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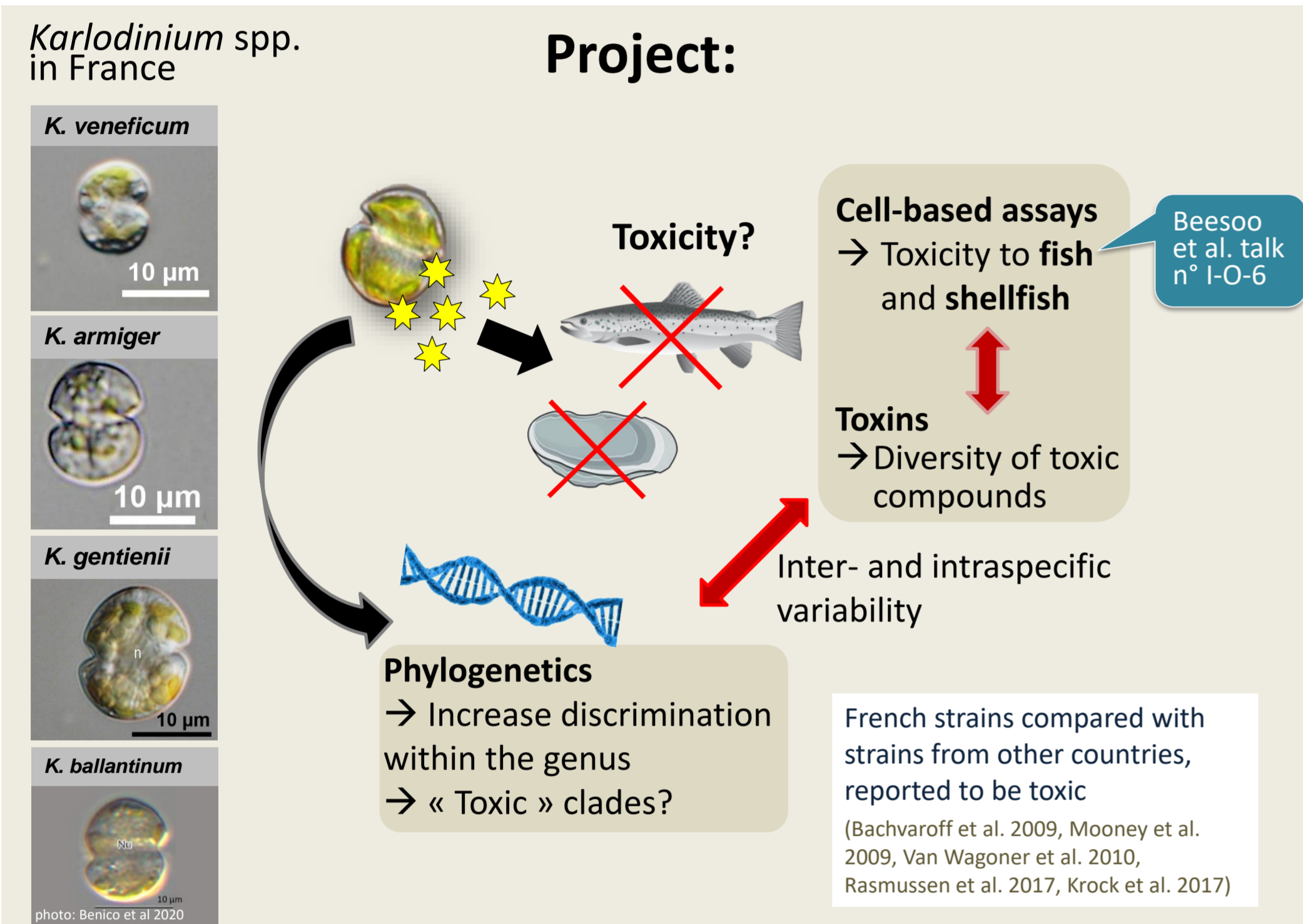
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Poster / speed talk  
n° I-ST-1

