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The fluorescence in situ hybridization - an innovative method in the circular solutions for resources recovery in Municipal enterprise for waste treatment - Sofia

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- ✓ *New technologies*
- ✓ *New indicators*
- ✓ *New control systems*

The circular economy and the sustainable development impose a waste minimization and a continual use of resources. New technologies and innovative control systems have been developed and implemented in waste treatment plants.

This study aims to present the role of the fluorescence *in situ* hybridization as a tool in the management of different resource recovery technologies.

SWOT analysis of the FISH

Strengths

Precise technique for control
The fixed samples could be storage long time
It detects the cultivable and the non-cultivable microorganisms
Gives information about the quantity of the target microorganisms
Gives information about the localization of the target microorganisms
Gives the opportunity to be studied different groups of microorganisms
The software is able to extract information better than human and could avoid the subjectivism in the images interpretation
The dveing with DAPI, a step of the FISH protocol, gives additional information about the polyphosphate accumulation

Weaknesses

A multi-step protocol that is a precondition for experimental mistakes
The use of each different probe requires a different concentration of the hybridization and the washing buffer as well as different hybridization time
It includes the use of expensive chemicals and equipment
The study of some groups of microorganisms requires the implementation of different methods for cell wall permeability
It requires qualified personnel
It is necessary the presence of a specialized software for microbial ecology and FISH

Opportunities

An opportunity for observation of the hybridized samples on a scanning confocal laser microscope for removing of the out of focus fluorescence and studying the structure in deepness
Different variations of the FISH methods exist

Threats

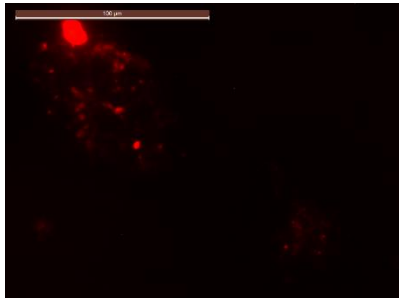
An unspecific bonding of the probe to the sample
Background fluorescence
Experimental mistakes
Unvenetration of the probe in the cell due to different specifics of the microorganisms cell walls

ЕВРОПЕЙСКИ СЪЮЗ
ЕВРОПЕЙСКИ СТРУКТУРНИ И ИНВЕСТИЦИОННИ ФОНДОВЕ
clean circle
ОПЕРАТИВНА ПРОГРАМА НАУКА И ОБРАЗОВАНИЕ ЗА ИНТЕЛИГЕНТЕН РАСТЕЖ

Case study – Municipal enterprise for waste treatment – Sofia (MEWT)

Fig. 1 Biological treatment plant "Han Bogrov"

Fig. 2 Wastewater treatment plant "Sadinata"



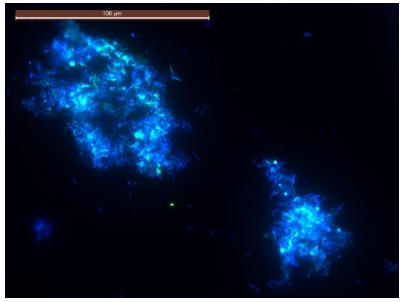
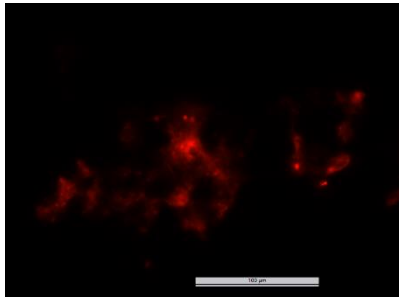


Fig. 3 FISH for Family Methanobacteriaceae and a DAPI dyeing in a digester of BTP "Han Bogrov"



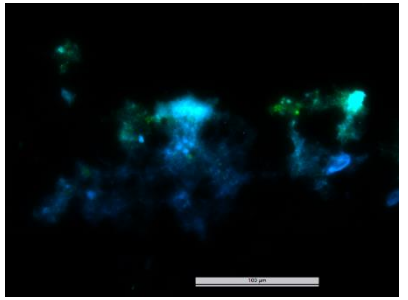


Fig. 4 FISH for Pseudomonas sp. and a DAPI dyeing in a model experiment with activated sludge from WWTP "Sadinata"

