催化学报 » 2011, Vol. 32 » Issue (10) :1584-1591 DOI: 10.1016/S1872-2067(10)60262-8

研究论文

丁二酸酐修饰对漆酶稳定性和除酚效率的影响

熊亚红*, 高敬忠, 郑坚鹏, 邓乃康

华南农业大学理学院生物材料研究所, 广东广州 510640

XIONG Yahong*, GAO Jingzhong, ZHENG Jianpeng, DENG Naikang

Institute of Biomaterial, College of Science, South China Agricultural University, Guangzhou 510642, Guangdong, China

- 摘要
- 参考文献
- 相关文章

Download: PDF (371KB) HTML (1KB) Export: BibTeX or EndNote (RIS) Supporting Info

摘要 采用 (NH4)2SO4 分步沉淀法对诺维信中国公司生产的漆酶制剂 DeniLite IIS 进行了纯化, 并用丁二酸酐 (SA) 对纯化酶进行了化学修饰, 运用三硝基苯磺酸法、紫外光谱法及荧光光谱法对修饰效果进行了初步表征, 比较了天然酶和修饰酶的 pH 稳定性、热稳定性及除酚效率. 结果表明, 修饰酶的平均氨基修饰度为 85%, 其紫外吸收峰和荧光发射峰均出现蓝移, 而且紫外吸收减小、荧光强度增加. 尽管采用 SA 化学修饰未能改变漆酶的最适反应温度, 但使其最适反应 pH 值由 4.5 提高到 5.5, 并且使酶活提高 60%. 与天然酶相比, 修饰酶的 pH 稳定性和热稳定性更高, 催化效率 (kcat) 和酶与底物的亲和力 (kcat/Km) 分别提高了 53% 和 122%, 对邻、间和对苯二酚的除酚效率分别提高了 48%, 57% 和 18%. 这预示着这些修饰漆酶可望应用于工业生产和酚类污染废水的治理.

关键词: 丁二酸酐 漆酶 化学修饰 稳定性 动力学 除酚效率

Abstract: Chemical modification is a useful method to change the properties of enzymes. Laccase is a phenol oxidase belonging to a multicopper protein, which catalyzes the oxidation of many phenolics. DeniLite IIS, a commercial laccase preparation from the Novozymes China Company, was purified by ammonium sulfate fractional precipitation. Succinic anhydride (SA) was used as a modifier for the chemical modification of the purified laccase. The effects of modification were characterized using the 6-trinitrobenzene sulfonic acid method, ultraviolet spectroscopy, and fluorescence spectroscopy. The pH stability, thermal stability, and the phenolics removal efficiency for the native and modified laccases were compared. The results showed that the average amino modification yield of the modified laccase was 85% and the modified laccase had a blue shifted ultraviolet peak and fluorescence emission peak as well as a decrease in the ultraviolet absorbance and an increase in the fluorescence intensity. Although chemical modification with SA did not change the optimum temperature for the catalysis of the laccase, it caused the optimum pH of the catalysts to shift from 4.5 to 5.5 and the enzymatic activity increased by 60%. Compared with the native laccase, the modified laccase exhibited remarkably higher pH stability and thermal stability and its catalysis efficiency (k_{cat}) and substrate affinity (k_{cat}/K_m) increased by 53% and 122%, respectively. The phenolics removal efficiency (o-, m-, p-dihydroxybenzene) of the modified laccase increased by 48%, 57%, and 18%, respectively. These results indicate that the modified laccase with higher stability and higher efficiency is suitable for application in industrial production and for the treatment of phenolics-polluted water.

Keywords: succinic anhydride, laccase, chemical modification, stability, kinetics, phenolics removal efficiency

收稿日期: 2011-05-16; 出版日期: 2011-09-02

引用本文:

熊亚红, 高敬忠, 郑坚鹏等 .丁二酸酐修饰对漆酶稳定性和除酚效率的影响[J] 催化学报, 2011,V32(10): 1584-1591

XIONG Ya-Hong, GAO Jing-Zhong, ZHENG Jian-Peng etc .Effects of Succinic Anhydride Modification on Laccase Stability and Phenolics Removal Efficiency[J] Chinese Journal of Catalysis, 2011,V32(10): 1584-1591

链接本文:

http://www.chxb.cn/CN/10.1016/S1872-2067(10)60262-8 或 http://www.chxb.cn/CN/Y2011/V32/I10/1584

Service

- 把本文推荐给朋友
- 加入我的书架
- 加入引用管理器
- Email Alert
- RSS

作者相关文章

- 熊亚红
- 高敬忠
- 郑坚鹏
- 邓乃康

[1] ayer A M, Staples R C. Phytochemistry, 2002, 60: 551

[2] aldrian P. FEMS Microbiol Rev, 2006, 30: 215

[3] iontek K, Antorini M, Choinowski T. J Biol Chem, 2002, 277: 37663

[4] hurston F. Microbiology, 1994, 140: 19

[5] 阳, 蒋国翔, 牛军峰, 王颖, 呼丽娟. 化学进展 (Li Y, Ji-ang G X, Niu J F, Wang Y, Hu L J. Prog Chem), 2009, 21: 2028

[6] hin-ya Y, Aye H N, Hong K J, Kajiuchi T. Enzyme Microb Technol, 2005, 36: 147

[7] ann C M, Qi D F, Distefano M D. Curr Opin Chem Biol, 2001, 5: 696

[8] yer P V, Ananthanarayan L. Process Biochem, 2008, 43: 1019

[9] 建忠, 宋海燕, 翁丽萍, 计亮年. 分子催化 (Liu J Z, Song H Y, Weng L P, Ji L N. J Mol Catal(China)), 2002, 16: 475

