

在嬰兒點頭痙攣病人和紅藻胺酸誘發抽搐之幼鼠其親皮質素釋放激素及細胞激素的表現

The Expression of Corticotropin-Releasing Hormone(CRH) and Cytokines in Patients of Infantile Spasm and in Neonatal Rats with Kainate-Induced Seizure

中文摘要

嬰兒點頭痙攣(IS)是嬰兒時期特有的一種癲癇疾病。病嬰對一般抗痙攣藥物治療效應較差，然而對投與促腎上腺皮質素(ACTH)或類固醇的治療有明顯較佳的效果。ACTH的分泌主要是受到下視丘的親皮質素釋放激素(CRH)的調控，且已證實CRH此神經荷爾蒙(neurohormone)在人類幼年癲癇或幼鼠痙攣的發生扮有相當重要的角色。因而有學者提出「CRH的過量產生導致IS發生」之假說，此外在動物實驗方面，於幼鼠腦室注射CRH比起成鼠能更快速的引起痙攣。近年來許多研究發現細胞激素具有調節內分泌的功能，並參與許多中樞神經系統疾病的過程進展。基於以上原因，為更深入了解CRH在癲癇所參與的病理機轉，我們測量IS病人其周邊血液CRH濃度的變化是否和IS有關？或在以紅藻胺酸(KA)引發癲癇的出生後7天、14天、30天老鼠其抽搐程度的加重是否能使CRH和細胞激素(IL-1, IL-6, TNF- α)在周邊或腦中表現增加？實驗結果指出，不論是IS病人血漿中CRH濃度(0.546(0.184 ng/ml, n=12, p<0.001)或是年齡相符的一般癲癇病人(0.294(0.032 ng/ml, n=13, p<0.01)都較控制組病人(0.135(0.022 ng/ml, n=12)為高。在注射KA後3小時的出生30天老鼠(P30)其血中CRH濃度(0.368(0.06 ng/ml, n=4, p<0.05)明顯較控制組為高；但在注射KA後1小時的P14天老鼠其血中CRH濃度(0.260(0.04 ng/ml, n=6)和控制組比起則無統計上的差異。此外以反轉錄聚合酶連鎖反應方法半定量測量到CRH mRNA在P7、P14天注射KA老鼠的大腦皮質部位之表現量分別較控制組增加了7.84倍及1.55倍；另外IL-1 α 、IL-6、TNF- α mRNA也在P7天注射KA老鼠的大腦皮質部位增加表現量。以上結果顯示支持CRH的過度產生和IS有極大的相關，並推測在腦部發育過程中CRH可能是一個致痙攣前驅物(pro-convulsant)，而因癲癇所導致CRH在大腦皮質的表現增加，進而活化細胞激素可能導致腦部不正常之過度興奮現象而對癲癇程度的進展極為重要。

英文摘要

Infantile spasm (IS), is a form of epilepsy specifically occurring at infant stage, is refractory to conventional anticonvulsant treatment but is exceptionally sensitive to adrenocorticotrophic hormone (ACTH) treatment. Corticotropin-releasing hormone (CRH), a major regulatory factor of ACTH secretion in pituitary gland, is secreted by the hypothalamus. It has been demonstrated that CRH plays an important role in the pathogenesis of infantile spasm. Some researchers have proposed the hypothesis that "excessive production of CRH leads to IS". In addition, in animal experiments, intracerebroventricular injection of CRH in neonatal rats induced seizures more rapidly than in adult rats. In recent years, many studies have found that cytokines have a regulatory function on endocrine and participate in the progression of many central nervous system diseases. Based on the above reasons, to further understand the pathogenesis of CRH in epilepsy, we measured the change of CRH concentration in peripheral blood of IS patients and whether it was related to IS? Or in neonatal rats with kainic acid (KA) induced epilepsy, whether the increase of seizure severity could increase the expression of CRH and cytokines (IL-1 α , IL-6, TNF- α) in peripheral blood or brain? The results showed that, whether in IS patients plasma CRH concentration (0.546 (0.184 ng/ml, n=12, p<0.001) or in age-matched general epilepsy patients (0.294 (0.032 ng/ml, n=13, p<0.01) were higher than control group patients (0.135 (0.022 ng/ml, n=12). In rats injected with KA for 3 hours at P30, CRH concentration in blood (0.368 (0.06 ng/ml, n=4, p<0.05) was significantly higher than control group; but in rats injected with KA for 1 hour at P14, CRH concentration (0.260 (0.04 ng/ml, n=6) and control group were not significantly different. In addition, reverse transcription-polymerase chain reaction (RT-PCR) method was used to semi-quantitatively measure CRH mRNA expression in the cerebral cortex of P7 and P14 rats injected with KA. The results showed that CRH mRNA expression in the cerebral cortex of P7 and P14 rats injected with KA was 7.84 and 1.55 times higher than control group, respectively. In addition, IL-1 α , IL-6, and TNF- α mRNA expression in the cerebral cortex of P7 rats injected with KA was also increased. The above results support the hypothesis that excessive production of CRH is closely related to IS, and it is speculated that CRH may be a pro-convulsant in the development of the brain, and the increase of CRH expression in the cerebral cortex caused by epilepsy may lead to excessive excitation phenomenon in the brain, which is very important for the progression of epilepsy.

ary, has been implicated as an important neuro-hormone participating in seizure generation particularly in early life of both human and animals. In human, excessive production of CRH is speculated as one of the pathological mechanisms underlying the generation of IS. In animal, intracerebral injection of CRH potently produces seizure behavior since early life. Recently, cytokines also participated in communication between immunity and central nerve system diseases. To further understand the role of CRH in pathological mechanisms of seizure, we investigated whether increased plasma CRH level is associated with IS and whether seizure activity will increase the expression of CRH and cytokines (IL-1, IL-6 and TNF- α) in circulation and in brain of neonatal rats. Plasma CRH level in IS patients (0.546(0.184 ng/ml, n=12, P<0.001) or seizure patients (0.294(0.032 ng/ml, n=13, P<0.01) is significantly higher as compared to control (0.135(0.022 ng/ml, n=12). The plasma CRH level of postnatal 30-day (P30) rats at 3h after kainate (KA)-injection (0.368(0.06 ng/ml, n=4) is significantly higher (p<0.05) than that of control, whereas, plasma CRH level show no statistically difference in P14 rats at 1h after KA-injection (0.260(0.04 ng/ml, n=6) when compare to that of control. Furthermore, an 7.84 and 1.55 fold increased expression of CRH mRNA in cortex was observed in P7 and P14 rats with KA-induced seizure by RT-PCR analysis. In addition, IL-1, IL-6 and TNF- α mRNA expression were increased in cortex of P7 rats with KA-induced seizure. These data support the hypothesis that over-production of CRH is likely associated with IS, and seizure activity induced by KA may also upregulate the CRH and cytokines expression in neonatal developing brain. Given that CRH is a pro-convulsant in developing brain, increase of CRH in cortex elicited by seizure may further potentiate the progression of the activity of seizure.