



SUPPLEMENTARY ONLINE DATA Nogo-A couples with Apg-1 through interaction and co-ordinate expression under hypoxic and oxidative stress

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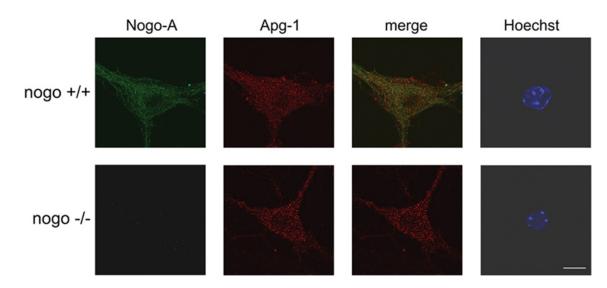


Figure S1 Subcellular localization of Nogo-A and Apg-1 in cultured hippocampal neurons

Hippocampal neurons from wild-type and Nogo-/- mice were cultured for 20 days in vitro before being fixed and stained for Nogo-A and Apg-1. Single-plane confocal analyses illustrate the subcellular localization of these two proteins in the wild-type and of Apg-1 in the Nogo-knockout. Scale bar, 10 μ m.

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Table S1 List of NiG candidate interactors identified by two different MS screens

All proteins are listed which were identified in two independent MS screens as NiG-interactor candidates. Following elution from the NiG column, proteins were either subjected to brief SDS/PAGE, followed by in-gel digestion and LC-MS/MS (four independent experiments), or were TCA-precipitated and, after digestion, subjected to LC-MS/MS (two independent experiments). The criteria for inclusion were at least two hits with the gel-based approach without any detection in the controls, and two hits with the TCA-based approach without any detection in the controls. Two peptides per protein were the minimum for a positive protein identification. Peroxiredoxins were addressed separately because Prdx2 was described as an interactor of NiG [1]. As shown in the Table, we have also found peroxiredoxins, including Prdx2, in both MS screens, albeit with a lower frequency and/or weaker sample compared with the control ratio than other proteins. CaMKII (Ca²⁺/calmodulin-dependent protein kinase II) had a very good frequency and sample compared with control ratio in the gel-based approach, but we could not confirm interaction with endogenous Nogo-A via co-immunoprecipitation, suggesting that CaMKII interacts with the isolated NiG domain, but not with full-length Nogo-A.

Screen	Protein name	Accession number	Frequency of hits (sample/control)	Remarks
Gel approach (four independent experiments)	CaMKIIα	EDL09793.1	$4 \times / 0 \times$	We could not verify interaction via co-immunoprecipitation using brain lysate
	Apg-1	NP_035150	3×/0×	Interaction is characterized in the present paper
	Clathrin heavy chain 1	NP_001003908, XP_181312	3×/0×	
	F-actin capping protein α	EDL13878	$2 \times 0 \times$	
	Dynamin 3	EDL39302.1	$2 \times 0 \times$	
	Protein phosphatase 1, regulatory subunit 7	EDL39961	$2 \times / 0 \times$	
	Prdx1	NP_035164	$2 \times / 2 \times$	
	Prdx4	AAH19578	$1 \times 0 \times$	
TCA approach (two independent experiments)	Apg-1	NP_035150	$2 \times / 0 \times$	Interaction is characterized in the present paper
	Prdx1	NP 035164	$2 \times / 2 \times$	
	Prdx2	NP_035693	$2 \times /1 \times$	Interaction with NiG/Nogo-A was described in [1].
	Prdx5	P99029	1×/0×	

REFERENCE

1 Mi, Y. J., Hou, B., Liao, Q. M., Ma, Y., Luo, Q., Dai, Y. K., Ju, G. and Jin, W. L. (2012) Amino-Nogo-A antagonizes reactive oxygen species generation and protects immature primary cortical neurons from oxidative toxicity. Cell Death Differ. **19**, 1175–1186

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