In situ Measurements of Cardiac Activity Characteristics of Indigenous Macrobenthic Invertebrates for Water Quality Bioindication

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Efficiency of monitoring of water resources has fundamental value for rational aquatic ecosystems quality management. Now for water resources management the special legislations and directives are developed for conducting biological monitoring of potential danger of harmful and emerging substances in surface, soil, sea, waste and drinking waters (Water Frame Directive, BSAP, HELCOM).

They know about their habitat quality better than we

Living organisms very well “know” the state of the environment they habitat. This “knowledge” is displaying in various way. It could be sharp changes of behavior or physiological reactions in a case of accidental environmental toxic pollution, or slow change of physiological parameters as a result of absorption and accumulation of pollutants.

Why benthic invertebrates?

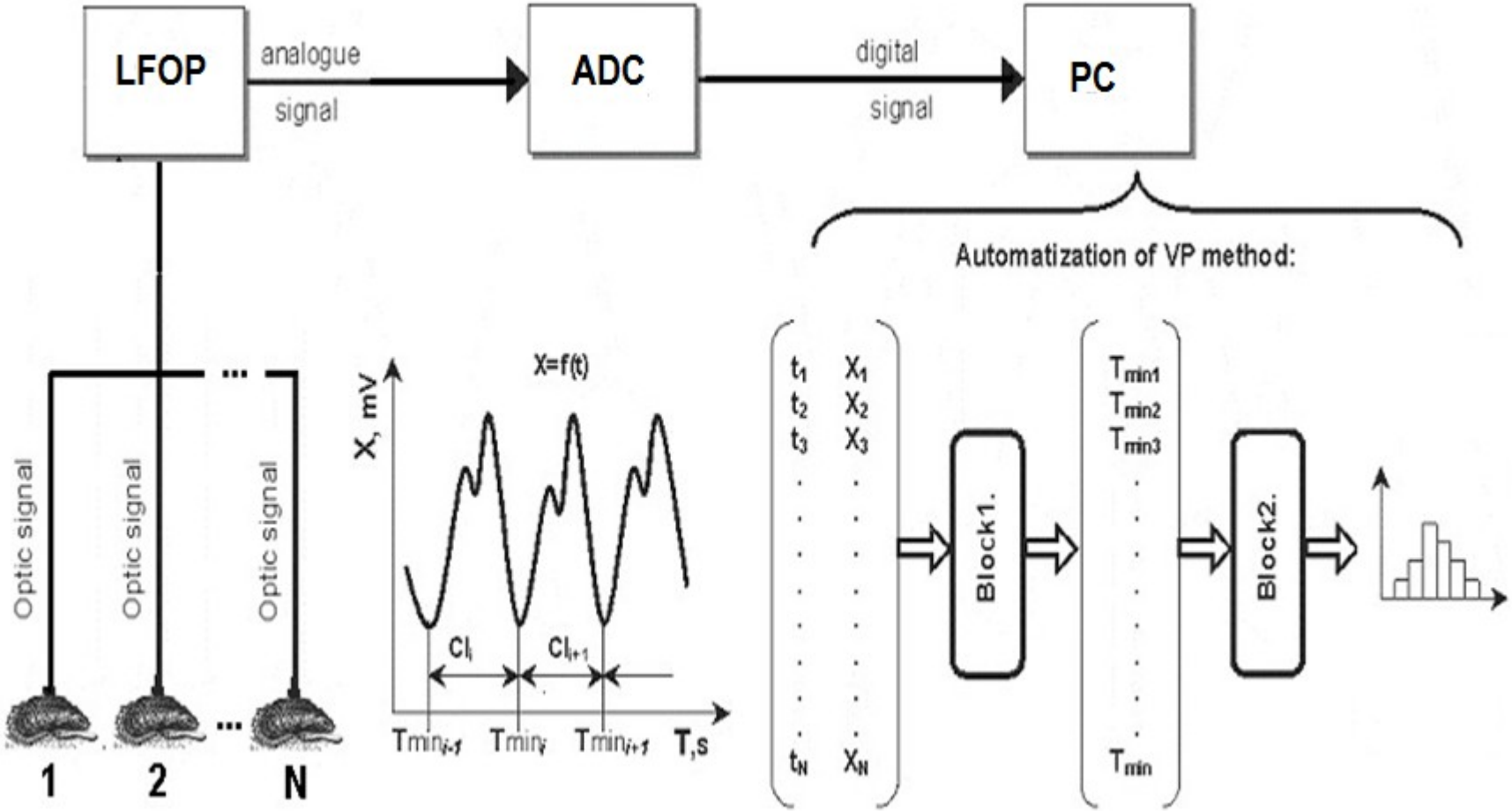
- Wide spread organisms in aquatic ecosystems
- Organisms with low locomotor activity or sedentary organisms
- Shell-clad species (relevant for attachment sensors on their exoskeleton)

Why cardiovascular system?