## Potentially ichthyotoxic *Karlodinium* spp. in France

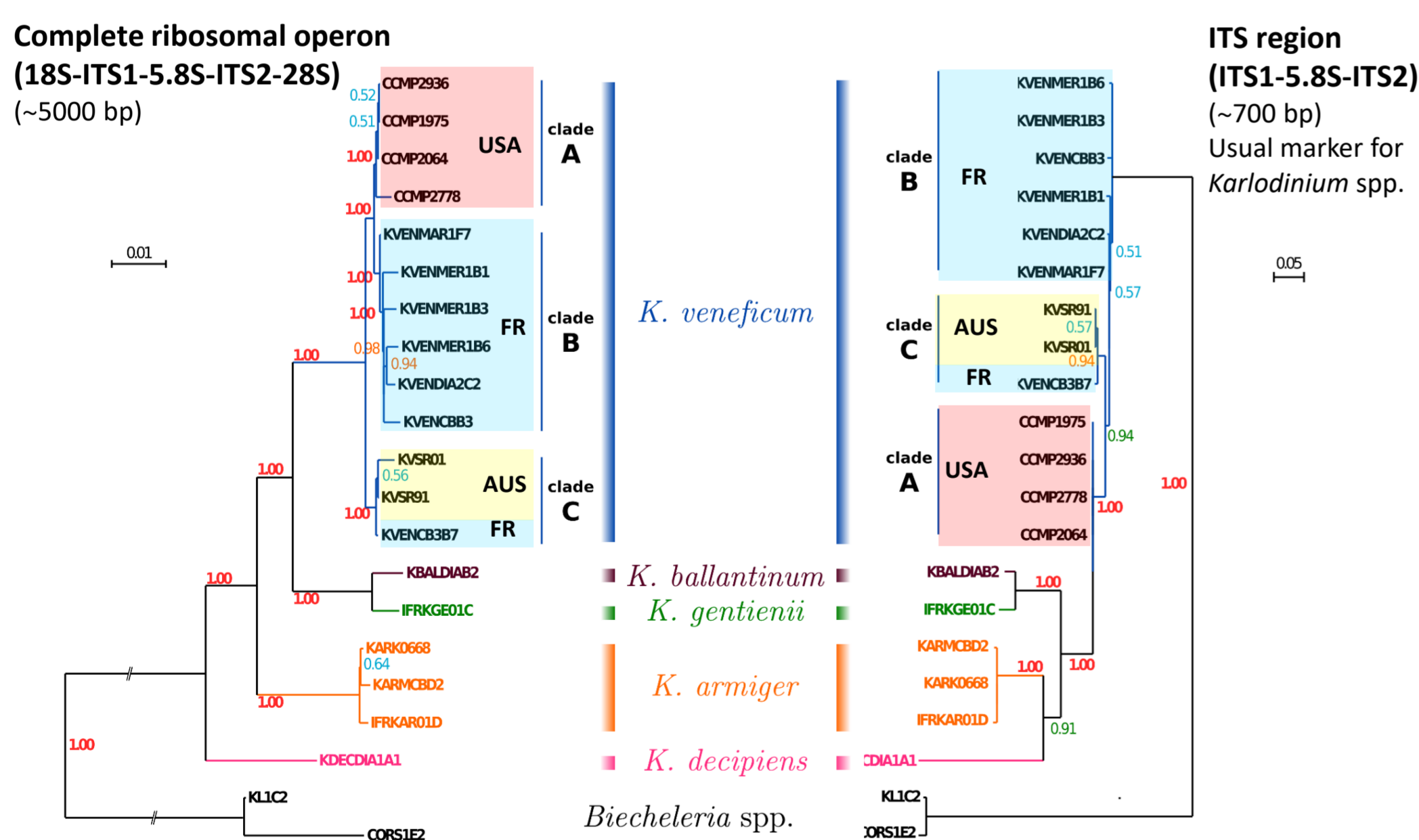
- Ichthyotoxic microalgae produce compounds that lead to fauna mortality, with ecological and economical impacts, particularly in the aquaculture industry (e.g. Hallegraeff et al. 2017).
- *Karlodinium* spp. blooms have induced fish and shellfish kills in several regions of the world. But toxicity is variable among species and strains (Bachvaroff et al. 2009, Krock et al. 2017).
- In France: presence of *Karlodinium* species (Nézan et al. 2014), but toxicity of French strains is unknown.

→ Are French *Karlodinium* spp. at ichthyotoxic risk?



