Summary

In order to fertilize the egg, sperm cells orient in a chemical gradient of factors that are secreted by the egg. This process is called chemotaxis. The chemotactic factor of sperm cells of the sea urchin *Arbacia punctulata* is the peptide resact. Resact binds to a receptor-guanylyl cyclase (GC) which synthesizes cGMP from GTP. The increase in the cGMP-concentration leads to the opening of Ca^{2+}-channels - either directly or indirectly. The present study includes the biochemical and functional characterization of Arbacia-GC as well as first experiments concerning the adaptation of sperm cells.

The GC density in the flagellum of *A.punctulata* sperm was calculated to be ~ 400 GC molecules/μm^2. With two different experimental approaches, I found a 12 - 24-fold higher GC density. Densitometric analysis of Coomassie-stained protein gels and resact binding assays gave values of ~ 4.700 and ~ 11.000 GC-molecules/μm^2, respectively. In Cross-link experiments I could identify the resact-receptor as a trimer formed from GC-subunits. The binding assay revealed that the receptor down-regulates its binding affinity by negative cooperativity. The receptor senses resact-concentrations varying by four orders of magnitude. By means of GC-density, resact affinity, and cGMP-measurements in quench-flow experiments I determined a rate of cGMP synthesis of 4 - 5 cGMP/GC* s^{-1}. The enzymatic activity of the Arbacia-GC is fairly similar to that of GC-E from vertebrate photoreceptor cells. The synthesis of 4 to 5 cGMP/GC s^{-1} is sufficient to evoke a Ca^{2+}-signal upon binding of a single resact-molecule.

After a signaling pathways has been activated it also has to be turned off. This can be achieved at different levels. My experiments show that the resact-receptor is inactivated by dephosphorylation with a half-life of ~ 200 ms. Pioneering experiments concerning the adaptation of sperm cells showed unexpected and interesting results: Sperm cells that have been preincubated with a background concentration of resact (0,005 to 5 nM) showed a larger Ca^{2+}-signal in response to a given resact concentration (testpuls) than sperm cells that have not been preincubated with resact: So interestingly, sperm cells show an increase in sensitivity over a wide range of resact-background concentrations. Sensitization of sperm cells increases with the concentration of the testpuls. The Ca^{2+}-signal becomes smaller – sperm cells desensitize - when testpulses are given on a high resact-background concentration.