- One of the main physiological systems
- Fast-response
- Reflects other visceral systems reactions
- Rhythmicity

In 1999 a fibre optical non-invasive method of cardiac activity monitoring in selected invertebrates with exoskeleton was developed by our scientific group, and now we have experience in measurements of cardiac activity in about 25 freshwater and marine species of crustaceans (crayfish, crabs) and mollusks (Bivalvia and Gastropoda).

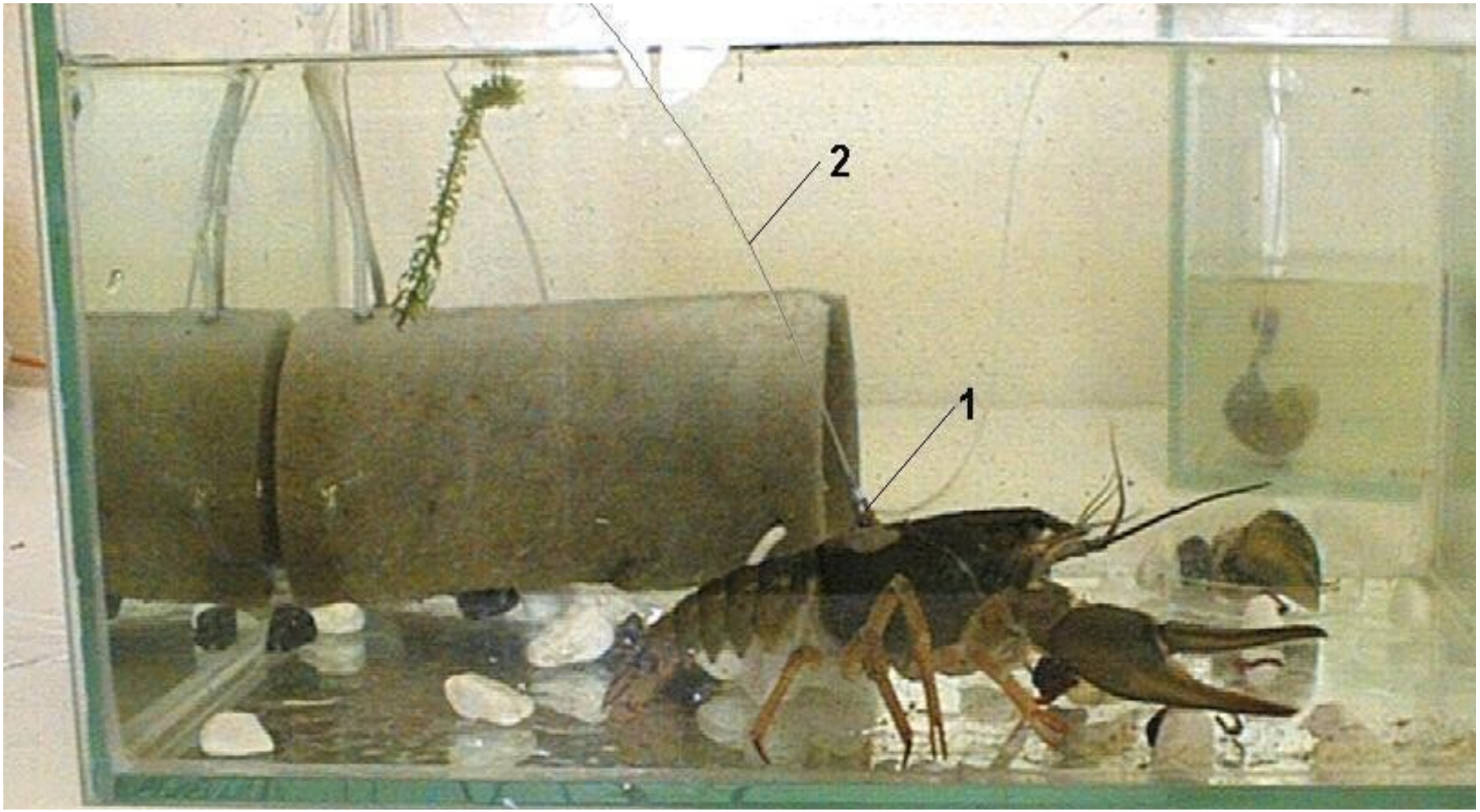
Block-scheme of experimental set up and automatic data processing



Stages of data analysis for a sample of cardiac intervals (usually 50 or 100 intervals) to obtain the variational pulsometry characteristics: heart rate (HR), standard deviation (SD).

