The kallikreinogenase activity of human mast cell tryptase.

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Tryptase is a tetrameric serine protease [1] which is present almost exclusively in the secretory granules of mast cells [2]. There has been some controversy over the extent to which tryptase may contribute to the generation of kinins which has been reported following mast cell activation [3,4]. Earlier studies suggested that tryptase does not generate bradykinin from either low molecular weight kallikreinogen (LMWK) [5] or high molecular weight kallikreinogen (HMWK) [6]. In contrast Proud et al. [7] in seeking to purify the kallikrein-like enzyme from mast cells found that tryptase and kallikreinogenase activities co-chromatographed, and concluded that tryptase may be the enzyme in mast cells responsible for kinin generation. However, tryptase-induced release of bradykinin from LMWK was optimal at pH 5.5, and the activity was relatively low at neutral pH casting doubt on its biological significance. We have re-examined this issue and have investigated the potential of tryptase to cleave the synthetic chromogenic substrates for kallikreins S-2266, S-2302 (both Kabi-Vitrum) and CH-848 (Ferring), and the natural substrates LMWK and HMWK.

Tryptase was purified from human lung tissue using high salt extraction, octyl agarose and heparin agarose column chromatography as described previously [8]. Antiserum specific for urinary kallikrein did not cross-react with the tryptase on Ouchterlony immunodiffusion. Peptide CH-848 was prepared by solution peptide synthesis [9] using a (7+2) fragment coupling strategy. Heptapeptide Ac-Arg-Pro-Pro-Leu-Met-Pro-OH previously synthesised by S.P.P.S. and dipeptide H-Phe-Arg. PNA.2HCl were coupled by the BOP activation method, and CH-848 purified by reversed phase MPLC. HMWK was purified from plasma by chromatography as described previously [8]. Antiserum against bradykinin was a generous gift from Dr L. Schwartz, Malmö General Hospital. Financial support from the National Asthma Campaign is gratefully acknowledged.


![Fig. 1. Cleavage of CH-848 (0.5 mM) by tryptase (0.5 µg/ml) over the pH range 3.0 to 10.0.](image-url)