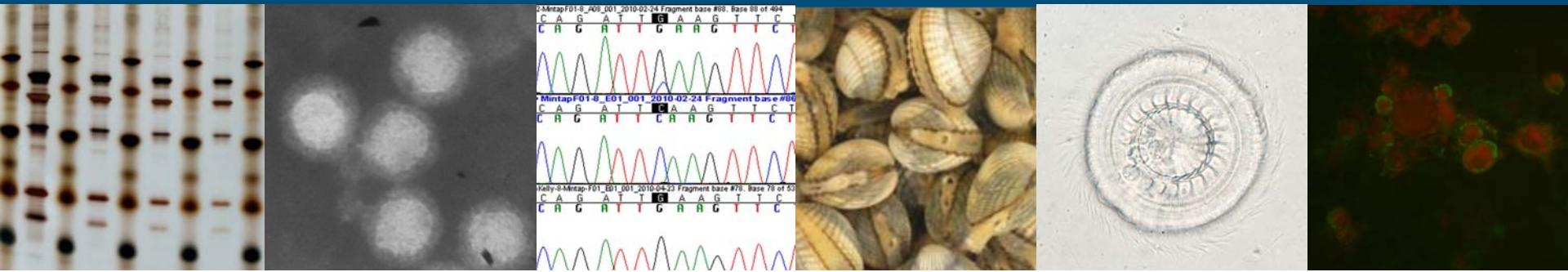


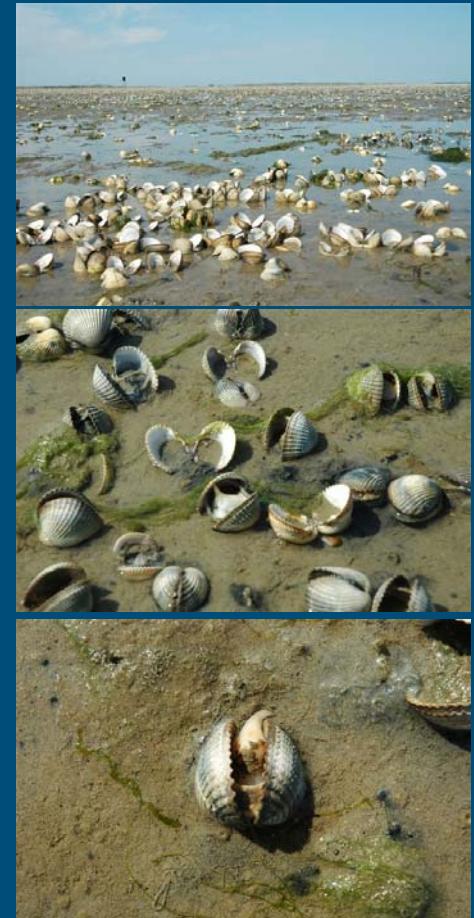
# Summer mortality of cockles *Cerastoderma edule* associated with digenean trematodes in the Oosterschelde, the Netherlands