Mussels with the fibre-optical sensors on their valves.

Set-up for registration and analysis of the cardiac activity of mussels (from 8 to 16 animals simultaneously)



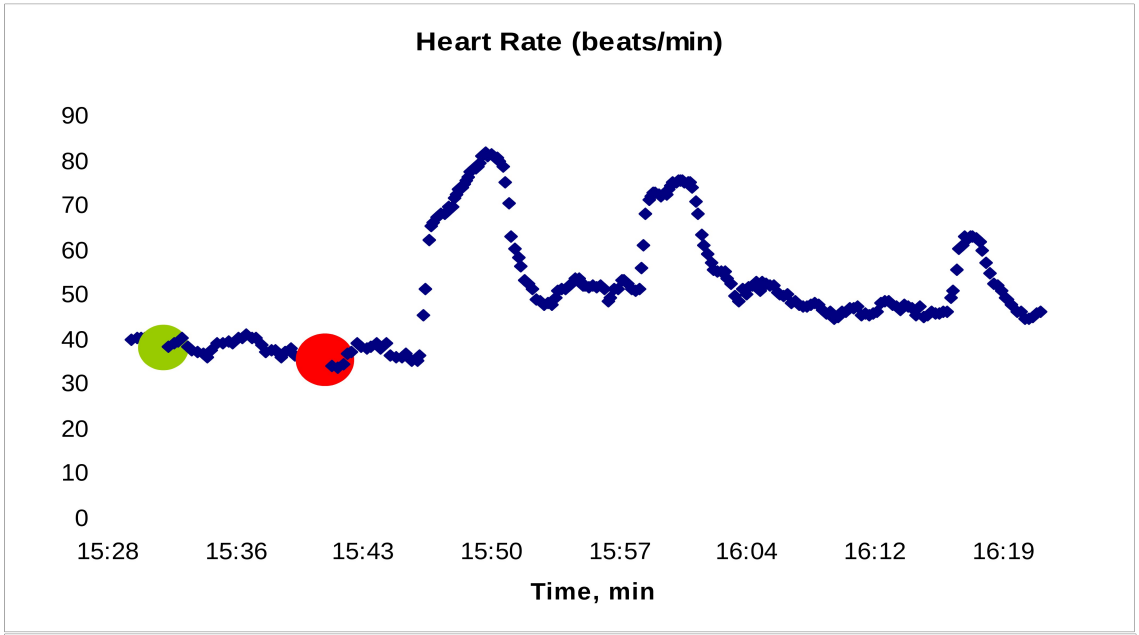
Crayfish with attached fiber-optical sensor on its carapace.