Waste management

Resources recovery

СТОЛИЧНО ПРЕДПРИЯТИЕ ЗА ТРЕТИРАНЕ НА ОТПАДЪЦИ

New technologies and innovative control indicators

Fluorescence in situ hybridization analysis for different groups of microorganisms

FISH as a tool in the management of the biotechnologies for resource recovery in MEWT

The FISH technique is based on hybridizing fluorescently labeled probes to ribosomal rRNA in permeabilized whole microbial cells. The method makes it possible within a relatively short time to retrieve information on phylogenetic identities of the cells directly in a sample, and since it also maintains the morphology of the cells it also supplies information on the spatial distribution as well as the number of identified microorganisms.

- FISH for the target microorganism
- DAPI (4′,6-diamidino-2-phenylindole) dyeing

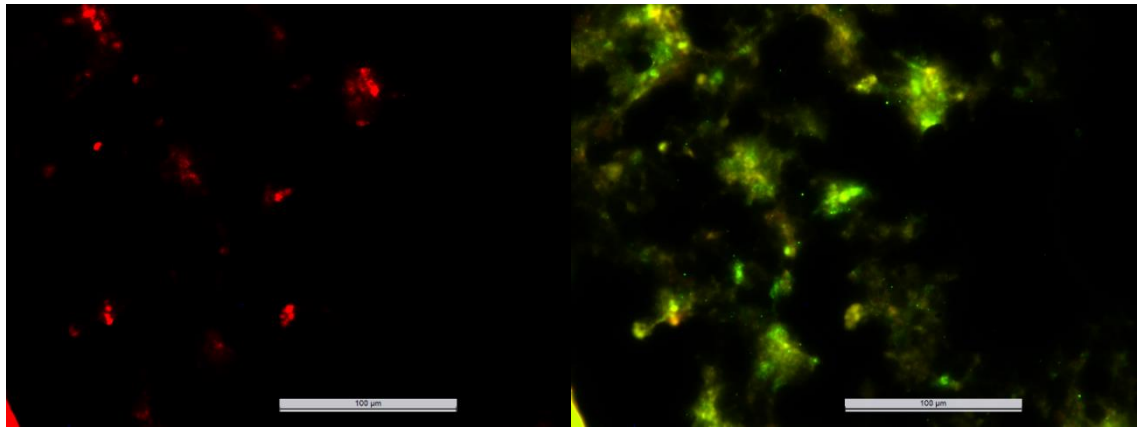


Fig. 5 FISH for Alcaligenes sp. and EUB mix in a model experiment for landfill treatment with an activated sludge from WWTP "Sadinata"

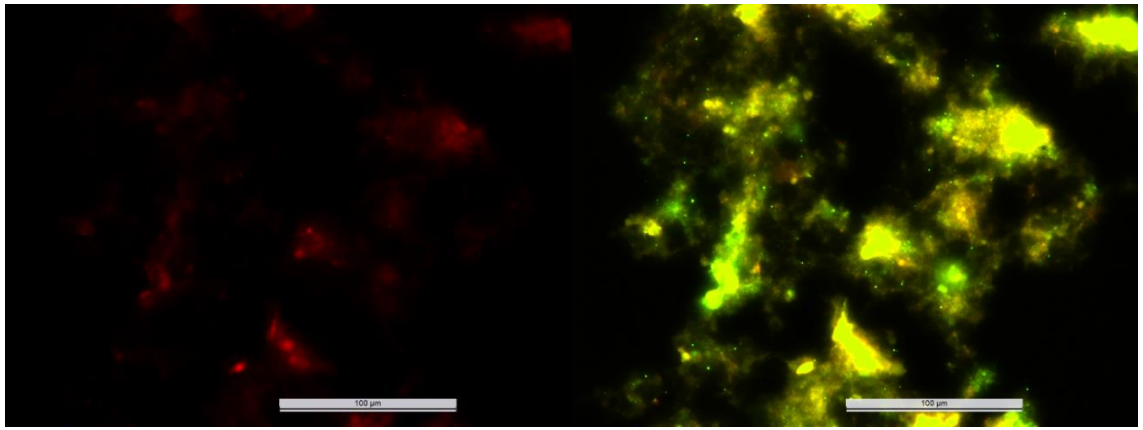


Fig. 6 FISH for Paracoccus sp. and EUB mix in a model experiment for landfill treatment with an activated sludge from WWTP "Sadinata"