[10] DeSantis G, Jones J B. Curr Opin Biotechnol, 1999, 10: 324

[11] Vandertol-Vanier H A, Vazquez-Duhalt R, Tinoco R, Pickard M A. J Ind Microbiol Biotechnol, 2002, 25: 214

[12] Schroeder M, Heumann S, Silva C J S M, Cavaco-Paulo A, Guebitz G M. Biotechnol Lett, 2006, 28: 741

[13] 李苏, 闫明, 戈钧, 刘铮. 化工学报 (Li S, Yan M, Ge J, Liu Z. J Chem Industry Eng (China)), 2007, 58: 3046

[14] Forde J, Tully E, Vakurov A, Gibson T D, Millner P, Ó'Fágáin C. Enzyme Microb Technol, 2010, 46: 430

[15] Song H Y, Yao J H, Liu J Z, Zhou S J, Xiong Y H, Ji L N. En-zyme Microb Technol, 2005, 36: 605

[16] 熊亚红, 苏健鸿, 刘小平. 分子催化 (Xiong Y H, Su J H, Liu X P. J Mol Catal(China)), 2010, 24: 435

[17] 熊亚红, 吴巧儿, 苏健鸿. 分子催化 (Xiong Y H, Wu Q E, Su J H. J Mol Catal(China)), 2010, 24: 529

[18] Bourbonnais R, Paice M G. Appl Microbiol Biotechnol, 1992, 36: 823

[19] Laemmli U K. Nature, 1970, 227: 680

[20] Lowry O H , Rosebrough N J, Farr A L , Randall R J. J Biol Chem, 1951, 92: 265

[21] Habeeb A F S A. Anal Biochem, 1966, 14: 328

[22] Zofia Olempska-Beer. 61st JECFA-Chemical and Technical Assessment (CTA): Laccase from Myceliophthora Thermo-phila expressed in Aspergillus Oryzae. Geneva: FAO, 2004. 4

[23] Glazer A N, Smith E L. J Biol Chem, 1961, 236: 2942

[24] 张丽, 秦德志, 杨维春, 刘巧茹, 王莉. 分析测试学报 (Zhang L, Qin D Z, Yang W C, Liu Q R, Wang L. J Instru Analy), 2010, 29: 721

[25] Khaparde S S, Singhal R S. Biores Technol, 2001, 78: 1

[1] 闫朝阳, 兰丽, 陈山虎, 赵明, 龚茂初, 陈耀强*.高性能 $Ce_{0.5}Zr_{0.5}O_2$ 稀土储氧材料的制备及其负载的单 Pd 三效催化剂[J]. 催化学报, 2012,33(2): 336-341

[2] 宋明娟, 邹成龙, 牛国兴, 赵东元.$(NH_4)_2SiF_6$ 预处理改善 SBA-15 介孔材料的水热稳定性[J]. 催化学报, 2012,33(1): 140-151

[3] 李霞, 杨霞珍, 唐浩东, 刘化章*.载体对合成气制甲烷镍基催化剂性能的影响[J]. 催化学报, 2011,32(8): 1400-1404

[4] 华新雷 1,*, 王立刚 1, 徐勇华 1, 王尤崎 1, 高滋 2.费托反应本征动力学中合成气分压对反应速率的影响[J]. 催化学报, 2011,32(7): 1242-1249

[5] 金辰, 邱顺晨, 朱月香*, 谢有畅.具有优异热稳定性的磷修饰氧化钛及其对水中污染物的降解[J]. 催化学报, 2011,32(7): 1173-1179

[6] 姚艳玲, 方瑞梅, 史忠华, 龚茂初, 陈耀强.La_2O_3 对 Pd 密偶催化剂性能的影响[J]. 催化学报, 2011,32(4): 589-594

[7] 李巧灵 1, 张元华 1, 陈世萍 1, 方维平 1,2, 杨意泉 1,2.反映工艺条件对管式反应器催化反应影响的转化率方程[J]. 催化学报, 2011,32(3): 446-450

[8] 韩伟, 贾玉心, 熊国兴, 杨维慎.介孔-微孔复合材料的水热稳定性及其催化裂化性能[J]. 催化学报, 2011,32(3): 418-427

[9] 姚艳玲, 何胜楠, 史忠华, 龚茂初, 陈耀强.BaO 含量对 Ba 改性 Al_2O_3 及其负载的 Pt-Rh 密偶催化剂性能的影响[J]. 催化学报, 2011,32(3): 502-507

[10] 方向青, 王钰宁, 邓秀娟, 吴海虹, 吴鹏, 刘月明, 何鸣元.Ti-MWW 催化氯丙烯环氧化反应动力学行为[J]. 催化学报, 2011,32(2): 333-339

[11] 王德峥.Langmuir-Hinshelwood 动力学的有效实验条件[J]. 催化学报, 2010,26(8): 972-978

[12] 苏继新;张慎平;马丽媛;屈 文;张明博.Au/SBA-15 的制备及其催化 CO 氧化反应性能[J]. 催化学报, 2010,31(7): 839-845

[13] 刘俊龙;薛会福;黄秀敏;吴培豪;黄信炅;刘尚斌;申文杰 .预吸附吡啶增强二甲醚在丝光沸石上羰基化反应的稳定性[J]. 催化学报, 2010,31(7): 729-738

[14] 陈闪山;朱银华;李伟;刘维佳;李力成;杨祝红;刘畅;姚文俊;陆小华;冯新.含 TiO2(B) 介孔氧化钛材料的制备、特性和应用[J]. 催化学报, 2010,31(6): 605-614

[15] 马俊红;冯媛媛;张贵荣;王安杰;徐柏庆 .氧化钨对 Pt-RuOxHy 电催化剂甲醇氧化性能的促进作用[J]. 催化学报, 2010,31(5): 521-524