



Early mastitis detection:
Can Lactoferrin evaluation by EU
funded project MOLOKO biosensor
help?

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Multiplex phOtonic sensor for pLasmonic-based Online detection of contaminants in milK

PROJECT REFERENCE: 780839

START/END: Jan 2018 – March 2022

TOTAL COST: EUR 6,036,381.25

EU CONTRIBUTION: EUR 5,479,159

TOPIC: ICT-30-2017 Photonics KET 2017

12 partners from 8 European countries





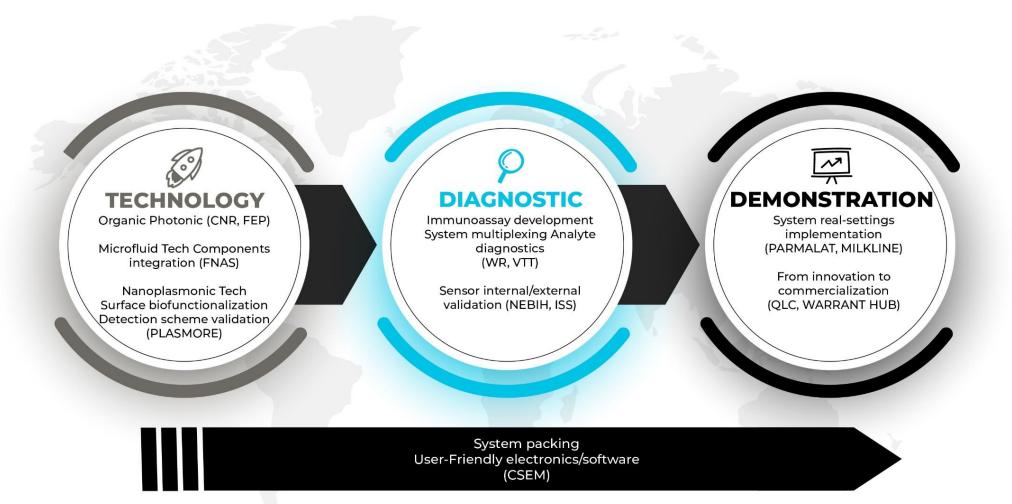
























Parmalat End User Role:

Why a milk processor so involved in an animal health project?

How to achieve to Food Safety goal in the milk supply chain?

Along the very long value chain of milk production and collection, the more **upstream the detection** of analytes for safety and quality, the greater the benefits for all the actors in the value chain













List of target molecules

FOOD SAFETY

Antibiotics

Staphylococcus aureus enterotoxin A and B



k-Casein B





Aflatoxin M1

B-Casein A2

Lactoferrin









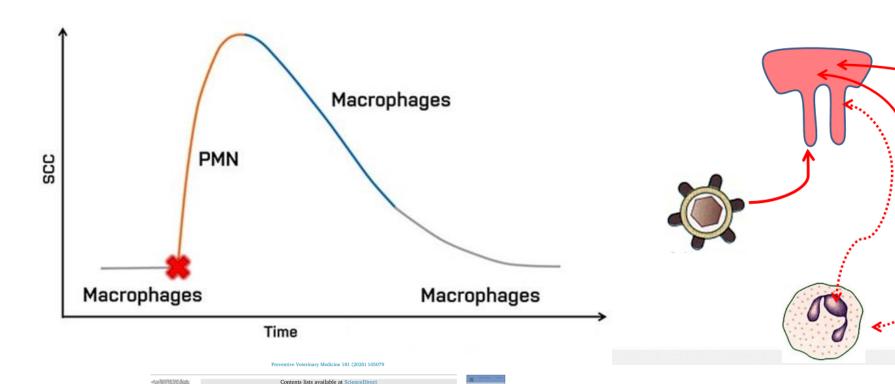




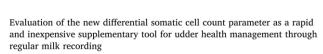




Early mastitis detection: Why Lactoferrin?



Neutrophils migrate from blood to the mammary gland in response to infection and constitute a major host defense mechanism



Preventive Veterinary Medicine

Daniel Schwarz^{0, st}, Debora E. Santschi^b, Jean Durocher^b, Daniel M. Lefebvre^l
*POSS Andytical A/S, Fost All: 1, 3400 Hillered, Demark
*Icaturus, St-Ande-Biellerus, Quide, Canada







Innate immune

defences

cells)

Inflammatory response

Infection is not controlled

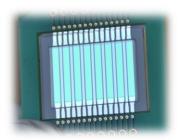
(epithelial







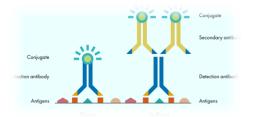
Sensor key components:



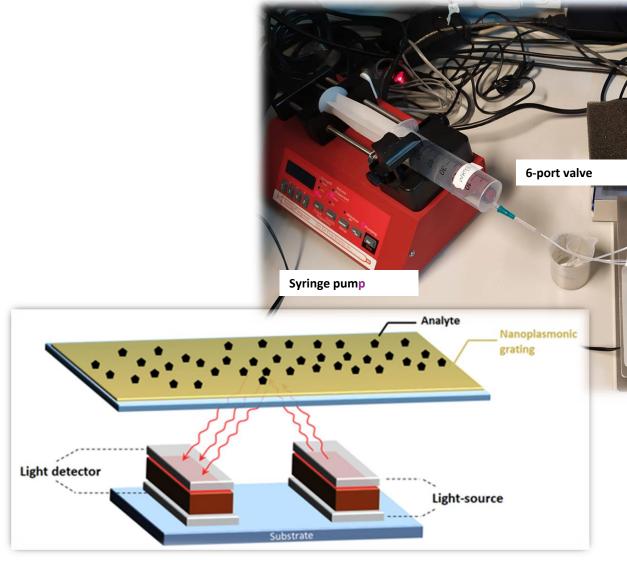
Photonic Organic Module



SPR Sensing Surface



Immunoassay Technology





Load Sample

OPM and Fluidic

MOLOKO sensor







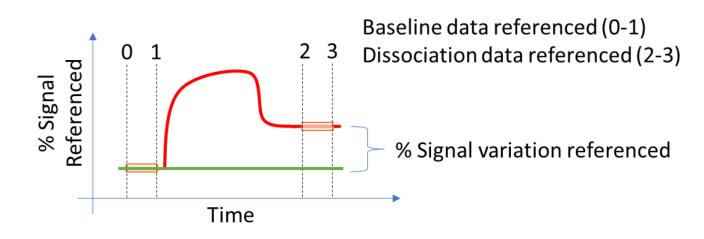


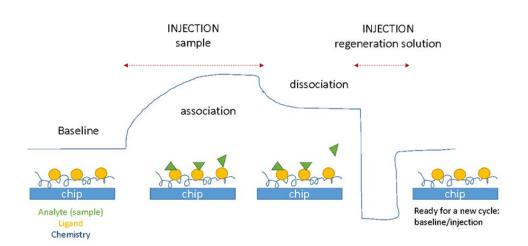






Lactoferrin - Dose-response curve in manual controlled MOLOKO sensor





Sensorgram type for protein tests or calibration of assays and milk samples measurements.

The signal from the reference channel is shown in green and the specific assay channel in red.





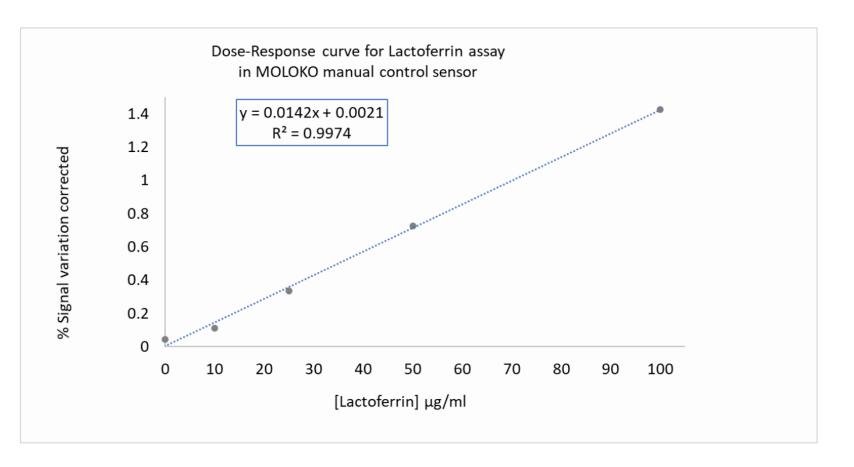








Lactoferrin - Dose-response curve in manual controlled MOLOKO sensor



The Dose-response curve showed a linear correlation between the signal of the MOLOKO biosensor and the concentration of LF.

The LOD for the LF assay was calculated as 3 times the signal of the noise and corresponds to 8.8 µg/ml.