Marc Engelsma, Ineke Roozenburg, Michal Voorbergen-Laarman



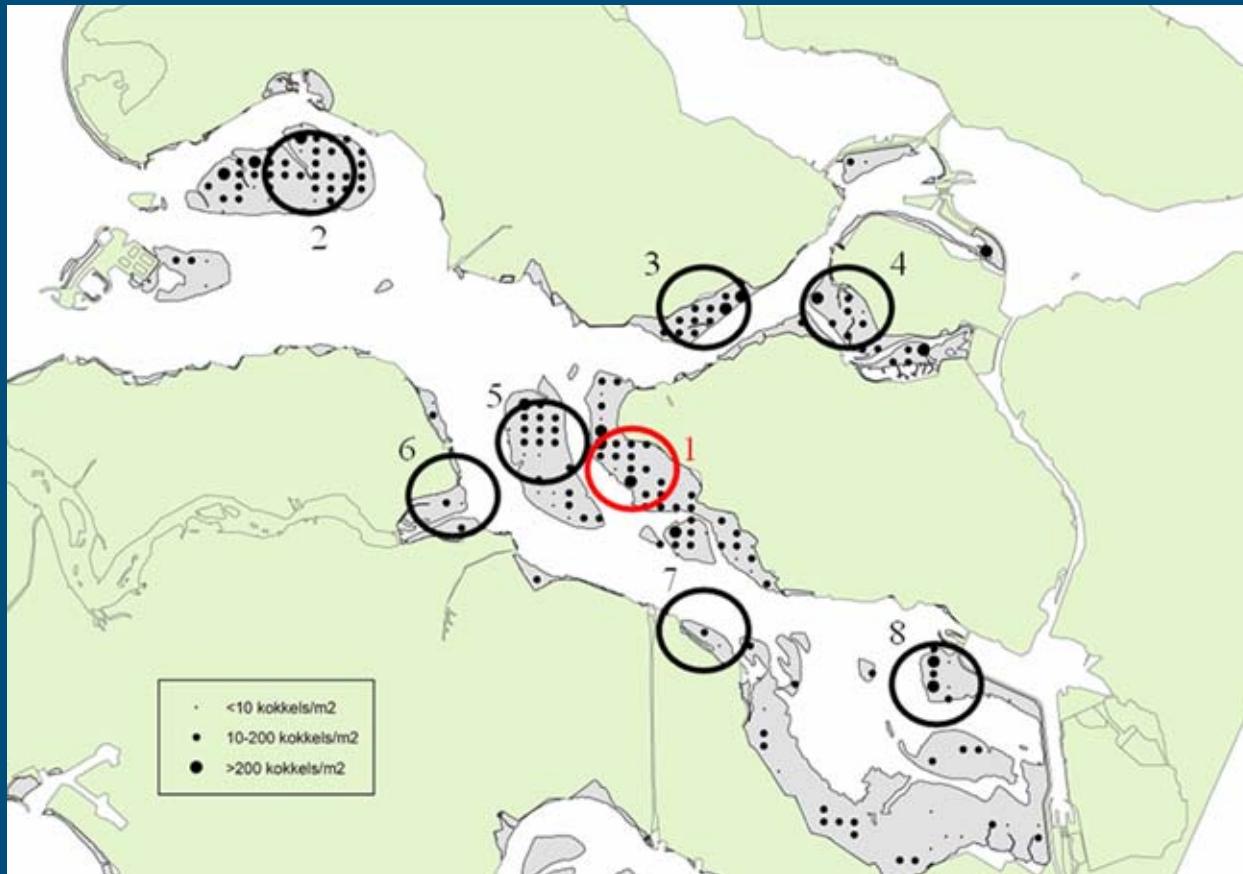
# Cockle mortality in the Oosterschelde

- June 2010, large number of live, moribund and dead cockles reported on the surface at one location in the Oosterschelde
- Parasite infection suspected
- After initial investigation at the site a survey was set up to investigate other sites in the Oosterschelde
- Also from two sites in the Wadden Sea a large number of surfacing cockles was reported



