

## **Supplementary Information**

**Manuscript title:** Zika virus exposure affects neuron-glia communication in the hippocampus of adult rats

**Author list:** Larissa Daniele Bobermin\*<sup>1</sup>, André Quincozes-Santos\*<sup>1,2</sup>, Camila Leite Santos<sup>1</sup>, Ana Paula M. Varela<sup>3</sup>, Thais F. Teixeira<sup>3</sup>, Krista Minéia Wartchow<sup>1</sup>, Lílian Juliana Lissner<sup>1</sup>, Amanda da Silva<sup>1</sup>, Natalie K. Thomaz<sup>1</sup>, Lucélia Santi<sup>4,5,6</sup>, Walter O. Beys-da-Silva<sup>4,5,6</sup>, Paulo M. Roehe<sup>3</sup>, Patrícia Sesterheim<sup>7</sup>, Jorge A. Guimarães<sup>5,6</sup>, Carlos-Alberto Gonçalves<sup>1,2</sup>, Diogo Onofre Souza<sup>1,2</sup>

\* These authors contributed equally to this work.

<sup>1</sup>Programa de Pós-Graduação em Ciências Biológicas: Bioquímica, Instituto de Ciências Básicas da Saúde, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil

<sup>2</sup>Departamento de Bioquímica, Instituto de Ciências Básicas da Saúde, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil

<sup>3</sup>Departamento de Microbiologia, Imunologia e Parasitologia, Instituto de Ciências Básicas da Saúde, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil

<sup>4</sup>Faculdade de Farmácia, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil

<sup>5</sup>Centro de Pesquisa Experimental, Hospital de Clínicas de Porto Alegre, Porto Alegre, RS, Brazil

<sup>6</sup>Programa de Pós-Graduação em Biologia Celular e Molecular, Centro de  
Biotecnologia, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil

<sup>7</sup>Centro de Cardiologia Experimental, Instituto de Cardiologia/Fundação Universitária  
de Cardiologia, Porto Alegre, RS, Brazil

