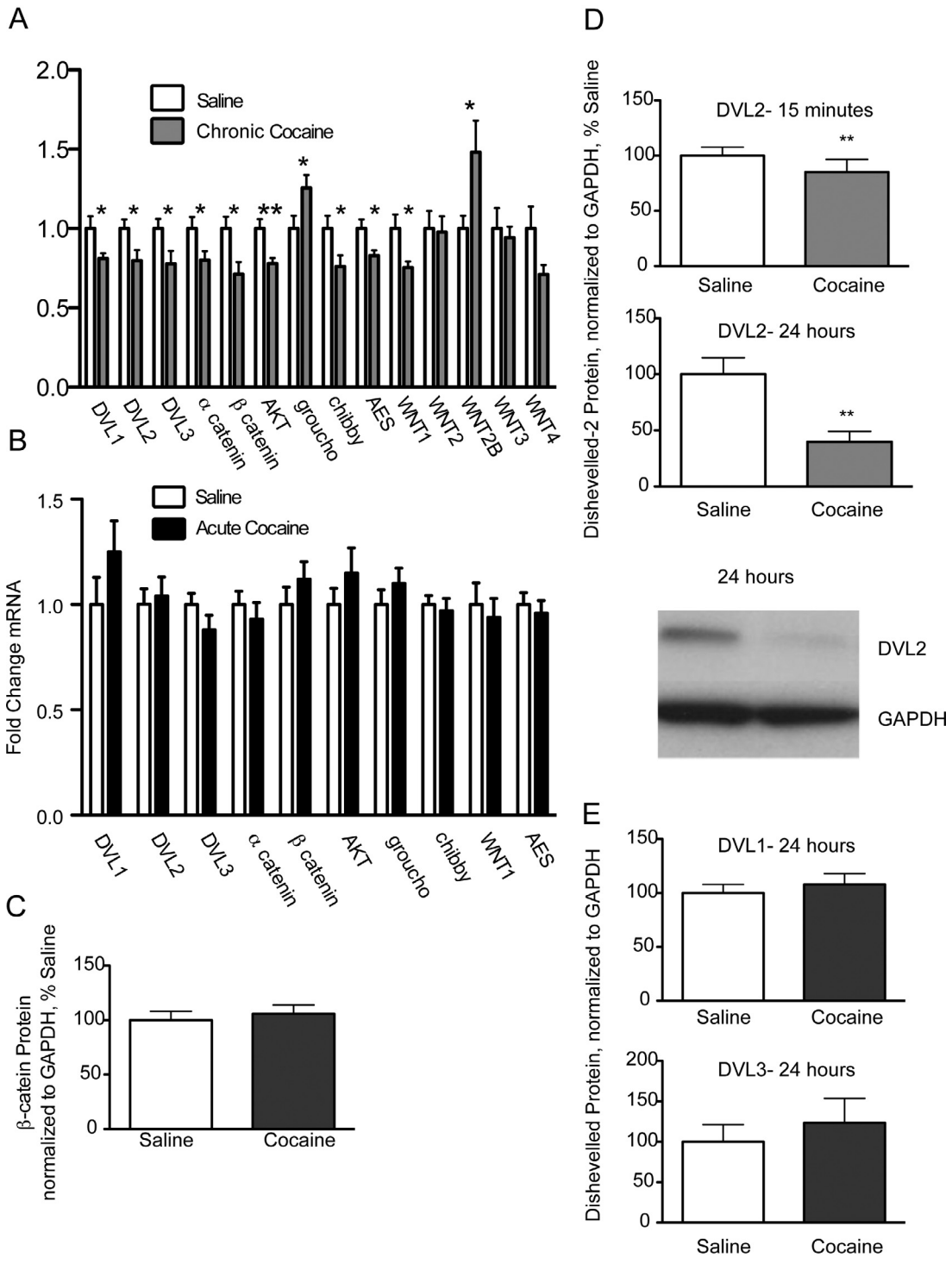


**Fig. 1.** Chronic cocaine regulation of Dishevelled-2 (DVL2) expression in NAc.
(A) 7 days of experimenter-administered cocaine (20 mg/kg) IP injections leads to widespread downregulation of Wnt signaling molecules as compared to saline 24 h after the last dose ($n = 10$/group, $^{*}p < 0.05$, $^{**}p < 0.01$). (B) A single 20 mg/kg cocaine injection leads to no such changes ($n = 9$/group, $p > 0.05$). (C) There is no change in β-catenin protein expression 24 h after chronic cocaine ($n = 10$/group, $p > 0.05$). (D) There is decreased Dishevelled-2 (DVL2) protein expression both 15 min and 24 h after chronic cocaine (top panel-15 min: $^{**}p < 0.01$, $n = 9$-10/group; bottom panel and blot- 24 h: $^{**}p < 0.01$, $n = 6$/group). (E) There is no regulation of Dishevelled-1 (DVL1) or Dishevelled-3 (DVL3) protein expression 24 h after chronic (7 days) cocaine ($n = 11$/group, $p > 0.05$).

branch of Wnt signaling that our laboratory has found to be critical in mediating resilience to stress [3]. However, signaling through Dishevelled also regulates distinct, "non-canonical" pathways [6,7]. For example, it can affect calcium signaling and $Ca^{2+}$/calmodulin-dependent protein kinase 4. Notably, mouse Dishevelled-1 and -2 can in addition activate certain small GTPases, including Rac1 [8]. Humans and mice express three Dishevelled homologs: 1, 2, and 3. Although there is functional redundancy between these three isoforms, an individual knockout approach has demonstrated distinct phenotypes of each protein. For example, Dishevelled-1 knockout mice, but not mice lacking other Dishevelled isoforms, have social interaction abnormalities [9,10]. In contrast, Dishevelled-2 or -3