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N-乙基-N-亚硝基脲诱发大鼠胶质瘤的实验研究*

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提 要 采用人工合成的N-乙基-N-亚硝基脲(ENU,60 mg/kg体重),经胎盘和皮下给药分别在妊娠晚期孕鼠子代和3天龄Wistar大鼠经12个月诱发出神经系统胶质瘤。成瘤率分别为73.2%和68.3%(胶质瘤为65.9%和63.4%);胶质瘤以少突-星形细胞瘤和少突胶质细胞瘤为主,呈小灶性、多灶性、混合性,并伴有灶性和/或弥漫性胶质增生存在。结果表明,这两种胶质瘤模型为深入研究胶质瘤的发生、生长及分化提供了良好的途径。本文讨论了灶性胶质母细胞增生在胶质瘤发生学上的意义。

关键词 神经胶质瘤/化学诱导;乙基亚硝基脲;疾病模型,动物;大鼠
中图法分类号 R739.264

脑胶质瘤因其部位的特殊性使临床病理学研究难以深入,故建立有效而稳定的脑胶质瘤动物模型很有意义。国外应用乙基亚硝基脲(ethylnitrosourea,ENU)诱发动物胶质瘤并应用于肿瘤病理学研究的报道很多[1~3],国内于1990年首次合成了ENU并用于SD大鼠恶性神经鞘瘤诱发实验[4]。我们最近亦采用了自行合成的ENU建立诱发Wistar大鼠胶质瘤的两种模型,以探讨体内胶质瘤的发生与防治。

1 材料与方法

1.1 诱发肿瘤实验

ENU合成后[4],用0.01 mol/L PBS(pH7.20)在一定温度下搅拌约3 min即可缓慢溶解ENU,分别配制成0.5%和1% ENU溶液,过滤除菌,按60 mg/kg体重给药。

1.1.1 经孕鼠胎盘给药致子代鼠成瘤:选用正常妊娠的健康Wistar大鼠(体重330~350 g)10只,随机分为ENU1和对照1两组。ENU1组(7只)在妊娠晚期(距预产期前7 d)经尾静脉一次性缓慢注射1%ENU溶液约2 ml/只。

1.1.2 经新生幼鼠皮下给药致瘤:选用3 d龄健康Wistar大鼠7窝,共69只幼鼠(体重3.5~4.4 g),随机分为ENU2和对照2两组。ENU2组于幼鼠右肩胛部(25只)或腰骶部(29只)、一次性皮下注射0.5%ENU溶液(每只约50 μl)。

对照1和对照2组则以相应体积的0.01 mol/L PBS(pH7.20)代替ENU在上述部位分别注射。

1.2 取材及标本处理

分别于注射后第4、6、8个月末每组随机选2只活杀取材,其余动物于第12个月末取材。乙醚麻醉,颈动脉插管,4%多聚甲醛灌注固定,迅速取脑、脊髓和各系统主要脏器及所有肉眼所见的肿瘤。完整取出脑组织后于不同平面至少切成7片观察。标本分别用4%甲醛、4%多聚甲醛(4℃)固定,对可疑肿瘤病灶迅速取电镜标本。常规进行光镜、电镜制样。部分标本进行抗GFAP、S-100蛋白和CD3、CD20免疫组化(ABC法)以明确诊断。

2 结果

2.1 动物的一般情况

ENU1组孕鼠给药后精神差,24 h后恢复正常。仔鼠娩出后抵抗力较弱(共娩出仔鼠49只,其中有8只先后死于非肿瘤因素)。ENU2组幼鼠给药后活动及食欲差,一周内易死亡(共死亡13只)。

上述所有给药动物成年后繁殖力低下。对照2组共2只大鼠死于非肿瘤性疾病。死于非肿瘤因素的动物均经全身解剖病检证实。

2.2 诱发胶质瘤情况(诱瘤结果见附表)

