中南大学学报(医学版) 2011, 36(11) 1071-1078 DOI: 10.3969/j.issn.1672-7347.2011.11.007 ISSN: 1672-7347 CN: 43-1427/R

论著

Pilocarpine诱导小鼠癫痫持续状态发作后海马神经元的兴奋激活、损伤和死亡

刘建新[1], 唐锋儒[2], 刘勇[1]

1. 西安交通大学医学院神经生物学研究所, 环境与疾病相关基因教育部重点实验室, 西安 710061;
2. 新加坡国立大学淡马锡实验室, 新加坡 117411

摘要：

目的: 研究癫痫持续状态发作后海马神经元兴奋激活、细胞损伤和细胞死亡的发生和相互关系。方法: 采用pilocarpine诱导Swiss小鼠癫痫持续状态(status epilepticus, SE)模型,分别以c-Fos,Fluoro Jade B和CFV染色分析SE后不同时间点齿状回和CA1区锥体细胞的兴奋激活、损伤和细胞存活状况。结果: 在齿状回颗粒细胞层,c-Fos阳性细胞在SE后1,2和24 h增多($P<0.01$或0.05),但各组齿状回颗粒细胞层均无明显Fluoro Jade B阳性细胞,CFV染色标记的阳性细胞数量在对照组和各实验组之间差异无统计学意义($P>0.05$);门区神经元在SE后没有明显的c-Fos诱导表达,但SE后2和24 h,门区Fluoro Jade B阳性细胞数量较对照组增多($P<0.01$),CFV染色显示SE后1 d门区残存的神经元数量较对照组减少($P<0.01$); CA1区锥体细胞层c-Fos阳性细胞数量在SE后30 min, 1,2和24 h后增多 ($P<0.01$ 或0.05),Fluoro Jade B阳性细胞数量在SE后2和24 h也较对照组增多($P<0.01$),但CFV染色CA1区锥体细胞数量在各组间差异无统计学意义($P>0.05$)。结论: 齿状回颗粒细胞、门区中间神经元以及CA1区锥体细胞在SE后的兴奋激活、颗粒细胞的损伤和死亡之间无直接的必然关系。

关键词： 癫痫持续状态 海马 c-Fos 神经元

Neuron activation, degeneration and death in the hippocampus of mice after pilocarpine induced status epilepticus

LIU Jianxin[1], TANG Fengru[2], LIU Yong[1]

1. Institute of Neurobiology, Environment and Genes Related to Diseases Key Laboratory of Education Ministry, School of Medicine, Xi'an Jiaotong University, Xi'an 710061, China;
2. Temasek Laboratories, National University of Singapore, Singapore 117411, Singapore

Abstract:

Objective To examine the occurrence of neuron activation, neurodegeneraion and cell death, and the correlation among them in the hippocampus after status epilepticus. Methods CFV, Fluoro Jade B and c-Fos staining were done at multiple time points after pilocarpine induced status epilepticus.Results In the stratum granulosum of dentate gyrus, c-Fos positive neurons increased significantly at 1 h, 2 h and 1 d after status epilepticus ($P<0.01$ or 0.05). However, almost no Fluoro Jade B staining cell was found in the stratum granulosum in the experiment and control groups, and no obvious difference was shown on the numbers of CFV staining cells in this area among all groups. In the hilus of dentate gyrus of different groups, there were no c-Fos positive neurons in all groups. In the hilus, the number of Fluoro Jade B staining cells significantly increased at 2 h and 1 d after the status epilepticus ($P<0.01$), and the number of CFV staining neurons dramatically decreased 1 d after the status epilepticus ($P<0.01$) compared with the control. In the stratum pyramidale of CA1, the numbers of c-Fos positive neurons at 30 mim, 1 h, 2 h and 1 d, and Fluoro Jade B staining cells at 2 h and 1 d after the status epilepticus significantly increased($P<0.01$ or 0.05), while no obvious difference in the number of CFV staining cells in the stratum pyramidale of CA1 among different groups was shown.Conclusion There is no direct correlation among cell activation, neuron degeneration and cell death in the hippocampus of mice after pilocarpine induced status epilepticus.

Keywords: status epilepticus hippocampus c-Fos neuron

收稿日期 2011-04-06 修回日期 网络版发布日期

DOI: 10.3969/j.issn.1672-7347.2011.11.007

基金项目:

This work was supported by Research Fund for the Doctoral Program of Higher Education of China (20100201120067) and Guang Hua Medical Research Grant from Xi'an Jiaotong University,P.R. China (GH0203411).