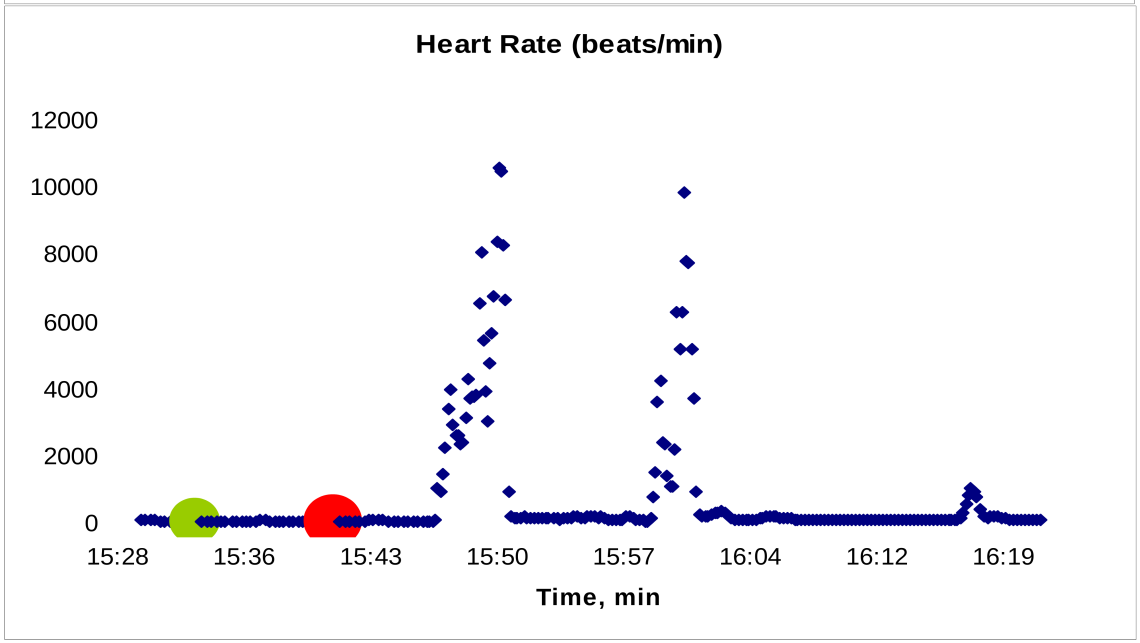
1 – sensor holder glued on the carapace;
2 – fibre-optic cable

A few modifications of bioelectronic system for industrial biomonitoring of surface and sewage water quality BioArgus based on this method were developed and used in industrial operation about 8 years at more than 10 water supply stations and wastewater treatment plants of St.-Petersburg and Khabarovsk.

Crayfish are used there as bioindicators of water quality.

Photograph of system for industrial biological water quality monitoring BioArgus mounted at water supply station.





The experiments with toxic military chemical agents of neural action

It is known that critical salinity for freshwater species is in the range of 7-8 psu (V.Khlebovich, 1974). Freshwater crayfish demonstrate salinity tolerance in the range up to 12 psu. The explanation is in the high salinity of their hemolymph. Kholodkevich (2013) proposed to use *Astacus leptodactylus*, dwelling Neva Bay, as test organism in described bioelectronic system for monitoring along FerryBox route (max salinity 6 psu). Preliminary studies showed that keeping of crayfish in water of mentioned salinity during 2 months did not lead to significant changes in their functional state. This give rise to principle possibility to advancement of FerryBox system by involving in of bioelectronic system. Creation of such bioanalytical complex will significantly increase the ecological importance of carried out *in situ* measurements with the minimum increase of operational expenses.