附表 乙基亚硝基脲诱发大鼠胶质瘤的结果

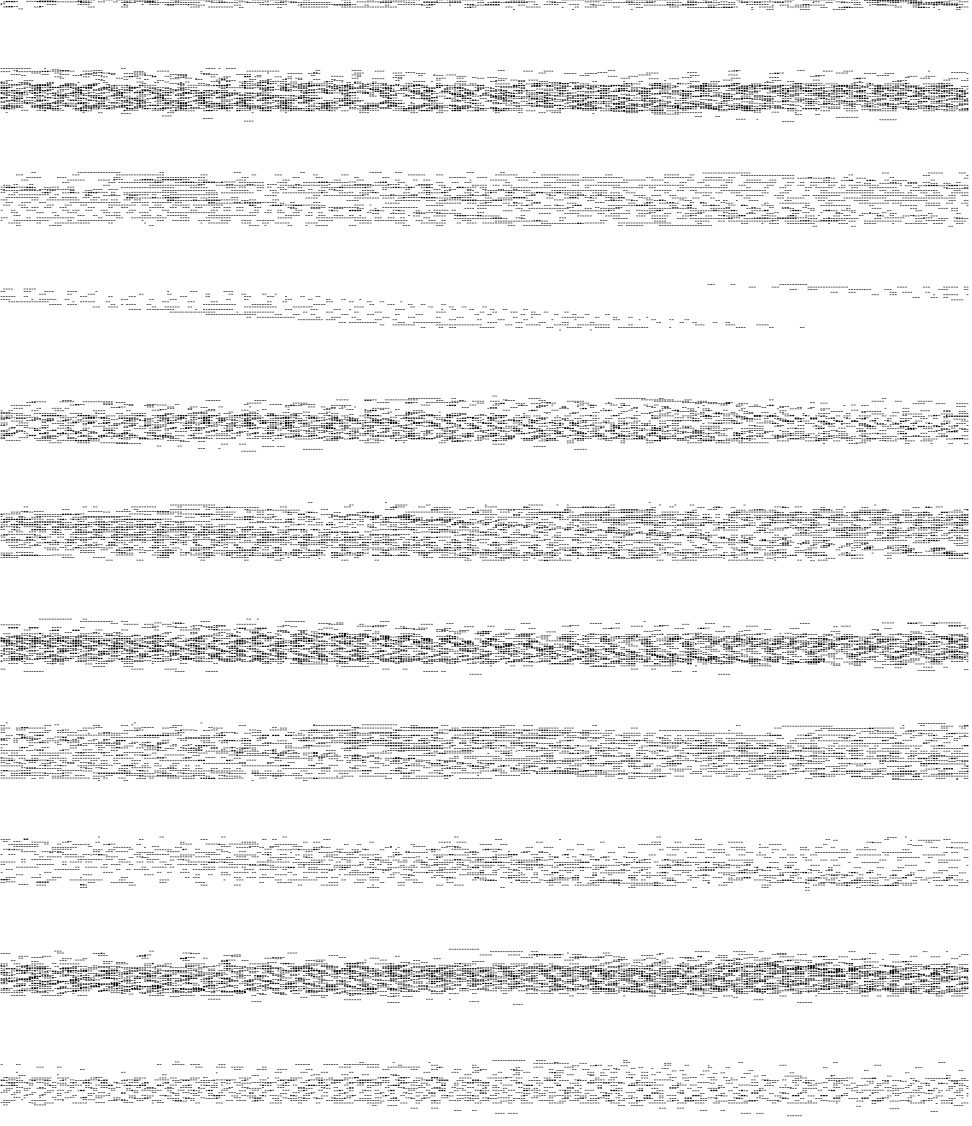
Tab The result of ethylnitrosourea-induced gliomas in rats

Groups	N	Nt	Glioma foci					Types of gliomas		Other tumors
			total	Cb	Cl	Bs	Sc	mixed(Ⅰ/Ⅱ)	oligo(Ⅰ/Ⅱ)	
ENU2	41	27	34	24	4	3	3	25(8/17)	9(4/5)	8
control2	30	0	0	0	0	0	0	0	0	0
ENU2	41	26	30	21	2	5	2	21(12/9)	9(6/3)	5
control2	13	0	0	0	0	0	0	0	0	0