Vliehors, Wadden Sea  
Picture Dick Bruin

# Collection sites



Picture Karin Troost

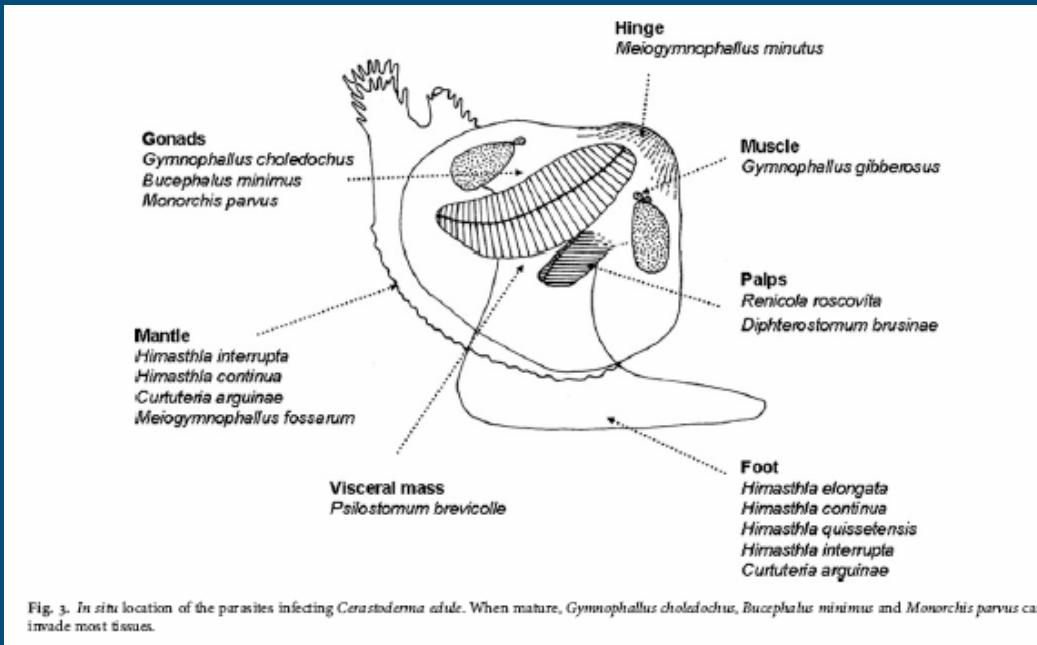


# Methods

- Cockles were collected at low tide from 8 sites in the Oosterschelde and 2 sites in the Wadden Sea
- At each site a batch of 30-60 surfacing and 30-60 buried cockles were collected
- At the lab 20-30 individuals were investigated from each batch by squash preps of different tissues on the presence of digenean trematodes.
- From 2 sites in the Oosterschelde and the 2 sites in the Wadden Sea histological slides were prepared.
- Remaining cockles were fixed in EtOH.



# Identification of trematodes in cockles



## Additional help (by email):

Matt Longshaw (CEFAS, UK)

Xavier de Montaudouin (Univ. Bordeaux, FR)

David Thieltges (NIOZ, NL)

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## REVIEW

# Digenean trematode species in the cockle *Cerastoderma edule*: identification key and distribution along the north-eastern Atlantic shoreline

XAVIER DE MONTAUDOUIN<sup>1</sup>, DAVID W. THIELTGES<sup>2</sup>, MÉRIAME GAM<sup>3</sup>, MANUELA KRAKAU<sup>4</sup>, SUZANA PINA<sup>5</sup>, HOCEIN BAZAIRI<sup>3</sup>, LAURENT DABOUINEAU<sup>6</sup>, FERNANDA RUSSELL-PINTO<sup>5</sup> AND K. THOMAS JENSEN<sup>7</sup>

# Trematodes, cockle 2<sup>nd</sup> intermediate host

## *Himastla* species

Location: mantle, foot

1<sup>st</sup> host: periwinkle

Final host: waterbirds



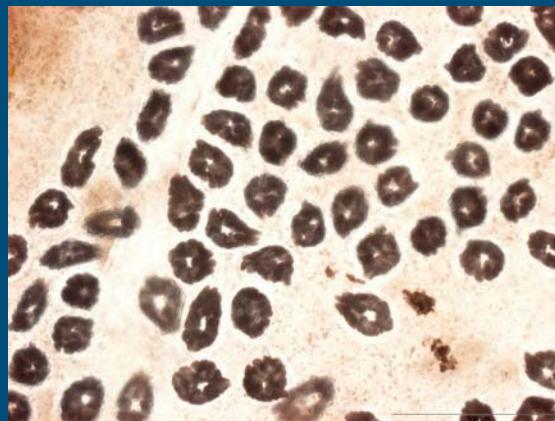
## *Parvatrema minutus*

(Syn. *Meiogymnophallus minutus*)

Location: cyst near hinge

1<sup>st</sup> host: furrow shell

Final host: oystercatcher



- Although trematodes with the cockle as second intermediate host can be present at high prevalence impact on the population is generally low