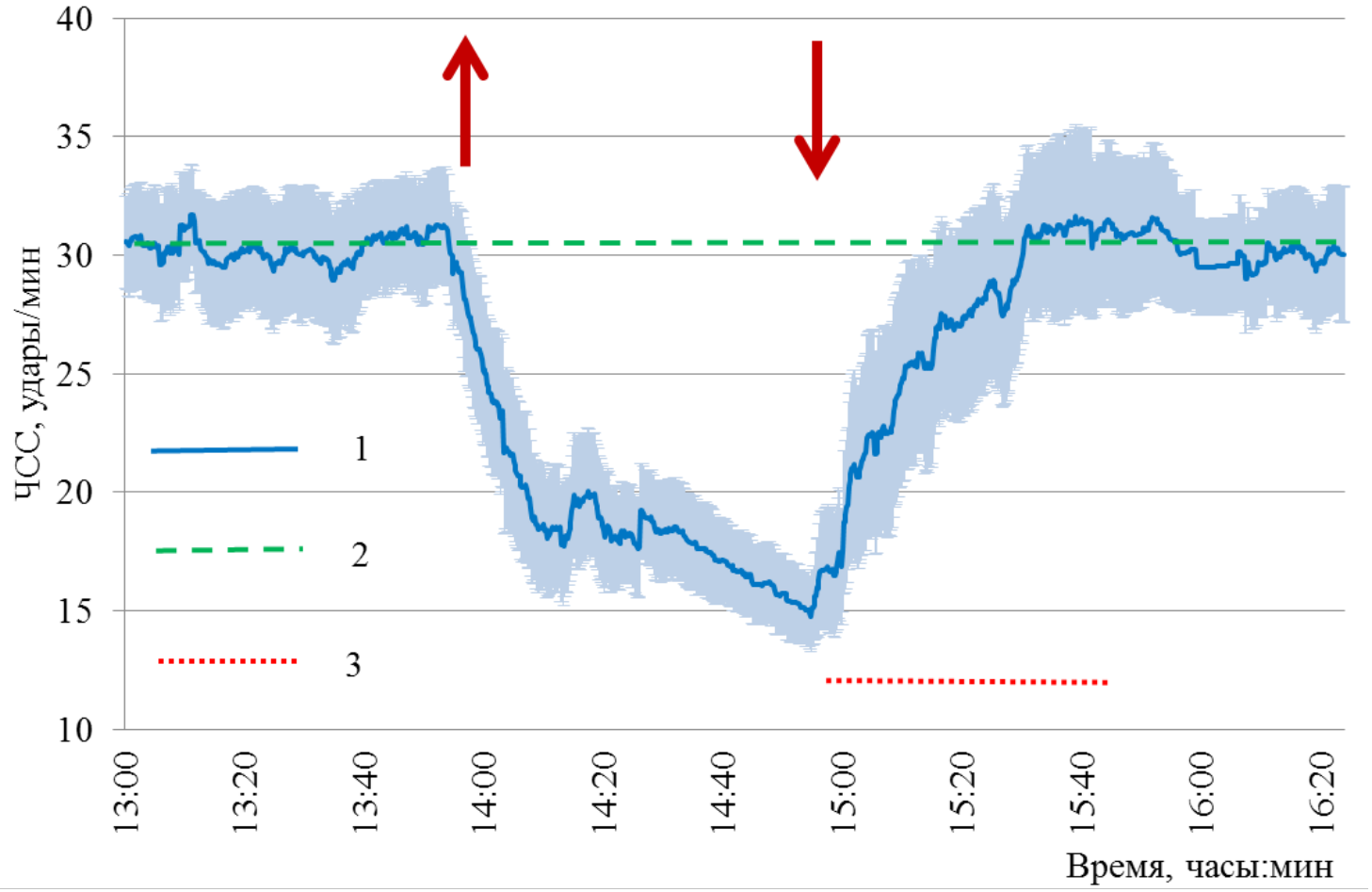
To evaluate the physiological state of sentinel species used, it was proposed to apply *active bioindication method* based on short-term standardized stress load (Kuznetsova et al., 2008; Kholodkevich et al., 2009). This allows to measure recovery of cardiac activity (and organism as a whole) after loading by evaluation of adaptive capacities of species exposed to toxic action (chronic or acute).

The main proposed ecotoxicological biomarkers worked out in our part of the BONUS/BEAST project were:

- duration of recovery of heart rate (T_{rec}, mins) after rapid change of medium salinity, test used as functional load;

- coefficient of HR variation (CV, %) over the group of tested organisms after removal of load.

