with DNA in sperm nuclei at the end of spermiogenesis. Protamine-like proteins are one of the three types of Sperm Nuclear Basic Proteins, and represent a structurally and functionally intermediate group of proteins between the histone and protamine type. Protamine-like proteins represent the major acid-soluble protein components of the mussel *Mytilus galloprovincialis* sperm chromatin and consist of the protamine-like proteins PL-II, PL-III and PL-IV. The aim of this study was to investigate the antibacterial activity of these proteins since, to date, there are reports on bactericidal activity of protamines and histones, but not on protamine like proteins. We tested the bactericidal activity of these proteins against Gram-negative bacteria: *Proteus mirabilis*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Salmonella typhmurium*, *Enterobacter aerogenes*, *Enterobacter cloacae*, and *Escherichia coli* as well as on Gram-positive bacteria: *Enterococcus faecalis* and two different strains of *Staphylococcus aureus*. The results show that *Mytilus galloprovincialis* protamine-like proteins exhibited bactericidal activity against all bacterial strains tested with different minimum bactericidal concentration values, ranging from 15.7 to 250 µg/mL and also on the clinical isolates of the same bacterial species. Interestingly, these proteins were active against some bacterial strains tested that are resistant to conventional antibiotics. For their possible therapeutic use, we investigated the tossicity of these proteins. We found that these proteins showed very low toxicity as judged by red blood cell lysis and viability MTT assays and seem to act both at the membrane level and within the bacterial cell. Antibacterial proteins have a potential as alternative treatments to standard antibiotic therapies but oral administration would most likely result in the proteins being degraded in the digestive system. In order to analyze this aspect we generated an in vitro model of gastrointestinal digestion of PL-proteins and tested the bactericidal activity of the product obtained on a Gram-positive and a Gram-negative strain. We obtained the same results with respect to undigested protamine-like proteins on the Gram-positive bacterium. In conclusion, this work presents the first evidence obtained for *Mytilus galloprovincialis* of bactericidal activity of protamine-like-proteins.

Hemolymph extraction sites and 3d-visualization of the cardiovascular system and related structures of the blue mussel (*Mytilus edulis*)

M Eggermont[1], P Cornillie[2], N Nevejan[1], P Bossier[1], M Dierick[3], D Adriaens[4], P Sorgeloos[1], T Defoirdt[5], AM Declercq[1§]

[1]Laboratory of Aquaculture and Artemia Reference Center, Faculty of Bioscience Engineering, Ghent University, Ghent, Belgium

[2]Department of Morphology, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium

[3]Department of Physics and astronomy, Ghent University, X-Ray Engineering bvba Zwijnaarde, Belgium

[4]Research Group Evolutionary Morphology of Vertebrates, Ghent University, Ghent, Belgium

[5]Center for Microbial Ecology and Technology, Faculty of Bioscience Engineering, Ghent University, Ghent, Belgium

Bivalve hemolymph is used in a broad range of research domains such as eco(toxico)logy and immunology. However, the lack of a detailed description of hemolymph withdrawal protocols and locations (adductor muscles and heart) raises questions regarding the exact origin of the aspirated hemolymph and doesn't exclude the possibility of contamination with other body fluids which may have led to biased conclusions. A good description of the species-specific anatomy is lacking for many bivalves but is essential for a correct hemolymph withdrawal.

In this study we visualized and discussed the cardiovascular anatomy of the blue mussel (*Mytilus edulis*) and generated three-dimensional (3D) reconstructions based on micro-CT and histological images. Other structures, such as the gastro-intestinal system, the muscular system and body cavities, were included as well because of their close relationship to the cardiovascular system.

Hemolymph withdrawn from the posterior adductor muscle originates from small spaces and fissures between the muscle fibers that are connected to at least one hemolymph supplying artery, more specifically the left posterior gastro-intestinal artery. Hemolymph withdrawal from the heart is less straightforward. It is possible to puncture the pericard, anterior aorta and ventricle to collect a limited volume of hemolymph, however caution should be taken for contamination from the pallial cavity. Drainage of the pallial fluid prior to hemolymph extraction is therefore essential.

The different hemolymph extraction sites were clearly visualized in 3D. This study resulted simultaneously in a detailed description and visualization of the anatomy of *Mytilus edulis* useful to many research areas. Furthermore the described protocols and techniques to visualize the anatomy in 3D can easily be reproduced and adapted to other bivalve species.

More with mussels

AC Smaal[1,2]

[1]Aquaculture and Fisheries Group, Wageningen University and Research, Wageningen, The Netherlands

[2]HZ University of Applied Sciences, Aquaculture Team, Vlissingen, The Netherlands

Marine bivalves like mussels have been cultivated for ages and are recognised as a sustainable low food chain resource that acquires feed from natural resources in their environment. They provide a rich source for human nutrition and an associated economic value for local communities. Total mussel aquaculture production amounted 1.9 million tons in 2015 with a landing value of 3.2 billion US $.

Besides human nutrition, the mussels provide food for birds and benthos, a habitat for a large number of species, they regulate water quality and sequester carbon and nitrogen. Mussels are used to