## Phylogenetics

- Sequencing: whole ribosomal operon of rDNA (MinION Nanopore: long sequences but errors, Mi-Seq Illumina: low error rate but fragmentation).
- Bioinformatic analysis and phylogenetic trees (Bayesian method)



→ Better inter- & intraspecific discrimination using whole ribosomal operon vs ITS region

- Interspecific variability: essentially in ITS 1 & 2, but also in the rest
- Intraspecific variability: all along the operon (24 sites discriminative of a clade)

→ *K. veneficum*: 3 clades according to geographical origin

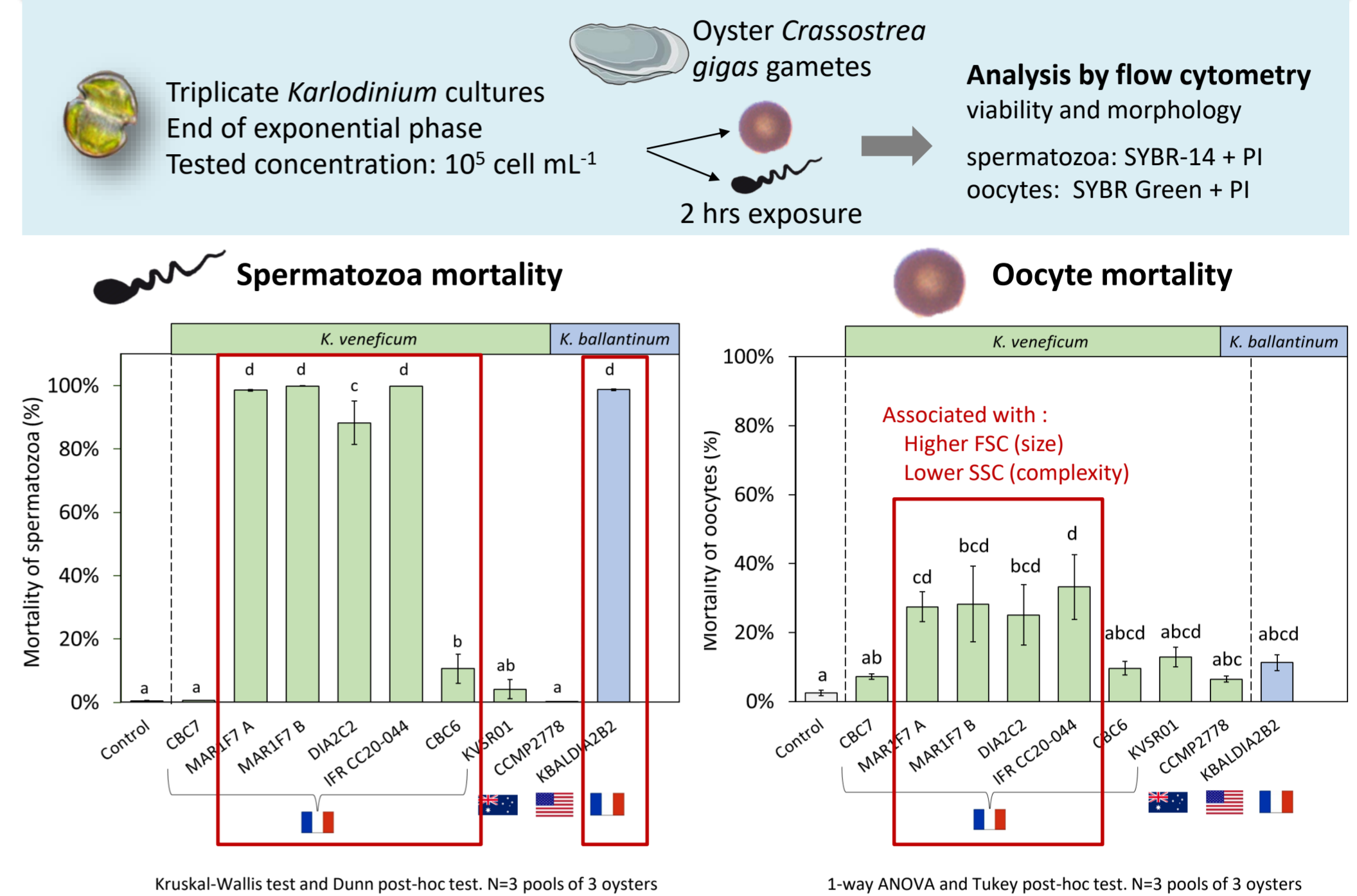
## Conclusions and perspectives

- Better phylogenetic discrimination within *Karlodinium* genus using whole ribosomal operon compared with usual ITS and LSU regions
- Different phylogenetic clades within *K. veneficum*, according to geographical origin and unrelated to toxicity
- French strains of *Karlodinium* spp. can affect bivalve cells as much as other foreign strains previously reported to be toxic
- Risk for bivalve reproduction needs to be evaluated at bloom realistic concentrations
- Toxicity of *K. ballantinum* and *K. veneficum* due to different mechanisms? → **Toxin analyses to complete and micropredation to study**

