“Bioinformatic sequence analysis to determine relationships within the fetuin-proteinfamily”

Bachelor’s thesis

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Fetuins are essential in various developmental processes, for example, in fertilization, embryo development, aging and apoptosis. Those members of the cystatin-superfamily play an important role in diseases such as Alzheimer and in the immune defense to cancer.

The differentiation of fetuins is of two types: fetuin-A, also known as alpha-2-Heremans-Schmid glycoprotein (AHSG), and fetuin-B. These plasma-proteins have two cystatin-like domains with a connecting linker in common. However, both types of fetuin differ strongly in their function: Whereas fetuin-B can inhibit certain astacins, which are members of the metalloprotease family, fetuin-A is not capable of this process. Negative charged amino acid side chains on the surface of fetuin-A have a function in mineralization of bone tissue and therefore prevent precipitation of calcium phosphate. The first part of this thesis reviews theoretical foundations regarding astacins, both types of fetuins, and their interaction with each other.

The aim of this research has been to obtain a comprehensive insight into relationships and evolution of fetuins. In a practical part, numerous sequences of fetuins were analyzed regarding their functional and evolutionary similarities and differences, and cross-examined between both fetuin-A and fetuin-B, as well as between fetuins of different species. Relevant sequences of amino acids were searched for in databases and analyzed by performing *multiple sequence alignment* and by creating phylogenetic trees. An analytical focus laid on certain regions that are essential for inhibiting astacins and for regulating mineralized matrix. Some models of fetuin-A and another member of the cystatin-superfamily, kininogen-1-like protein of the snake *Python bivittatus*, resulted from the previously created alignments from computer modeling for the purpose of analyzing and comparing predicted three-dimensional structures. The negatively charged amino acid patterns within the fetuin-A sequence were not yet highly conserved in species that only consist of cartilage instead of bones. We found that in zebrafish multiple genome duplications took place, which sequences were categorized into five groups. Furthermore, we found that antivenin proteins of snakes, which have the ability to inhibit metalloproteinases in the form of snake venom, are fetuin-A-like.

The results of this thesis support the assumption that fetuin-A and fetuin-B differentiated increasingly with the development of Osteichthyes. Furthermore, the hypothesis of fetuins emerging at the basis of vertebrates, is supported. Our findings suggest that fetuin-A emerged presumably earlier in evolution.