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论文

外源一氧化氮供体硝普钠对小麦种子萌发早期β-淀粉酶及其亚细胞分布的影响

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摘要:

采用药理学和生物化学实验技术, 研究了外源一氧化氮(nitric oxide, NO)供体硝普钠(sodium nitroprusside, SNP) 处理对小麦种子萌发早期β-淀粉酶及其亚细胞分布的影响。β-淀粉酶的专一性抑制剂g-环糊精可以明显抑制O.5 $mmol L^{-1}$ SNP诱导的小麦种子萌发, SNP还能诱导自由型β-淀粉酶酶活性, 上调β-淀粉酶基因转录本的水平。 Western blotting结果发现, SNP处理可以增加β-淀粉酶蛋白的表达; 运用NO专一性清除剂cPTIO及转录抑制剂放 线菌素D可以逆转上述转录本和蛋白水平的变化。免疫电镜亚细胞定位证实小麦胚内的β-淀粉酶主要定位于蛋白储 藏体内, 并可能附着在淀粉粒上, SNP处理增加了β-淀粉酶蛋白的分布。推测外源NO供体SNP在小麦种子萌发早期 不仅提高了自由型 β -淀粉酶的活性,还可能诱导了 β -淀粉酶的重新合成。

关键词: 一氧化氮 β-淀粉酶 转录调控 亚细胞定位 小麦种子

Effects of Exogenous Nitric Oxide Donor Sodium Nitroprusside on **B**-Amylase and Its Subcellular Localization during Early Stage of Seed Germination in Wheat

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Abstract:

Nitric oxide (NO) is an important signaling molecule with diverse physiological functions in plants. NO can break dormancy of seeds, and promote seeds germination under various stresses and normal growth conditions. In this report, treatment with 0.5 mmol ${\rm L}^{-1}$ sodium nitroprusside (SNP) promoted seeds germination by increasing β -amylase activity, while the application of α -cyclodextrin, a specific inhibitor of β -amylase, could apparently inhibit the process induced by SNP. Semi-quantitative RT-PCR analysis indicated that the application of SNP induced the free β -amylase activity, and up-regulated β -amylase transcript. West-ern-blotting result showed that SNP increased the protein level of β-amylase, whereas the cPTIO (a specific scavenger of NO) and actinomycin-D (a transcriptional inhibitor) reversed changes in transcriptional and the protein levels mentioned above. Subcellular localization studies of proteins via immunoelectron microscopy technique illustrated that SNP could increase the quantity of β-amylase which was predominantly located in protein storage vacuole, and adhered to starch granules. The above results suggest that SNP might not only enhance the activity of free β -amylase, but also induce the β amylase synthesis de novo at the early stage of wheat seed germination.

Keywords: Nitric oxide β-amylase Transcriptional regulation Subcellular localization Wheat seeds 收稿日期 2007-12-20 修回日期 1900-01-01 网络版发布日期 2008-09-12

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