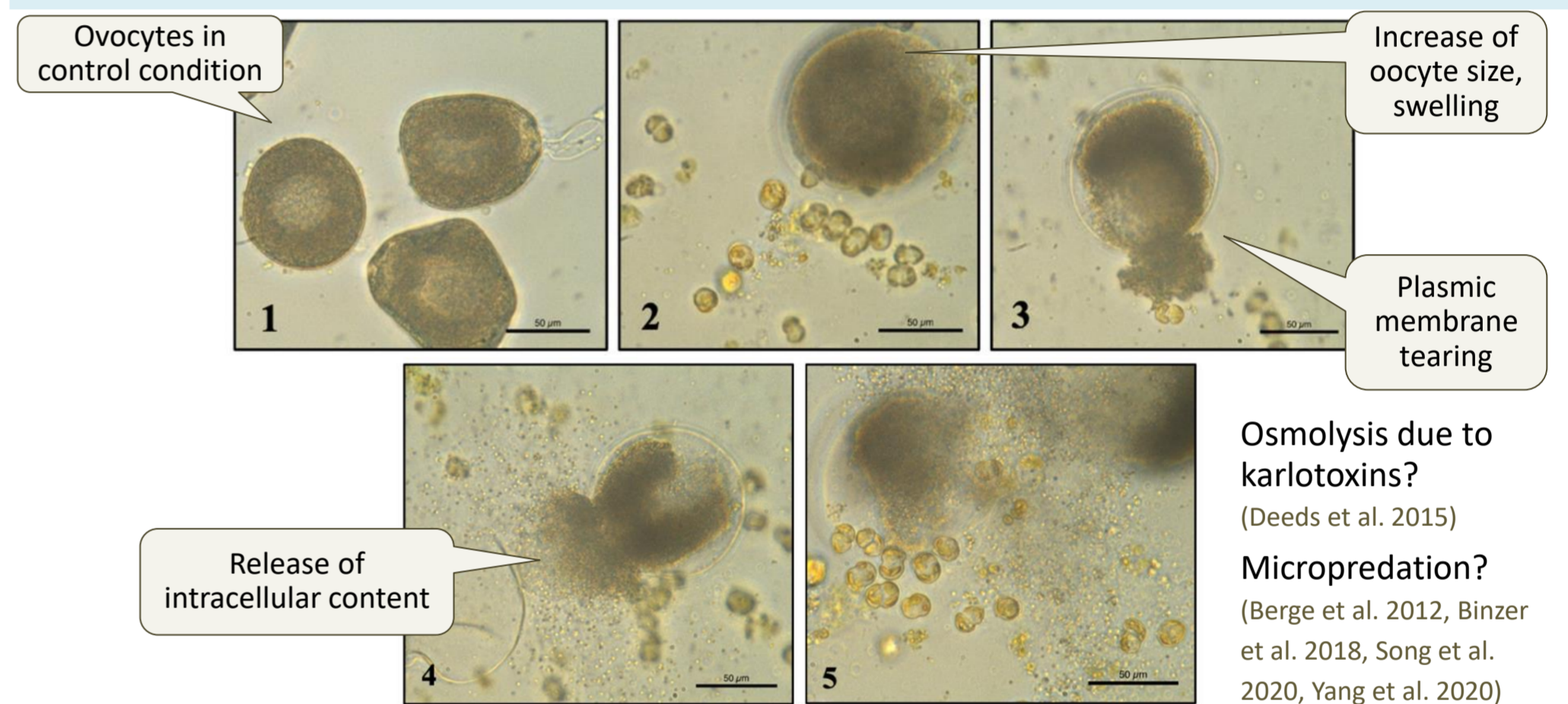
**References:** Adolf et al 2009, J. Phycol. 45, 176–192; Bachvaroff et al 2009, J. Phycol. 45, 137–153; Berge et al 2012, ISME J. 6, 1926–1936; Binzer et al 2018, PLoS One 13, e0199306; Deeds et al 2015, Aquat. Toxicol. 159, 148–155; Fu et al 2010, Aquat. Microb. Ecol. 59, 55–65; Hallegraeff et al 2017, Proceedings of the 17th ICHA, pp. 148–153; Krock et al 2017, Mar. Drugs 15, 391; Mooney et al 2009, J. Phycol. 45, 164–175; Nézan et al 2014, Harmful Algae 40, 75–91; Peng et al 2010, J. Am. Chem. Soc. 132, 3277–3279; Song et al 2020, Harmful Algae 99, 101926.; Yang et al 2020, Harmful Algae 93, 101177.