André Quincozes-Santos

Departamento de Bioquímica

Programa de Pós-Graduação em Ciências Biológicas: Bioquímica

Instituto de Ciências Básicas da Saúde

Universidade Federal do Rio Grande do Sul

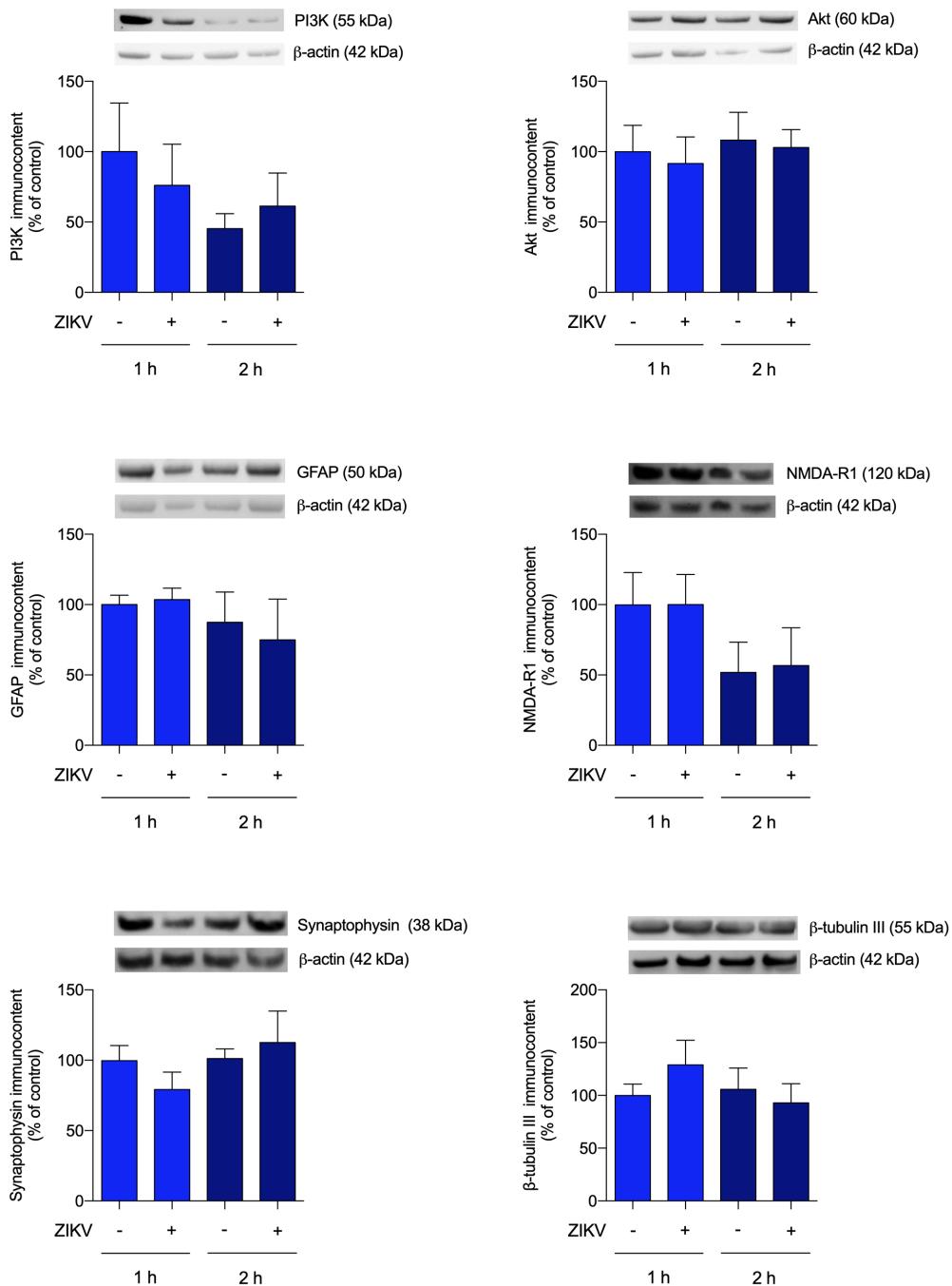
Rua Ramiro Barcelos, 2600 – Anexo

Bairro Santa Cecília

90035–003 Porto Alegre, RS, Brazil

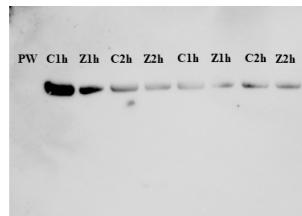
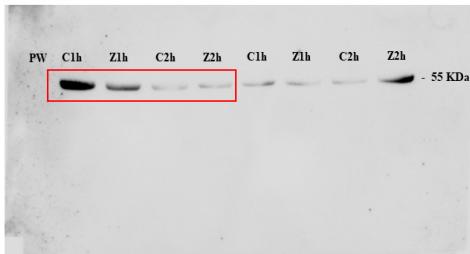
Phone: +55 51 3308 5567

E-mail: [andrequincozes@ufrgs.br](mailto:andrequincozes@ufrgs.br)



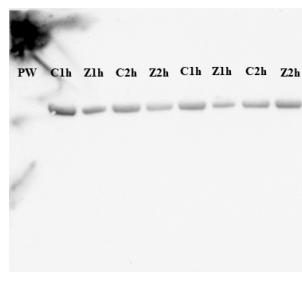
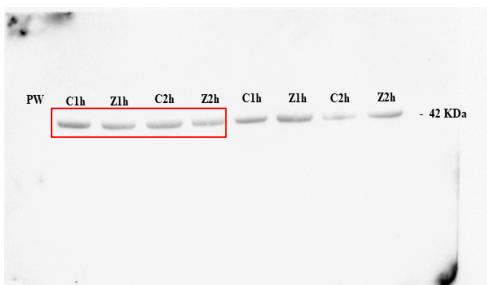
**Figure S1.** Effects of ZIKV on immunocontent of proteins related to neuronal and glial functions. Hippocampal slices were infected with  $10^5$  PFU of ZIKV (non-infection controls were simultaneously carried out). Western blotting analysis for (a) PI3K; (b) Akt; (c) GFAP; (d) NMDA-R1; (e) synaptophysin; (f) β-tubulin III were performed in the hippocampal slices after 1 h or 2 h incubation. Bars represent means  $\pm$  SEM of at least three experimental determinations, analyzed by one-way ANOVA followed by Tukey's test. Representative western blotting images are shown in the graphs.