# Trematodes, cockle 1<sup>st</sup> intermediate host

## *Bucephalus minimus*

Location: gonads, viscera

2<sup>nd</sup> host: goby

Final host: sea bass

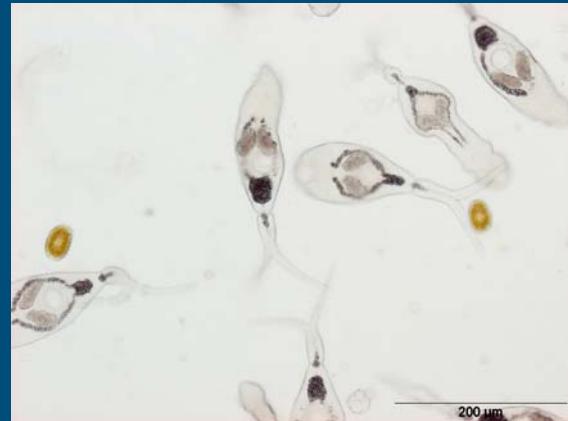


## *Gymnophallus choledochus*

Location: gonads, viscera

2<sup>nd</sup> host: polychaetes, cockle

Final host: waterbirds



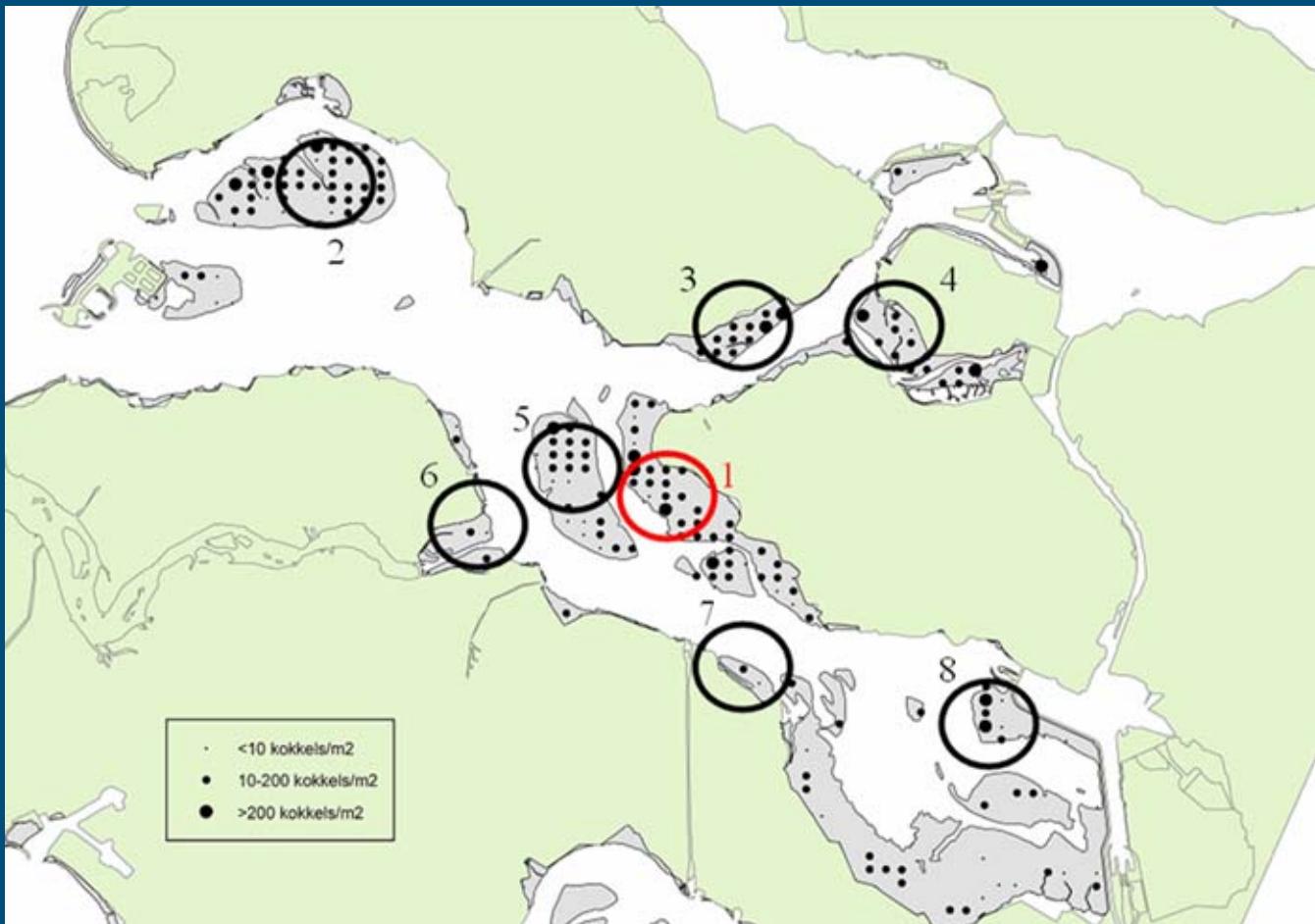
- Trematodes with the cockle as first intermediate host have been associated with summer mortalities of cockles