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## Toxicity to bivalve cells



**Light microscope:** oocytes exposed to Control or to French *K. veneficum* MAR1F7 strain



→ Identification of cytotoxic French strains (more toxic than foreign strains)

→ High intraspecific variability

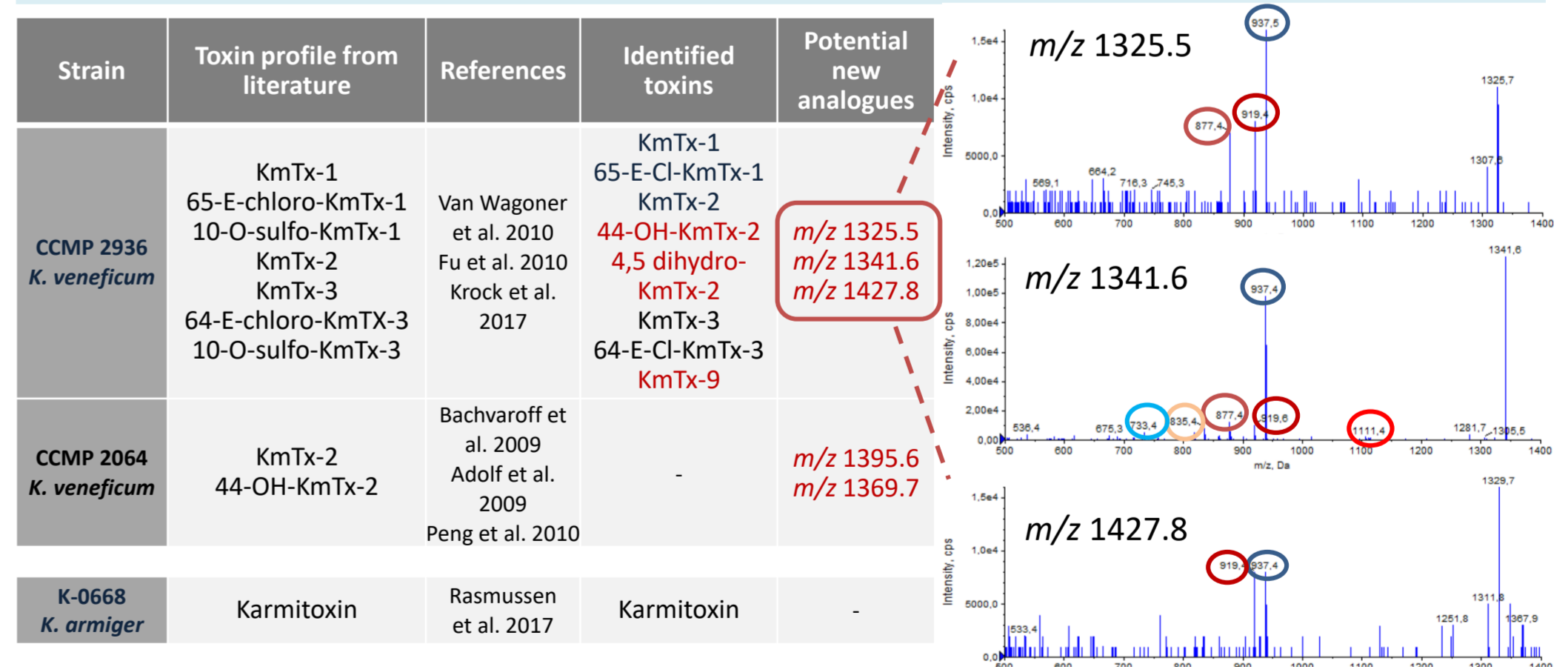
→ Role of karlotoxins vs micropredation?

→ First time *K. ballantinum* is reported to be toxic (similar results toward hemocytes – not presented)

→ Potential effects on oyster reproduction: to be assessed at bloom realistic concentrations of *Karlodinium* spp. ( $10^4$ – $10^2$  cell mL<sup>-1</sup>)

## Toxin analyses

- *Karlodinium* spp. cultures, end of exponential phase
- Methanolic extractions and analyses by LC-MS/MS (EPI and MRM modes)



→ First analyses of USA and Spanish strains suggest **putative new analogues and variation of karlotoxin composition** compared to profiles previously described in the literature

→ To be continued

## Fundings



Let It Bi, Biotox, Biotox-cell, Mecatox projects