

Cloning and Nucleotide Sequence of the Acid Protease-encoding Gene (*pepA*) from *Aspergillus oryzae*

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We have cloned a genomic DNA sequence encoding the acid protease (PEPA) from *Aspergillus oryzae* using a 0.6-kb fragment as a probe. This fragment was amplified by the polymerase chain reaction (PCR) using oligonucleotide primers designed from the partial amino acid sequences of peptide fragments of the purified protein. Nucleotide sequencing analysis has shown that the cloned gene (designated *pepA*) encodes 404 amino acid residues and contains 3 putative introns ranging in length from 50 to 61 nucleotides. The deduced amino acid sequence of the *A. oryzae* PEPA has a high degree of homology (67%) to the *A. awamori* PEPA. Comparison with the amino acid sequence of *A. awamori* PEPA suggests that the *A. oryzae* PEPA may consist of a 78 amino acid prepro-peptide and 326 amino acid mature protein. The amino acid composition of the mature protein was almost consistent with that of the acid protease purified from *A. oryzae* reported previously. Southern hybridization analyses showed that the *pepA* gene exists as a single copy in the *A. oryzae* chromosome. The cloned gene was found to be functional, since transformants of *A. oryzae* containing multiple copies of the *pepA* gene showed a 2-6 fold increase in acid protease activity compared with the recipient strain.