**PI3K (55 kDa)**



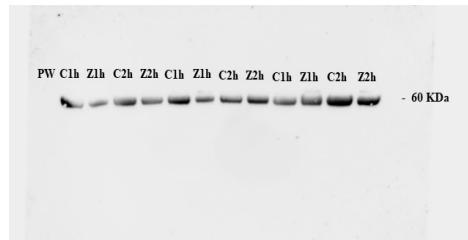
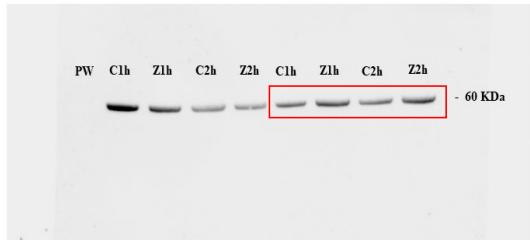
PW, Protein weight  
C1h, Control 1 h  
Z1h, ZIKV 1 h  
C2h, Control 2 h  
Z2h, ZIKV 2 h

**$\beta$ -actin (42 kDa) for PI3K analysis**

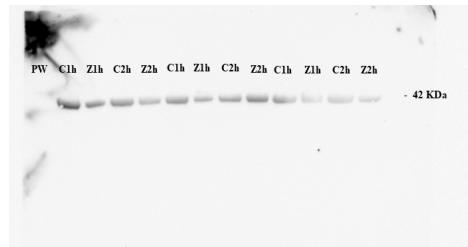
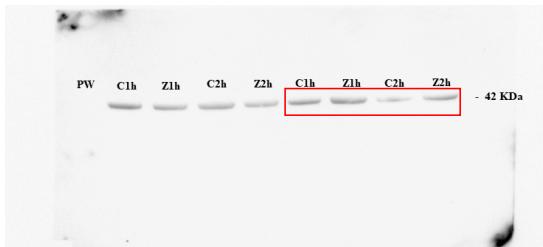


**Figure S2.** Full-length western blot membranes for PI3K detection and the respective  $\beta$ -actin controls. Red boxes highlight the bands used as representative images in the Figure S1.

### Akt (60 kDa)

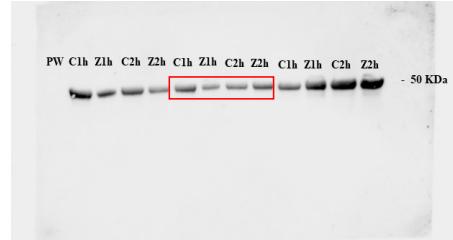


### $\beta$ -actin (42 kDa) for Akt analysis



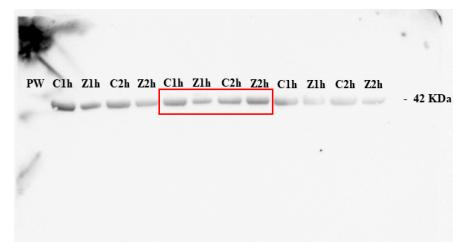
**Figure S3.** Full-length western blot membranes for Akt detection and the respective  $\beta$ -actin controls. Red boxes highlight the bands used as representative images in the Figure S1.

**GFAP (50 kDa)**



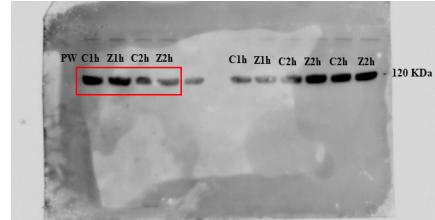
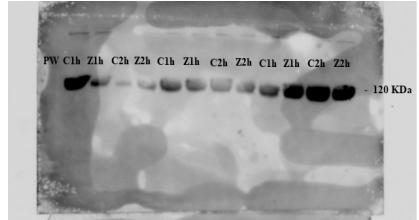
PW, Protein weight  
C1h, Control 1 h  
Z1h, ZIKV 1 h  
C2h, Control 2 h  
Z2h, ZIKV 2 h

**$\beta$ -actin (42 kDa) for GFAP analysis**



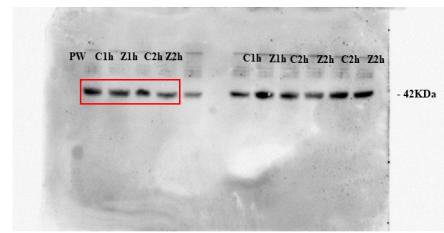
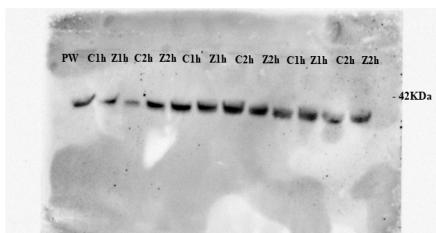
**Figure S4.** Full-length western blot membranes for GFAP detection and the respective  $\beta$ -actin controls. Red boxes highlight the bands used as representative images in the Figure S1.