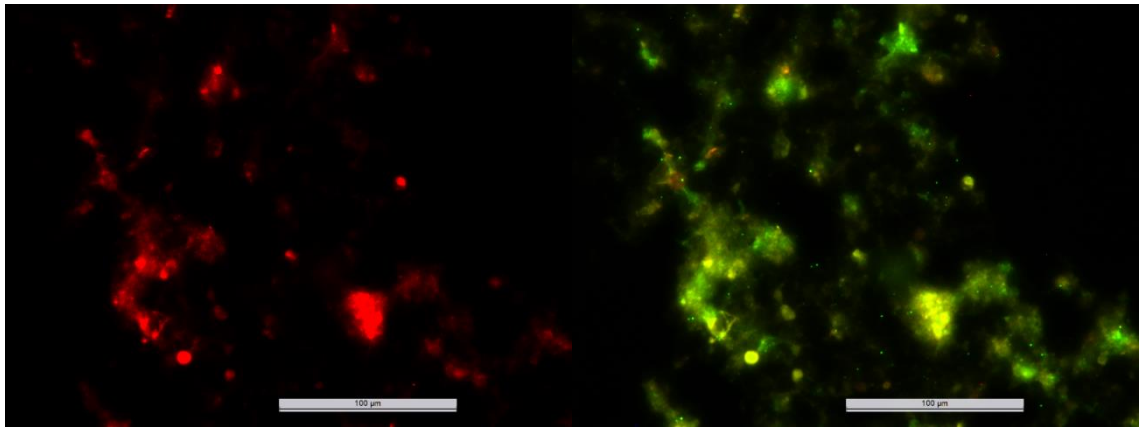


Fig. 7 FISH for cluster Azoarcus-Thauera. and EUB mix in a model experiment for landfill treatment with an activated sludge from WWTP "Sadinata"

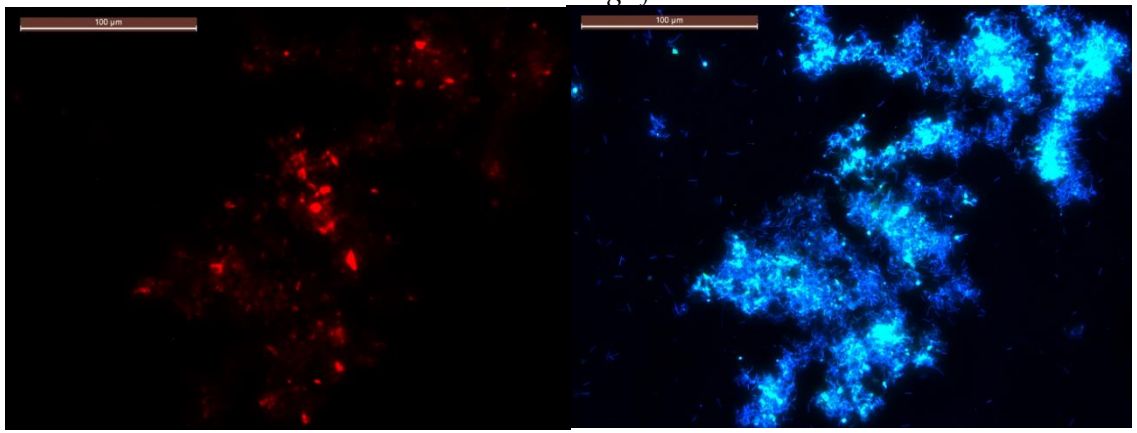


Fig. 8 FISH for domain Archaea. and DAPI dyeing in a digester of BTP "Han Bogrov"

The FISH has been proved its large potential as a method in the management of the resource recovery biotechnologies in MEWT giving information at the same time about the quantity of the target microorganisms, their localization, their relationships and thus it contributes to the understanding of their functioning.

FISH for the target microorganism

DAPI dyeing

FISH for EUB mix

Acknowledgments

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