Lactoferrin - On Farm



Real environment evaluation:

2 different herds

100 lactating cows with AMS

600 lactating cows with a milking parlour system

524 individual milk samples tested with gold standard ELISA





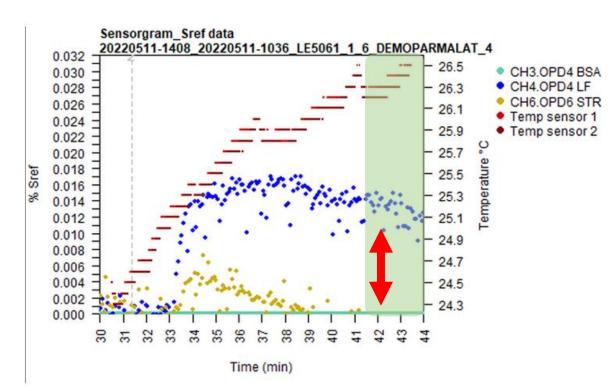


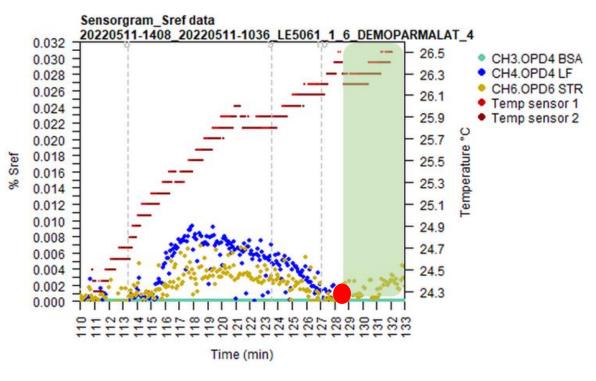






Lactoferrin - On Farm





Interaction of Lactoferrin at 100µg/ml in buffer.

Interaction of milk sample diluted 100 times.

Blue line shows signal from the specific channel for Lactoferrin detection. Green region corresponds to the dissociation phase in which the level of analyte is evaluated













Lactoferrin - On Farm

Individual milk samples have been also analysed for:

- 1. Fat
- 2. Protein
- 3. Lactose
- 4. Somatic Cell Count
- 5. Differential Somatic Cell Count
- 6. Bacteriological culture
- 7. F-
- 8. K+
- 9. Na+
- 10. Ca2+
- 11. Cl-
- 12. NO3-
- 13. NH4+
- 14. pH
- 15. Redox
- 16. Conductivity

And also this parameters have been taken into consideration:

- 1. DIM
- 2. Parity
- 3. Mastitis events

Using this others with an algorithm together with Lactoferrin threshold?





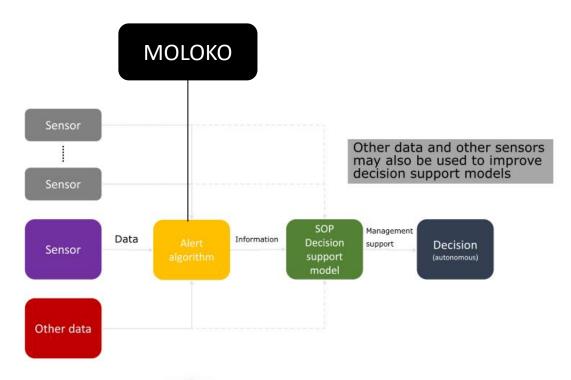








Next step - Precision Livestock Farming (PLF)















Detection results clinical mastitis

Necessary performance (ISO)

- Sensitivity 80% specificity 99% In practise
- Sensitivity 21% specificity 99%
- Sensitivity 50% specificity 90% In theory
- Sensitivity 57% specificity 98% Recently (with SCC):
- Sensitivity 90% specificity 90%

NMC san diego 2022 proceeding -Short course Sensor-based mastitis management (Henk Hogeveen, Ilka Klaas, David Kelton & Alfonso Zecconi)

What about others heard situations? Dry off, subclinical, etc.



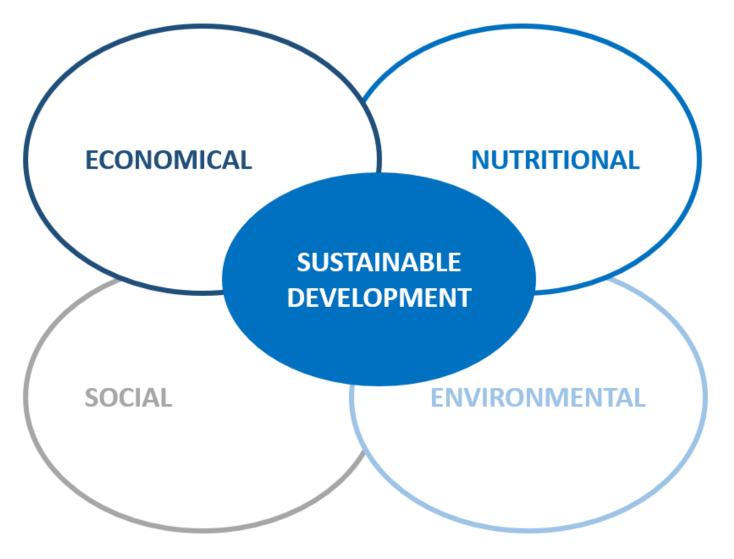








Conclusion:













Grazie per l'attenzione





www.moloko-project.eu