Muscle endopin 1, a muscle intracellular serpin which strongly inhibits elastase: purification, characterization, cellular localization and tissue distribution

In the present work, an endopin-like elastase inhibitor was purified for the first time from bovine muscle. A three-step chromatography procedure was developed including successively SP-Sepharose, Q-Sepharose and EMD-DEAE 650. This procedure provides about 300 microg of highly pure inhibitor from 500 g of bovine diaphragm muscle. The N-terminal sequence of the muscle elastase inhibitor, together with the sequence of a trypsin-generated peptide, showed 100% similarity with the cDNA deduced sequence of chromaffin cell endopin 1. Hence, the muscle inhibitor was designated muscle endopin 1 (mEndopin 1). mEndopin 1 had a molecular mass of 70 kDa, as assessed by both gel filtration and SDS/PAGE. According to the association rates determined, mEndopin 1 is a potent inhibitor of elastase (k_{ass}=2.41\times10^{7} \text{ M}^{-1}\text{s}^{-1}) and trypsin (k_{ass}=3.92\times10^{6} \text{ M}^{-1}\text{s}^{-1}), whereas plasmin (k_{ass}=1.78\times10^{3} \text{ M}^{-1}\text{s}^{-1}) and chymotrypsin (k_{ass}=1.0\times10^{2} \text{ M}^{-1}\text{s}^{-1}) were only moderately inhibited. By contrast, no inhibition was detected against several other selected serine proteinases, as well as against cysteine proteinases of the papain family. The cellular location of mEndopin in muscle tissue and its tissue distribution were investigated using a highly specific rabbit antiserum. The results obtained demonstrate an intracellular location and a wide distribution in bovine tissues.

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