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研究论文

小麦面粉Puroindoline蛋白的提取与纯化

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摘要 Puroindoline蛋白是小麦面粉中一种非常重要的蛋白质,不仅影响和决定了籽粒的硬度,而且有抗G+、G-菌以及抗真菌的作用.用含4% Triton X-114、 100 mmol/L pH7.8 Tris-HCl缓冲液处理小麦面粉来分离Puroindoline蛋白,经处理后得到的蛋白质混合溶液首先用分子筛葡聚糖G-75纯化,每个收集管内的组分经SDS-PAGE分析,分子量小于31 kD的蛋白质组分被回收和集中,回收的蛋白质组分经PEG20000浓缩后,再用离子交换柱羧甲基纤维素(CM-23)进行纯化.其洗脱液分别是双蒸水和NaCl,梯度为0.05～0.7 mol/L、8 mmol/L pH5.5的MES缓冲液,回收只含15 kD的蛋白质的组分,接着用PEG20000浓缩,最后冷冻干燥得到Puroindoline蛋白.

关键词 Puroindoline蛋白 面粉 提取与纯化 Triton X-114 SDS-PAGE

分类号 Q946.1

Extraction and Purification of Puroindoline from Wheat Flour

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Abstract Puroindoline, influenced and decided the wheat hardness and exhibiting activity against gram-positive bacteria, gram negative bacteria and fungi, is a very important protein in wheat flour. In this studies, the puroindoline was extracted from wheat flour in the 100 mmol/L Tris HCl pH7.8 buffer, containing 4% Triton X-114. Then the protein mixture including puroindoline was successively purified by Sephadex G-75, and each fraction was analyzed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis(SDS-PAGE). Fractions containing polypeptides with apparent molecular weights of less than 31 kD were pooled and loading on Carboxymethyl-cellulose(CM-23) after concentrated by PEG20000, and each fraction was analyzed as preceding. In those ways, they were eluted by distilled water and applying a gradient from 0.05 mol/L to 0.7 mol/L NaCl in 8 mmol/L MES pH5.5 buffer respectively. The fractions containing polypeptides apparent molecular weight 15 kD were pooled and concentrated by PEG20000, at last the puroindoline was obtained by freeze-dried.

Key words Puroindoline Flour Extraction and purification Triton X-114 SDS-PAGE

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