Reproductive barriers in the *Seminavis robusta* species complex and their involvement in species diversification

De Decker Sam, Pieter Vanormelingen, Koen Sabbe, and Wim Vyverman

Laboratory for Protistology and Aquatic Ecology, Biology Department, Ghent University
Krijgslaan 281-S8, 9000 Gent, Belgium
E-mail: Sam.DeDecker@UGent.be

With an estimated 200 000 species, diatoms are the most species-rich group of microbial eukaryotes (Vanormelingen *et al.*, 2008). They are responsible for around 20% of the net primary production on earth and play pivotal roles in global carbon and silica cycles (Nelson *et al.*, 1995; Smetacek, 1999). Despite this enormous diversity and ecological importance, the understanding of diatom speciation is largely uncharted territory. To date, several studies showed a large cryptic species diversity in microalgae, with a large variety of reproductive systems in some species complexes (Chepurnov & Mann, 1997; Sabbe *et al.*, 2004). Since sexual reproduction is an obligate stage in the life cycle of most diatoms (Chepurnov *et al.*, 2004), their evolutionary success may be related to this widespread variation in reproductive systems and to their highly sophisticated signalling systems during mating (Gillard *et al.*, 2013; Sato *et al.*, 2011). In this study, we collected a set of *Seminavis robusta* strains from the Veerse Meer and the Grevelingenmeer (the Netherlands) and the spuikom (Belgium). Genetic analysis showed that these strains form 3 cryptic species that are distinct but closely related. Sexual reproduction can be induced with high efficiency in intra-group crosses, while inter-group mating success is severely reduced. This poses an ideal scenario in which we can dissect the contribution of different possible reproductive barriers (e.i. mechanisms preventing gene flow) between emerging diatom species. For the identification of the most important reproductive barriers between the 3 cryptic *S. robusta* species, we will distinguish pre-zygotic barriers and post-zygotic barriers. The former include lack of recognition between sexual partners, while the latter include inviability or sterility of hybrid progeny. Both types of barriers prevent gene flow and thereby induce species diversification. Scoring of these reproductive barriers will be achieved by performing intra- and inter-group crosses in highly standardized laboratory conditions and assessing the contribution of different phases of the sexual process as a reproductive barrier. The role of the initial sexual signalling system as a barrier to gene flow will be unravelled using separate bioassays that are being developed at present. After identification of the most important barriers, genetic association studies (QTL mapping, GWAS) will be applied to identify the genomic regions underlying these barriers. This will result in candidate genes involved in speciation by sexual isolation. These results will contribute to testing the hypothesis the rapid evolution of reproductive isolation mechanisms contributes to the rapid diversification of diatoms. Furthermore, identification of candidate genes will pave the road for follow-up comparative and functional studies that will give us insight in the genetic players involved in the sexual process in *S. robusta*. This knowledge will be of major importance, since *S. robusta* is strongly emerging as a model species to study the life cycle regulation in diatoms (Chepurnov et al., 2008; Gillard *et al.*, 2008; Gillard *et al.*, 2013).

References

Chepurnov V.A. and D.G. Mann. 1997. Variation in the sexual behaviour of natural clones of *Achnanthes longipes* (Bacillariophyta). European Journal of Phycology 32(2):147-154.

Chepurnov V.A. *et al.* 2004. Experimental studies on sexual reproduction in diatoms. International Review of Cytology - a Survey of Cell Biology 237:91.

Chepurnov V.A. *et al.* 2008. In search of new tractable diatoms for experimental biology. Bioessays 30(7):692-702.

Gillard J. *et al.* 2013. Metabolomics enables the structure elucidation of a diatom sex pheromone. Angewandte Chemie-International Edition 52(3):854-857.

Gillard J. *et al.* 2008. Physiological and transcriptomic evidence for a close coupling between chloroplast ontogeny and cell cycle progression in the pennate diatom *Seminavis robusta*. Plant Physiology 148(3):1394-1411.

Nelson D.M. *et al.* 1995. Production and dissolution of biogenic silica in the ocean - Revised global estimates, comparison with regional data and relationship to biogenic sedimentation. Global Biogeochemical Cycles 9(3):359-372.

Sabbe K. *et al.* 2004. Apomixis in Achnanthes (Bacillariophyceae); development of a model system for diatom reproductive biology. European Journal of Phycology 39(3):327-341.

Sato S. *et al.* 2011. Novel sex cells and evidence for sex pheromones in diatoms. Plos One 6(10).

Smetacek V. 1999. Diatoms and the ocean carbon cycle. Protist 150(1):25-32.

Vanormelingen P., E. Verleyen, and W. Vyverman. 2008. The diversity and distribution of diatoms: from cosmopolitanism to narrow endemism. Biodiversity and Conservation 17(2):393-405.