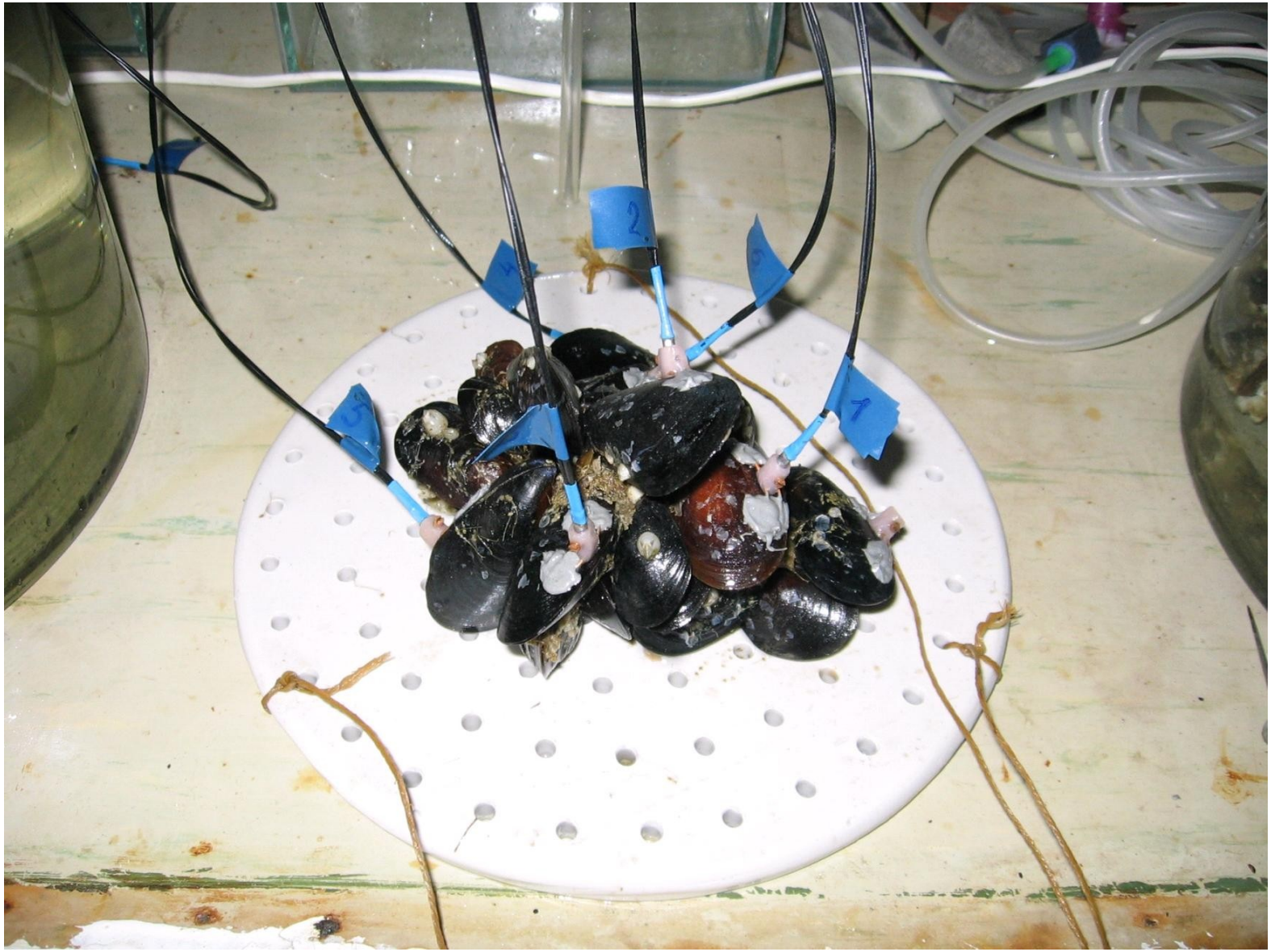
Procedure of HR recovery time calculation



HR trend under 1-h salinity change and during restoration of background pattern. Arrows indicate the period of salinity test. Time needed for restoration of background HR values (green line 2) after change to ambient salinity is indicated by red dotted line 3.

Salinity gradient and distribution of hydrobionts in the Baltic Sea sub-regions

Degree of danger of undesirable physico-chemical impacts on aquatic ecosystems most objectively can be estimated by their influences on the health of indigenous species of hydrobionts



Druse of mussels with the fibre-optic sensors on their valves before deployment in cages at study sites

Examples of the caging method in use

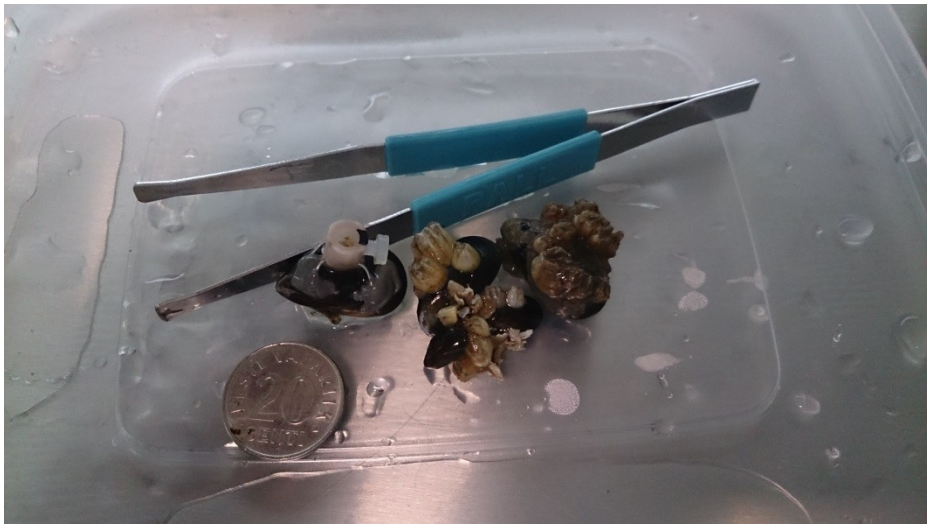
Assessment of biological effects of anthropogenic caused stress:

Method was approbated in the EU-funded projects for the Baltic Sea sub-regions assessment:

•BONUS+BEAST Program (Biological Effects of Anthropogenic Chemical Stress: Tools for the Assessment of Ecosystem Health);

•HYDROTOX (Water Toxicity Estimation)

Preliminary stage of future joint experiments (SRCES RAS, Russia and TTU



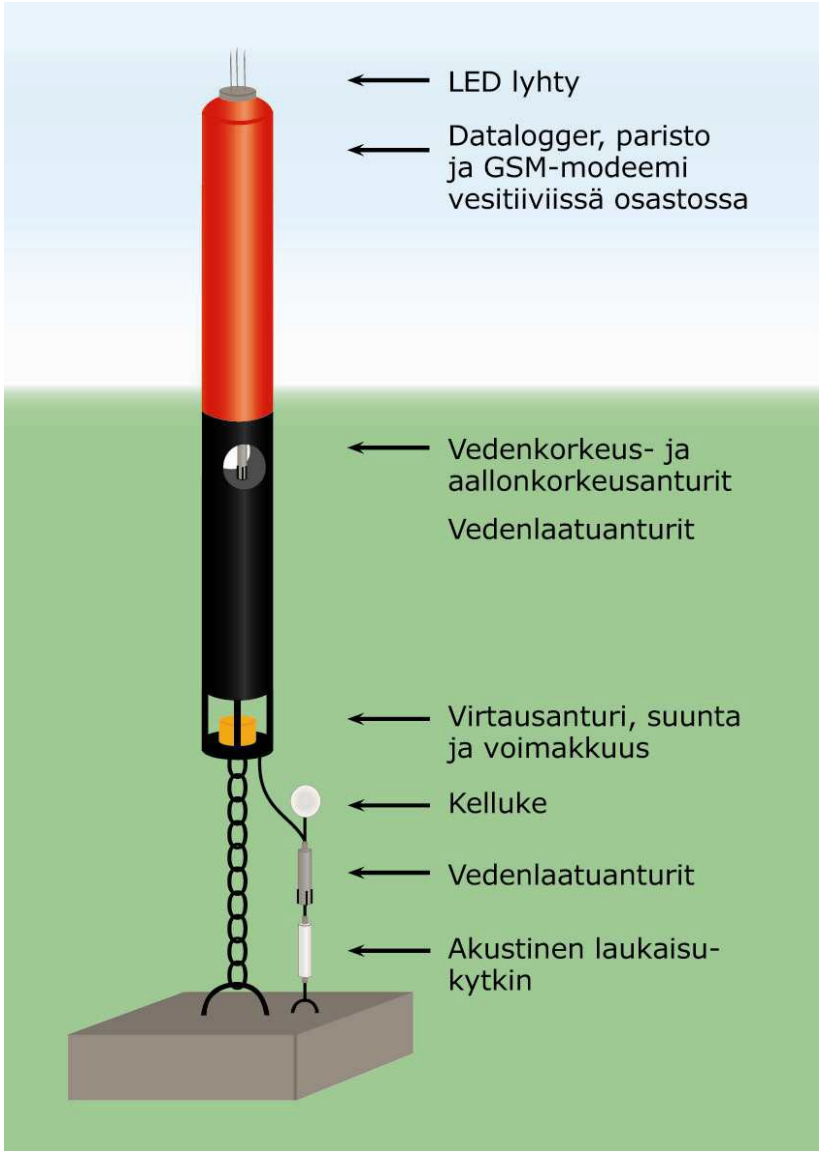
Monitor data according needs

Environmental data

- Oil spill detection
- Oxygen content
- Algae status
- Salinity
- Turbidity

Physical marine data

- Wave height
- Water level and tides
- Current; direction and strength
- Water temperature



CONCLUSIONS

- ***In situ*** **measurement of cardiac activity in selected invertebrates is a useful early diagnostic tool for surface water quality assessment based on responses of biota to environmental challenges;**
- **Described system in joint use with FerryBox could detect integrated biological effects of mixture of pollutants on marine and brackish water organisms;**
- **Ecological efficiency of autonomous underwater buoys could be significantly advanced by including in on-line bioelectronic system of cardiac activity monitoring in its structure accompanied with the use of caged organisms testing.**


Thank you
for attention!