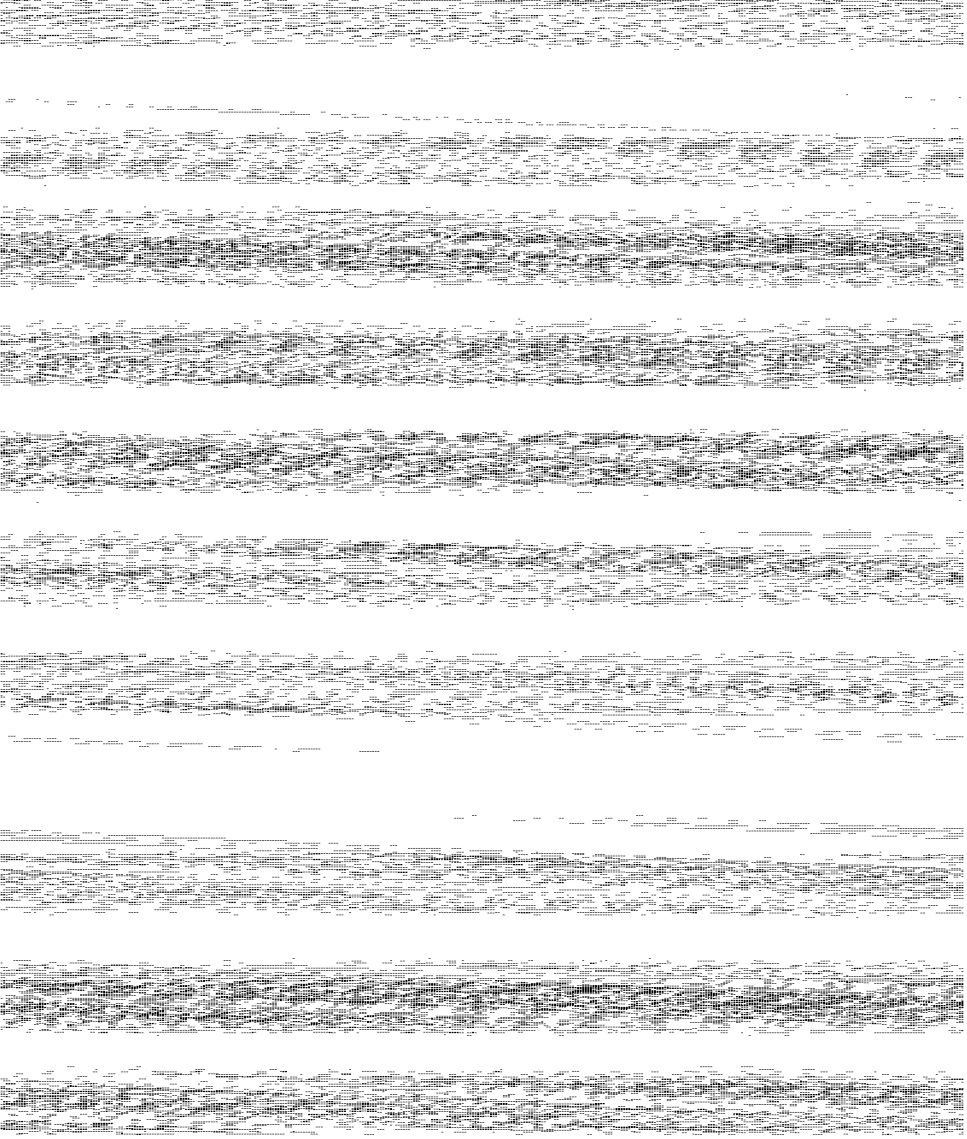
N: the numbers of rats observved, not including the rats died of other diseases rather than tumors; Nt: the number of rats with gliomas. Cb: cerebrum; Cl: cerebellum; Bs: brian stem; Sc: spinal cord; mixed: mixed oligodendro-astrocytomas; oligo: oligodendrogliomas; Ⅰ/Ⅱ number of grade Ⅰ/that of grade Ⅱ.