通讯作者: LIU Yong,E-mail:liuy5599@mail.xjtu.edu.cn;TANG Fengru, E-mail:tangfr@gmail.com

作者简介: LIU Jianxin, Ph.D., lecture, mainly engaged in the reserch of epilepsy.

作者Email: liuy5599@mail.xjtu.edu.cn; tangfr@gmail.com

参考文献：

[1] Cavalheiro E A, Leite J P, Bortolotto Z A, et al. Long-term effects of pilocarpine in rats: structural damage of the brain triggers kindling and spontaneous recurrent seizures [J] . Epilepsia, 1991, 32(6):778-782.

[2] Turski W A, Cavalheiro E A, Bortolotto Z A, et al. Seizures produced by pilocarpine in mice: a behavioral,

electroencephalographic and morphological analysis [J] . Brain Res, 1984, 321(2):237-253.

[3] Peng Z, Houser C R. Temporal patterns of fos expression in the dentate gyrus after spontaneous seizures in a mouse model of temporal lobe epilepsy [J] . J Neurosci, 2005, 25(31):7210-7220.

[4] Fabene P F, Andrioli A, Priel M R, et al. Fos induction and persistence, neurodegeneration, and interneuron activation in the hippocampus of epilepsy-resistant versus epilepsy-prone rats after pilocarpine-induced seizures [J] . Hippocampus, 2004, 14(7):895-907.

[5] Herrera D G, Robertson H A. Activation of c-fos in the brain [J] . Prog Neurobiol, 1996, 50(2/3):83-107.

[6] Schmued L C, Albertson C, Slikker W Jr. Fluoro-Jade: a novel fluorochrome for the sensitive and reliable histochemical localization of neuronal degeneration [J] . Brain Res, 1997, 751(1):37-46.

[7] Schmued L C, Hopkins K J, Fluoro-Jade B. A high affinity fluorescent marker for the localization of neuronal degeneration [J] . Brain Res, 2000, 874(2):123-130.

[8] Poirier J L, Capek R, De Koninck Y. Differential progression of dark neuron and Fluoro-Jade labelling in the rat hippocampus following pilocarpine-induced status epilepticus [J] . Neuroscience, 2000, 97(1):59-68.

[9] Mirzaeian L, Ribak C E. Immunocytochemical mapping of Fos protein following seizures in gerbils indicates the activation of hippocampal neurons [J] . Hippocampus, 2000, 10(1):31-36.

[10] Liu J X, Cao X, Tang Y C, et al. CCR7, CCR8, CCR9 and CCR10 in the mouse hippocampal CA1 area and the dentate gyrus during and after pilocarpine-induced status epilepticus [J] . J Neurochem, 2007, 100(4):1072-1088.

[11] Liu J X, Tang Y C, Liu Y, et al. mGluR5-PLCbeta4-PKCbeta2/PKCgamma pathways in hippocampal CA1 pyramidal neurons in pilocarpine model of status epilepticus in mGluR5+/+ mice [J] . Epilepsy Res, 2008, 82(2/3):111-123.

[12] Liu J X, Liu Y, Tang F R. Pilocarpine-induced status epilepticus alters hippocampal PKC expression in mice [J] . Acta Neurobiol Exp (Wars), 2011, 71(2):220-232.

[13] Parent J M, Yu T W, Leibowitz R T, et al. Dentate granule cell neurogenesis is increased by seizures and contributes to aberrant network reorganization in the adult rat hippocampus [J] . J Neurosci, 1997, 17(10):3727-3738.

[14] Scharfman H E, Goodman J H, Sollas A L. Granule-like neurons at the hilar/CA3 border after status epilepticus and their synchrony with area CA3 pyramidal cells: functional implications of seizure-induced neurogenesis [J] . J Neurosci, 2000, 20(16):6144-6158.

大鼠的镇痛效应
[J]. 中南大学学报(医学版), 2008,33(06): 481-487
14. 王岐本1,2， 蒙艳斌1， 黄庆红1， 谢乐斯3， 何旭峰2，潘爱华2.大鼠脑血管、神经元和星形胶质细胞的空间构筑[J]. 中南大学学报(医学版), 2008,33(07): 592-595
15. 吴志国， 龙莉莉， 肖波， 陈锶， 易芳 .匹罗卡品致痫大鼠海马神经肽Y中间神经元数目变化及其轴突出芽[J]. 中南大学学报(医学版), 2009,34(02): 93-98
