

## Antibacterial Activity of Bovine Lactoferrin-Derived Peptides

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### Executive Summary

Several peptides sharing high sequence homology with lactoferricin B (Lf-cin B) were generated from bovine lactoferrin (Lf) with recombinant chymosin. Two peptides were copurified, one identical to Lf-cin B and another differing from Lf-cin B by the inclusion of a C-terminal alanine (lactoferricin). Two other peptides were copurified from chymosin-hydrolyzed Lf, one differing from Lf-cin B by the inclusion of C-terminal alanylleucine and the other being a heterodimer linked by a disulfide bond. These peptides were isolated in a single step from chymosin-hydrolyzed Lf by membrane ion-exchange chromatography and were purified by reverse-phase high-pressure liquid chromatography (HPLC). They were characterized by N-terminal Edman sequencing, mass spectrometry, and antibacterial activity determination. Pure lactoferricin, prepared from pepsinhydrolyzed Lf, was purified by standard chromatography techniques. This peptide was analyzed against a number of gram-positive and gram-negative bacteria before and after reduction of its disulfide bond or cleavage after its single methionine residue and was found to inhibit the growth of all the test bacteria at a concentration of 8 mM or less. Subfragments of lactoferricin were isolated from reduced and cleaved peptide by reverse-phase HPLC. Subfragment 1 (residues 1 to 10) was active against most of the test microorganisms at concentrations of 10 to 50 mM. Subfragment 2 (residues 11 to 26) was active against only a few microorganisms at concentrations up to 100 mM. These antibacterial studies indicate that the activity of lactoferricin is mainly, but not wholly, due to its N-terminal region.