### NMDA-R1 (60 kDa)



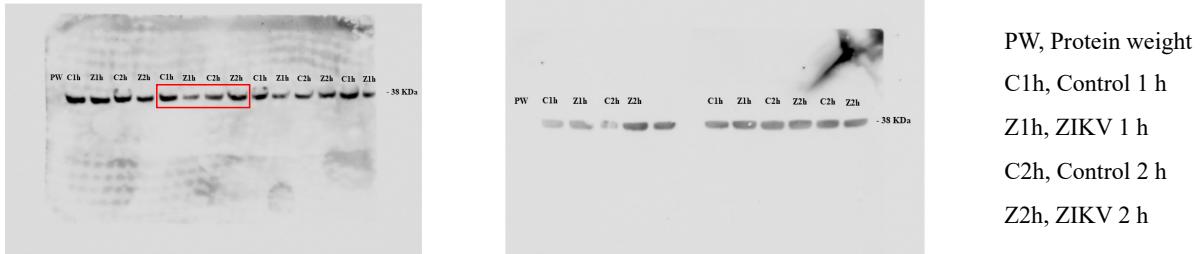
PW, Protein weight  
C1h, Control 1 h  
Z1h, ZIKV 1 h  
C2h, Control 2 h  
Z2h, ZIKV 2 h

### $\beta$ -actin (42 kDa) for NMDA-R1 analysis

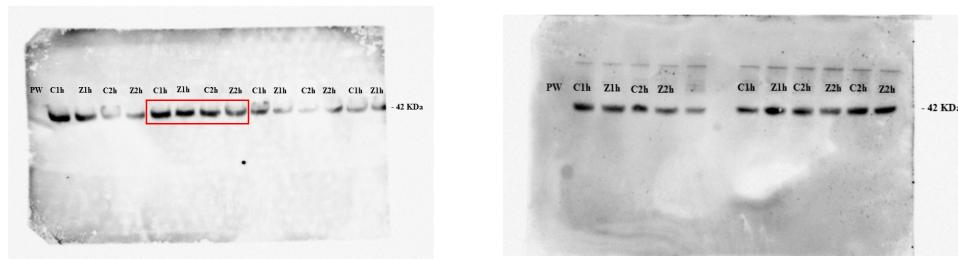


**Figure S5.** Full-length western blot membranes for NMDA-R1 detection and the respective  $\beta$ -actin controls. Red boxes highlight the bands used as representative images in the Figure S1.

### Synaptophysin (38 kDa)

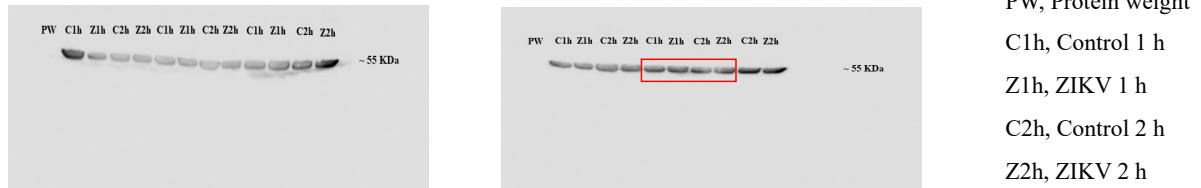


### $\beta$ -actin (42 kDa) for synaptophysin analysis

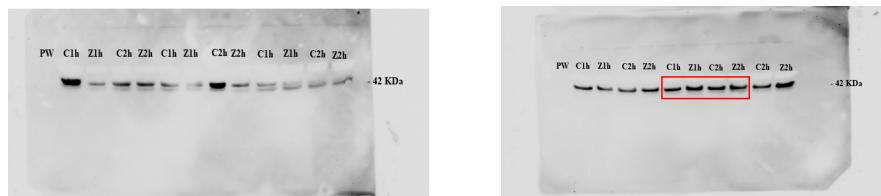


**Figure S6.** Full-length western blot membranes for synaptophysin detection and the respective  $\beta$ -actin controls. Red boxes highlight the bands used as representative images in the Figure S1.

### **$\beta$ -tubulin III (55 kDa)**



### **$\beta$ -actin (42 kDa) for $\beta$ -tubulin III analysis**



**Figure S7.** Full-length western blot membranes for  $\beta$ -tubulin III detection and the respective  $\beta$ -actin controls. Red boxes highlight the bands used as representative images in the Figure S1.