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不同壳聚糖浓度培养液对断奶仔猪外周血淋巴细胞中花生四烯酸代谢的影响

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Effects of Different Chitosan Concentrations in Medium on Arachidonic Acid Metabolism in Peripheral Blood Lymphocytes of Weaner Piglets

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摘要 为了研究壳聚糖对断奶仔猪外周血淋巴细胞(PBLs)中花生四烯酸(AA)代谢的影响, 试验分离提取断奶仔猪PBLs, 在含有0、40、80、160和320 $\mu\text{g}/\text{mL}$ 壳聚糖的培养液中进行培养48 h后, 测定培养液上清液中胞浆型磷脂酶A2(cPLA2)、环氧合酶-2(COX-2)和5-脂氧化酶(5-LOX)的活性及AA、前列腺素E2(PGE2)和白三烯B4(LTB4)的浓度; 测定细胞中cPLA2、COX-2和5-LOX mRNA的相对表达量。结果表明: 随着壳聚糖添加水平的增加, 淋巴细胞培养液中的cPLA2、COX-2和5-LOX的活性及AA、PGE2和LTB4浓度呈显著的二次曲线变化($P<0.05$); 淋巴细胞中的cPLA2、COX-2和5-LOX mRNA的相对表达量与壳聚糖添加水平也呈显著的二次剂量依赖关系($P<0.05$)。上述所有指标的最大值出现在壳聚糖的添加水平为80~160 $\mu\text{g}/\text{mL}$ 时, 而当壳聚糖的添加水平增加到320 $\mu\text{g}/\text{mL}$ 时其值有不同程度的降低。结果提示, 在本试验中, 适当水平的壳聚糖可以促进断奶仔猪PBLs内AA代谢网络途径中参与炎症或免疫调节的主要介质PGE2和LTB4的产生、关键控制酶cPLA2、COX-2和5-LOX的活性及其基因表达, 这可能是其发挥免疫调节功能的原因之一。

关键词: 壳聚糖 断奶仔猪 外周血淋巴细胞 花生四烯酸代谢 免疫功能

Abstract: This experiment was conducted to investigate the effects of different levels of chitosan in medium on the metabolism of arachidonic acid in peripheral blood lymphocytes of weaner piglet. The peripheral blood lymphocytes from weaner piglets were cultured in medium supplemented with 0 (control), 40, 80, 160 and 320 $\mu\text{g}/\text{mL}$ chitosan/mL medium, respectively. At the end of 48 h incubation, supernatants were collected for analyzing the activities of cytosolic phospholipase A2 (cPLA2), cyclooxygenase-2 (COX-2) and 5-lipoxygenase (5-LOX) as well as the concentrations of arachidonic acid (AA), prostaglandin E2 (PGE2) and leukotriene B4 (LTB4). The cells were harvested and analyzed for the mRNA expressions of cPLA2, COX-2 and 5-LOX genes. Results showed that chitosan increased the activities of cPLA2, COX-2 and 5-LOX as well as the concentrations of AA, PGE2 and LTB4 in supernatants in a quadratic dose-dependent manner ($P<0.05$). With the chitosan supplemental level increasing, the mRNA expressions of cPLA2, COX-2 and 5-LOX genes in peripheral blood lymphocytes was enhanced in a quadratic manner ($P<0.05$). In addition, the supplementation of 80 to 160 $\mu\text{g}/\text{mL}$ chitosan to the medium had higher values of all above indexes compared with those in other treatments. However, positive effects of chitosan tended to be suppressed when its supplemental level was increased to 320 $\mu\text{g}/\text{mL}$. It is concluded that appropriate level of chitosan can increase the concentrations of AA, PGE2 and LTB4, and improve the activities of cPLA2, COX-2 and 5-LOX as well as the mRNA expressions of cPLA2, COX-2 and 5-LOX genes in peripheral blood lymphocytes, which may be one of the reasons for chitosan to regulate the immune functions of weaner piglets.

Keywords: chitosan, weaner piglet, peripheral blood lymphocytes, arachidonic acid metabolism, immune function

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