ANALYSIS OF THE HYPEREXCITABILITY OF CA3 PYRAMIDAL NEURONS IN A MOUSE-MODEL PRESENTING THE INACTIVATION OF RAC1 AND RAC3 GTIPASES

Francesca Talpo1, Paolo Spaiardi1, Mauro Toselli1, Ivan De Curtis2, Gerardo Biella1

1) University of Pavia, Dept. of Physiology, via Forlanini 6-27100 Pavia, Italy;
2) Cell Adhesion Unit, San Raffaele Scientific Institute and University Vita-Salute San Raffaele, Via Olgettina 58-20132 Milano, Italy

INTRODUCTION

Rac1 and Rac3 are small GTPases, belonging to the Rho family. These proteins regulate the actin cytoskeleton and are co-expressed in the developing mammalian brain. Recently, double knockout mice (or Rac1 and Rac3 (Rac1/Rac3−/−) mice) were produced and characterized (Cortella et al., 2009). Rac1/Rac3−/− mice showed significant cytoarchitectonic modifications and an evident epileptic phenotype. These animals died approximately thirteen days after birth. The main cytoarchitectonic modification revealed by the anatomical analysis of Rac1/Rac3−/− mice was a drastic bilateral reduction of hilar mossy cells and of parvalbumin-positive GABAergic interneurons in the dorsal hippocampus. A reduction of parvalbumin-positive GABAergic interneurons was also observed in rats. These anatomical modifications appeared to result in a marked increase in excitability of the nervous tissue.

The aim of this work was to test electrophysiologically which neuronal elements and/or which synaptic connections were impaired in Rac1/Rac3−/− mice to produce the epileptic phenotype.

MATERIALS AND METHODS

Animal model. In this study, two groups of animals (PI1/F15) were used. The experimental group consisted of 9 Rac1/Rac3−/− mice. These animals did not express Rac1 and Rac3 at neuronal level. The control group consisted of 10 Rac3−/− mice, littermates of Rac1/Rac3−/− mice. These animals did not express Rac1 and Rac3 at neuronal level.

Electrophysiological methods. Whole-cell patch-clamp experiments in current clamp mode were performed on hippocampal CA3 pyramidal neurons (Rac1/Rac3−/− mice, n=15; Rac3−/− mice, n=17) in submerged slice preparation (300 μm thickness).

RESULTS

Following 4-AP perfusion spontaneous EPSPs frequency increases more in Rac1/Rac3−/− than in Rac3−/− mice (control mice)

The epileptogenic threshold of CA3 pyramidal neurons is lower in Rac1/Rac3−/− mice than in Rac3−/− control animals. The lack of neuronal Rac1 and Rac3 proteins impairs the migration of parvalbumin-positive GABAergic interneurons during the late phases of hippocampal development. The reduced number of GABAergic interneurons at hippocampal level in Rac1/Rac3−/− mice produces an imbalance of the hippocampal excitability, that gives rise to an evident epileptic phenotype.

CONCLUSIONS

- The epileptogenic threshold of CA3 pyramidal neurons is lower in Rac1/Rac3−/− mice than in Rac3−/− control animals.
- The lack of neuronal Rac1 and Rac3 proteins impairs the migration of parvalbumin-positive GABAergic interneurons during the late phases of hippocampal development. The reduced number of GABAergic interneurons at hippocampal level in Rac1/Rac3−/− mice produces an imbalance of the hippocampal excitability, that gives rise to an evident epileptic phenotype.

Bibliography