# Results analysis cockles on trematodes OS

- Results of first sampling (17 June 2011)
  - Slikken van de Dortsman
  - Yerseke Bank (reference site)

Site	Position	<i>B. minimus</i> (%)	<i>G. choledochus</i> (%)	<i>Himasthla</i> spp. (%)	<i>P. minutus</i> (%)
Slikken vd Dortsman	surface	<b>70</b>	4	81	56
Slikken vd Dortsman	buried	<b>52</b>	0	72	68
Yerseke Bank	high in tidal zone	12	0	12	0
Yerseke Bank	low in tidal zone	28	0	0	0

# Collection sites



# Results analysis cockles on trematodes

- Comparison of sites 7 sites in the Oosterschelde  
(30 June 2011)

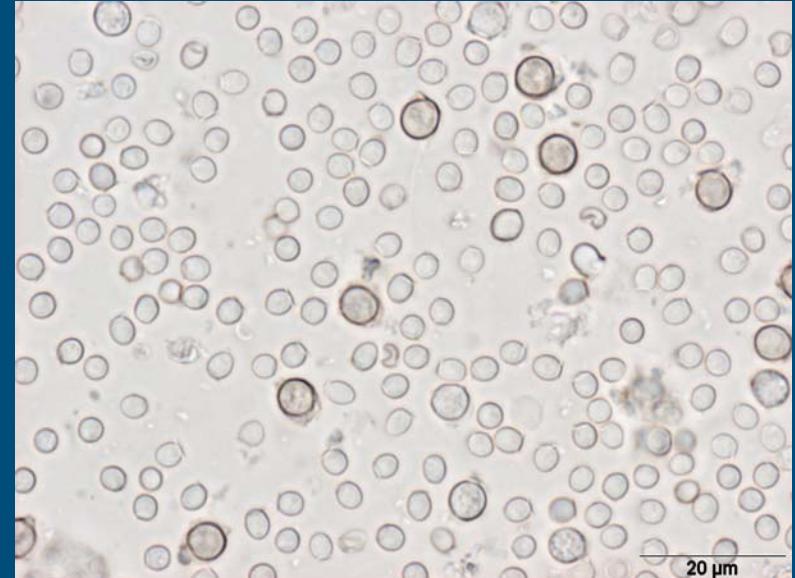
Surface	<i>B. minimus</i>	<i>G. choledochus</i>	<i>Himasthla</i> spp.		<i>P. minutus</i>	
Location	%	%	%	intensiteit	%	intensiteit
2 Roggenplaat	11	0	78	10 - >50	78	100 - > 400
3 Slikken van Viane	30	5	100	2 - >50	80	~50 - >200
4 Krabbenkreek	0	5	75	3 - >50	20	~10 - ~20
6 Slikken van Kats	70	5	100	~50 - >400	20	~10 - ~50
5 Vondelingsplaat	5	5	63	1 - 20	5	>100
1 Slikken van de Dortmans	40	20	100	>100 - >400	90	~50 - >400
8 Bergsediepsluis	25	0	85	1 - >50	10	~5 - ~50

Buried	<i>B. minimus</i>	<i>G. choledochus</i>	<i>Himasthla</i> spp.		<i>P. minutus</i>	
Location	%	%	%	intensity	%	intensity
2 Roggenplaat	0	0	90	20 - >100	80	>50 - >400
3 Slikken van Viane	10	0	80	2 - ~40	85	~50 - >200
4 Krabbenkreek	5	0	55	2 - >50	15	~5 - ~10
6 Slikken van Kats	25	5	100	~50 - >400	30	~20 - >50
5 Vondelingsplaat	5	0	75	1 - 20	0	-
1 Slikken van de Dortmans	30	0	95	~100 - >200	75	~50 - >200
8 Bergsediepsluis	25	0	80	~10 - >50	20	~10 - ~20



# Haplosporidian infections in cockles

- At 3 sites in the Oosterschelde cockles were observed with mass sporulation of a haplosporidian parasite
  - Vondelingsplaat 2/19
  - Bergsediëpsluis 1/20
  - Krabbenkreek 5/20



# Haplosporidian infections in cockles

- Of the 8 infected specimen 4 were double infected with two species!

