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论文

OPIDN致鸡大脑脑型肌酸激酶差异表达

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摘要：

目的 探讨有机磷化合物致迟发性神经毒性（OPIDN）鸡脑组织中脑型肌酸激酶同工酶（CK-BB）表达变化。方法 罗曼鹤母鸡24只，随机分成三磷甲苯磷酸酯（TOCP）染毒组（1 000 mg/kg）、苯甲基磺酰氟（PMSF）干预组（在染毒之前给予40 mg/kg PMSF）和对照组（生理盐水），每组8只。染毒第5和第21天，每组分别处死8只动物，分离大脑，提取总蛋白；用双向电泳和质谱分析技术，筛选相关差异表达蛋白。结果 蛋白双向电泳结果显示，在染毒第5天，TOCP组与对照组、PMSF干预组与对照组的CK-BB蛋白点灰度比值分别为0.858 3和0.913 3，各组间差异无统计学意义（$P>0.05$）；染毒第21天，TOCP组和PMSF干预组与对照组差异蛋白点CK-BB灰度比值分别为0.327 5与0.675 7，前者表达下调达3.0倍；对该蛋白点的质谱分析结果在Swiss-por数据库中进行比对，其与鸡种属、蛋白序列号为gi|45384340的creatine kinase B-type（CK-BB）蛋白的肽段匹配数为26，序列覆盖率为75%，评分值为1 603（可信度高），显示该蛋白点即为CK-BB蛋白。结论 OPIDN可致鸡大脑组织CK-BB蛋白表达显著下调，其变化可能与OPIDN模型动物神经组织损伤有关。

关键词： 迟发性神经毒性(OPIDN) 脑型肌酸激酶同工酶(CK-BB) 三磷甲苯磷酸酯(TOCP) 苯甲基磺酰氟(PMSF) 双向电泳分析

Differential expression of CK-BB in brain tissue of hens with OPIDN

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Abstract:

Objective To detect differential expressions of brain type isoenzyme of creatine kinase(CK-BB)in the brain of hens with organophophors compounds induced delayed nerutoxicity(OPIDN)and to provide target protein evidence for diagnosis of POIDN.Methods Twenty-four Roman hens were randomly divided into three groups:treated group(1 000 mg/kg triorthocresyl phosphate,TOCP),intervened group (40 mg/kg phenylmethanesul fonyl fluoride [PMSF] before TOCP treatment),and control group(tap water).Eight hens in each group were sacrificed at day 5 and day 20 after the treatments;then,the brain tissue of the hens were seperated at low temperature and total protein of the brain tissue were extracted.With solid phase PH gradient sodium dodecyl sulfate-polyacrylamide gel electrophoresis(SDS-PAGE)and two-dimensional electrophoresis and mass spectrometry,we detected differently expressed CK-BB protein.Results Compared with the control group,the difference in CK-BB expression was not significant at day 5($P<0.05$),while at day 20,the gray level ratio of CK-BB expression of treated-group and intervened-group were 0.327 5 and 0.675 7,with a three-fold down-regulation in the treated group.Mass spectrometry and Swiss-por data analysis identified CK-BB spot with peptide segments of 26,sequence coverage of 75%,and the assessment value of 1 603(≥45 for high reliability)in relation to the species of the hen and protein sequence number gi|45384340 of CK-BB.Conclusion The expression of CK-BB is down-regulated in the brain of hens with OPIDN,which may related to brain injury induced by OPIDN.

Keywords: organophosphorus compounds induced delayed neurtoxicity(OPIDN) brain type isoenzyme of creatine kinase(CK-BB) triorthocresyl phosphate(TOCP) phenylmethanesul fonyl fluoride(PMSF) two-dimensional electrophoresis

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