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2 Reifenberger G, Bilzer T, Seitz RJ, et al. Expression of vimentin and glial fibrillary acidic protein in ethylnitrosourea-induced rat gliomas and glioma cell lines. Acta Neuropathol, 1989,78:270
3 Vogeley KT, Reifenberger G, Bilzer T, et al. Immunochemistry of ethylnitrosourea-induced rat neurinomas, the RN6 neurinoma cell line and their transplantation tumors. Acta Neuropathol,1991,82(1):78
4 陶海鹏,杨光华,黄 跃. N-乙基-N-亚硝基脲的合成和动物实验. 华西医科大学学报,1990,21(2):163
5 Walker VE, Swenberg JA. Phenobarbital lacks promoting activity for neurogenic tumors in F344 rat transplacentally exposed to ethylnitrosourea. J Neuropathol Exp Neurol, 1989,48(3):263
6 Yoshino T, Motoi M, Ogawa K. Morphological maturation of tumor cells induced by ethylnitrosourea (ENU) in rat brain. I. Other tumors by administration of ENU in the late gestational stage. Acta Pathol Jpn,1985,35(6):1385
7 Yoshino T. Morphological maturation of tumor cells induced by ethylnitrosourea (ENU) in rat brain. II. Other tumors by administration of ENU in the mid-gestational stage. Acta Pathol Jpn,1985,35(6):1397
8 Galloway PG, Likavec MJ, Perry G. Immunohistochemical recognition of ethylnitrosourea induced rat brain microtumors by anti-Leu 7 mono-clonal antibody. Cancer Lett, 1990,49(3):243
9 文剑明,Emura M,Mohr U. 乙基亚硝脲诱导人和地鼠胚胎肺上皮细胞点突变比较. 中华肿瘤杂志,1991,13(4):253

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A study to induce brain glioma with N-ethyl-N-nitrosourea in rats

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Abstract In order to induce brain glioma, transplacental and subcutaneous injections of N-ethyl-N-nitrosourea (ENU, 60 mg/kg body weight) were given to the Wistar rats in late gestational stage and of 3 days of age. The rats were followed up until the end of the 12th month after drug administration. The rate of total tumor formation was 73.2% and 68.3% and that of brain glioma was 65.9% and 63.4% respectively. Histologically, the main types of glioma were mixed oligodendro-astrocytoma and oligodendroglioma. The tumors exhibited the microfocal, multifocal and mixed features with focal and/or diffuse proliferation of glial cells. These findings indicate that the 2 rat models of glioma induced with synthetic N-ethyl-N-nitrosourea were successful and provide a reliable approach for the study of the pathogenesis of cerebral glioma. In addition, the significance of the proliferation of focal glioblasts in the genesis of cerebral glioma was discussed.

Key words glioma/CI; N-ethyl-N-nitrosourea; disease models, animal; rats

幼儿巨大喉乳突状瘤一例

A case of child large laryngeal papilloma

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患儿男性,4岁。鼾声响4年,声嘶伴呼吸费力半年,加重2周以喉部新生物于1993年4月8日入院。患儿出生后不久偶然发现腭部有一赘生物,睡眠时鼾声比同龄婴儿响。一岁时腭部赘生物已长至黄豆大。院外曾行赘生物切除,组织病理报告为腭部乳突状瘤。手术后鼾声未减。1992年10感冒后出现声音嘶哑,活动后有呼吸困难和喉鸣,睡眠时鼾声加重。经一般治疗声嘶无改善,近二周出现明显吸气性呼吸困难的喉鸣,鼾声加重。睡眠时有呼吸暂停现象,并经常惊醒。

家族中无类似疾病史。其母妊娠期无异常。体格检查:发育正常,一般情况好。周身皮肤无异常包块。鼻翼扇动,吸气时可见明显"三凹"体征。心,肺,肝,脾未发现异常。间接喉镜下见喉前庭有巨大新生物,浅红色,表面粗糙不平,声门被新生物覆盖。入院诊断:喉新生物(性质待诊)。喉梗阻Ⅱ°。入院后于4月11日晚22点30分,突然出现Ⅲ°呼吸困难。患儿烦躁不安,嘴唇紫绀,大汗淋漓,不能平卧。即刻给予吸氧并行紧急气管切开

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