ID	Locatie	Position	Number of haplo species (fresh/histology)
10010825-A7	Vondelingsplaat	Surface	2 species
10010825-A8	Vondelingsplaat	Surface	1 species
10010825-E15	Bergsiediepsluis	Surface	2 species
10010825-N1	Krabbenkreek	Surface	1 species
10010825-N3	Krabbenkreek	Surface	1 species
10010825-N4	Krabbenkreek	Surface	2 species
10010825-N12	Krabbenkreek	Surface	2 species (mainly 1)
10010825-N18	Krabbenkreek	Surface	1 species

- With sequencing using conserved haplosporidian primers sequences of *Haplosporidium edule* and *Minchinia tapetis* could be obtained





Vliehors, Wadden Sea  
Picture Bram Fey

# Results analysis cockles on trematodes Wad



- Site: • Schiermonnikoog, Oude wal

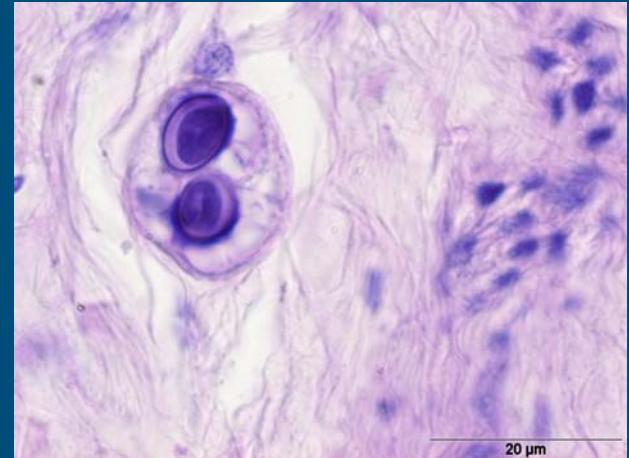
	<i>B. minimus</i>	<i>G. choledochus</i>	<i>Himasthla</i> spp.		<i>P. minutus</i>	
Position	%	%	%	intensity	%	intensity
Surface	0	0	15	~1 - ~10	45	>20 - >50
Buried	0	10	70	~2 - ~20	50	~20 - >50

- Site: • Vlieland, Vliehors

	<i>B. minimus</i>	<i>G. choledochus</i>	<i>Himasthla</i> spp.		<i>P. minutus</i>	
Position	%	%	%	intensity	%	intensity
Surface	43	10	60	1 - 7	87	>50 - >400
Buried	23	0	30	1 - 3	87	~100 - >400

# Results histological analysis cockles

- No listed diseases (OIE/EU) were observed by histology
- No indication for viral infections
- Prevalence of trematodes is reflecting the fresh observations.
- Few bacterial infections
- High prevalence of gregarine parasites (*Nematopsis* sp.) in cockles from "Slikken van de Dortsman"



# Summary and conclusions

- Digenean trematodes are part of the biodiversity in these area's
- However especially the high prevalence of *B. minimus* is exceptional
- *B. minimus* contributed for a large part to the mortality
- Initial cause/trigger is unknown



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IMARES, NL: Anneke van den Brink, Karin Troost, Tom Ysebaert

NIOZ, NL: David Thieltges

CEFAS, UK: Matt Longshaw, Kelly Bateman

Université de Bordeaux, FR: Xavier de Montaudouin

#### Collecting:

Oosterschelde: Gert-Jan van Veen and crew "Valk", Ad van Gool, Tim Schellekens

Wadden Sea: Jeroen Jansen, Arnold Bakker, Bram Fey and crew "Krukkel" and "Phoca"

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