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Commentary

# An Overview of Microbial Heparinases

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## DESCRIPTION

Heparinases (heparin lyases) are polysaccharide lyases that depolymerise glycosaminoglycans by eliminative cleavage. The catalytic system of polysaccharide lyases have three steps: Neutralisation of the negative charge on the C6 carboxylate anion, abstraction of the proton from C5 of uronic acid by a common base catalyst and proton sharing to the glycoside bond under cleavage by a general acid catalyst. Due to the contribution of the uronic acid carboxylic group, lyases only act on linkages on the non-reducing side of uronic acids. Polysaccharide lyases, which are extensive in nature and involved in diverse biochemical procedures, are one of five enzyme classes covered by the Carbohydrate-Active enzymes Database (CAZy) which groups enzymes into amino acid sequence-based families linking sequence to enzyme specificity and 3D structure. Polysaccharide lyases are presently divided into 40 families with heparinases found in PL families 8, 12, 13, 15, 21 and 37.

Heparinases act specifically on the glycosidic linkage joining the hexosamines and uronic acids exist in heparin and heparan sulphate *via* a β-elimination mechanism producing disaccharide/ oligosaccharide products with a C4–C5 double bond in the non-reducing end uronic acid moiety. Heparinases is divided into three groups based on substrate specificity: heparinase I, heparinase II and heparinase III, also mentioned as heparin lyase I, II and III, respectively.

Heparinase I acts principally on highly sulfated heparin while heparinase III specially cleaves the less sulfated heparan sulfate and heparinase II shows wider substrate specificity degrading sections of both high and low sulfation in heparan sulphate and heparin. First Heparinase activity was isolated from the bacteria Pedobacter heparinus and much of what is assumed about heparinase substrate specificity and defined.

Heparinase I cuts greatly sulfated sections and acts mainly at the glycosidic linkage joining N-sulfated glucosamine (GlcNS6S±3S) and 2-O-sulfated iduronic acid, that accounts for 75% to 95% of disaccharides in heparin. It has been stated that the linkage between GlcNS3S6S and IdoA2S which is found in the antithrombin III binding site is more liable to cleavage by heparinase I than the main heparin disaccharide unit of GlcNS6S linked to IdoA2S. While 2-O-sulfation of the IdoA residue or 6-O-sulfation of the GlcNS residue are not severely needed and cleavage of oligosaccharides lacking either sulfate group has been observed using augmented concentrations of heparinase I, the increased sulfation is better with the main catalytic effectiveness observed on the primary site, GlcNS6S-IdoA2S. Heparinase I display an importance for hexosamine-iduronic acid linkages over those containing glucuronic acid residues although increasing catalytic competence has been reported with increasing sulfation on the glucuronic acid residue and adjacent glucosamine residues. Hep I does not cleave at N-unsubstituted glucosamine (GlcNH3+) residues which are placed in heparan sulfate and have been connected with key cell events, even with 2-O- and/or 6-O-sulfation of the oligosaccharides.

It is notable that heparin/heparan sulfate may also be depolymerised by the mammalian enzyme heparanase, an endo-β-glucuronidase, which acts at the inside glycosidic bond linking glucuronic acid (GlcA) and N-sulfated glucosamine (GlcNS) through a hydrolytic mechanism with a speciality influenced by substrate size and sulfation pattern. Heparanase is involved in numerous pathological procedures including metastasis, tumour growth, angiogenesis and inflammation and as well as being vital for cancer advancement and metastasis, heparanase has also been associated with other diseases (diabetes, liver fibrosis, amyloidosis, bone necrosis, and Alzheimers disease) and is a target in the improvement of anti-tumour and anti-inflammatory therapies. The enzyme is also involved in normal physiological processes and occurs in a range of normal and malignant cells and tissues.

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