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论文

黄豆铁蛋白提取新方法及其与豌豆铁蛋白活性比较

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摘要:

铁蛋白是广泛存在于动物、微生物及植物体中的一种铁贮藏蛋白, 具有去除二价铁的毒性以及调节机体细胞铁代谢平衡的作用. 本文以黄豆种子为原料, 开发出黄豆铁蛋白提取新方法, 即将黄豆粗提液在55 °C下加热15 min, 再将冷却后的上清液分别用500 mmol/L MgCl₂和700 mmol/L柠檬酸三钠进行盐析. 离心得到的铁蛋白粗提液经透析后, 用DEAE-cellulose弱阴离子交换层析和Sephacryl S-300凝胶过滤层析进一步分离, 得到电泳纯的铁蛋白. Native-PAGE电泳测得分子量约为560000, SDS-PAGE电泳结果表明, 黄豆铁蛋白含有两种亚基, 分子量分别为28000和26500. 活性研究显示, 黄豆铁蛋白与豌豆铁蛋白铁氧化沉淀和还原释放反应的活性明显不同.

关键词: 黄豆; 铁蛋白; 纯化; 活性鉴定

New Method of Soybean Seed Ferritin Purification and Comparison of Activity with Pea Seed Ferritin

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Abstract:

Ferritin is a class of iron storage protein presents in all living organisms and serves the dual function of iron detoxification and iron storage. In this paper, a new method of soybean seed ferritin purification was reported. Crude extracts of soybean were heated at 55 °C for 15 min, then adjusted to 500 mmol/L MgCl₂, stored for 30 min at 4 °C. It was followed by addition of sodium citrate to final concentration of 700 mmol/L. The protein was further purified by ion exchange chromatography in DEAE-Sepharose and Sephacryl S-300 gel filtration columns. Native-PAGE purified the molecular weight of soybean ferritin was about 560000 and SDS-PAGE analyzed ferritin subunits were separated as two peptides(28000 and 26500). Subsequently, we compared the iron oxidative deposition and reductive release activities of soybean ferritin with those of pea seed ferritin, and found that their mechanisms of the iron oxidative deposition and iron release are different. Finally, a possible reason was given.

Keywords: Soybean seed; Ferritin; Purification; Activity identification

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