Antimicrobial peptides

A family of wound healers

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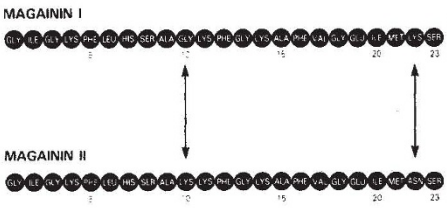
OOCYTES of the toad *Xenopus laevis* are an excellent system for studying RNA expression. Two recent papers by Zasloff and his colleagues[1,2] indicate that the surgical techniques involved in manipulating these eggs might fortuitously provide important information on how this animal has the remarkable ability to remain free of infection during wound healing with little or no post-operative care. A family of peptides (the magainins) isolated from the ventral skin of the toad has antimicrobial activity. Zasloff and colleagues have determined the amino-acid sequences of these compounds (see figure),



and their studies on some related synthetic peptides indicate certain structural requirements for biological activity. The authors suggest that the natural products provide a host-defence system to combat infection by microorganisms.

The magainins are the latest of a large number of biologically active peptides found in various organisms. The physiological role(s) of such compounds has not always been apparent, but there are promising exceptions to this rule. The peptide melittin, for example, is the main toxic component of honeybee venom and its structure has been studied in detail[3]. The monomeric melittin chain has 26 amino-acid residues (compared with the 23 residues of the magainins) and the peptide can form a highly amphiphilic helix. This allows it to integrate into membranes and its highly efficient lytic action could well stem from this limited penetration into the lipid bilayer. Indeed, a peptide containing 20–25 amino-acid residues is just about the right length for a putative transmembrane alpha-helix. The magainins too have the potential to form amphiphilic helices. But although they perturb protozoan membrane functions that are associated with osmotic balance, they do not cause haemolysis of human erythrocytes.

In contrast to the venomous associations of melittin, the cecropins have been hailed as constituents of a major antibacterial defence system in insects. Whereas melittin lyses bacteria and eukaryotic cells, cecropins are specific for the former. Insects lack lymphocytes and immunoglobulins and therefore their immune systems must differ from those of vertebrates. Injection of live non-pathogenic bacteria into pupae of *Hyalophora cecropia* induces a potent antibacterial activity involving cecropins A, B and D, the attacins and lysozyme[4]. Comparisons between the action of melittin, cecropins and magainins raise interesting questions about the structural requirements for the selective lytic activity of small membrane-active peptides.

Other peptides, including xenopsin and caerulein, have already been isolated from amphibian skin[5]. Xenopsin contains eight amino acids and its sequence closely resembles that of mammalian neurotensin, with which it shares certain biological properties[6]. Caerulein is a decapeptide with the same carboxy-terminal sequence as the mammalian hormones cholecystokinin and gastrin[7]. Again, certain biological properties are shared[7]. These skin peptides have obscure physiological roles although they too can potentially form amphiphilic helices, so a membrane-active or lytic function cannot be excluded. Clearly, however, magainins are distinct structurally from xenopsin and caerulein. Indeed, Zasloff and colleagues[1,2] claim that magainins have no similarity in amino-acid sequence to any catalogued protein in the Genebank database. Based on available evidence, therefore, the magainins are truly novel peptides.

Many, if not all, of those biologically active peptides that have been isolated are synthesized as precursors and the magainins are no exception. A complementary DNA clone has been isolated that encodes a 160-amino-acid portion of a precursor protein that includes in its sequence one copy of magainin I and two copies of magainin II. Each peptide has a common leader and a common trailer sequence of 6 and 7 amino acids, respectively. The two intervening sequences linking the three peptides and preceding each of the magainin II species are identical, apart from the substitution of glutamine for glutamic acid in one position. Similar, but less exact, internal homology has been reported for amino-acid sequences within the caerulein precursor as also purified from *X.laevis* skin[7]. What is particularly interesting for the magainin precursor is that both of the intervening peptides and the three biologically active magainins are each 23 amino acids long. This structure could have implications for processing. Liberation of xenopsin and caerulein seems likely to involve several processing steps[7] and a dipeptidylaminopeptidase activity,

Wounds on the abdomen remain free of infection.

already detected in the skin secretions of *X.laevis*, may well be involved at some stage. In contrast, the magainins could be liberated by a single cleavage event occurring next to the amino-terminal glycine residues of the active peptides and possibly by a single (tissue-specific?) cleavage event that liberates the carboxy terminus. The involvement of a dipeptidylaminopeptidase may be unnecessary. Such a processing mechanism would additionally liberate the intervening sequences as two intact, and almost identical, peptides each containing 23 amino acids. Could such peptides themselves have biological activity? They do not appear to be amphiphilic but neither do the defensins, biologically active antibiotic peptides that could function in mammalian phagocytic vacuoles[8].

It is an attractive idea that amphibians, which for centuries have featured prominently in witchcraft, could indeed possess a 'magical' ability to heal wounds (see photograph). Whether or not protective peptides similar to the magainins will be found in mammals and linked to such disorders as cystic fibrosis (discussed by Zasloff and colleagues[1,2]) is highly speculative. Biologically active peptides are fascinating compounds, but there are a lot of them around and precise physiological roles are hard to pin down. But as the magainins seem to be released within the skin and may be present in peritoneal and subdermal fluids, they are at least in a relevant location for fulfilling their putative role in wound healing. They could yet be of considerable clinical significance. □

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