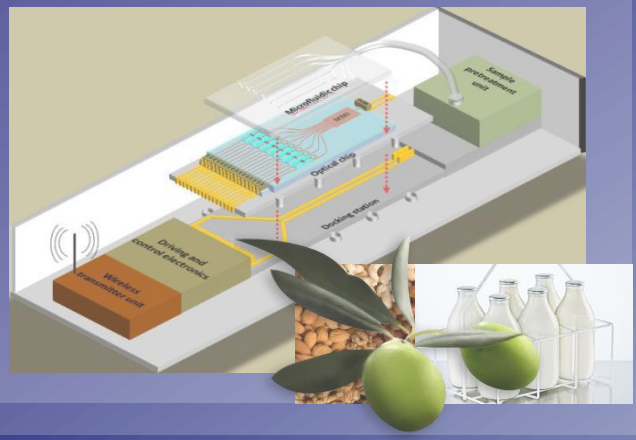
MNBS, Otranto (Italy), June 27-29, 2016

BIOFOS



Micro-ring resonator-based biophotonic system for food analysis

Funded under the seventh Framework Programme (FP7), ICT- STREP **FP7-ICT-2013-10, GA no: 611528**
Start date: 1/11/2013, Duration 36 months, 2.75M€

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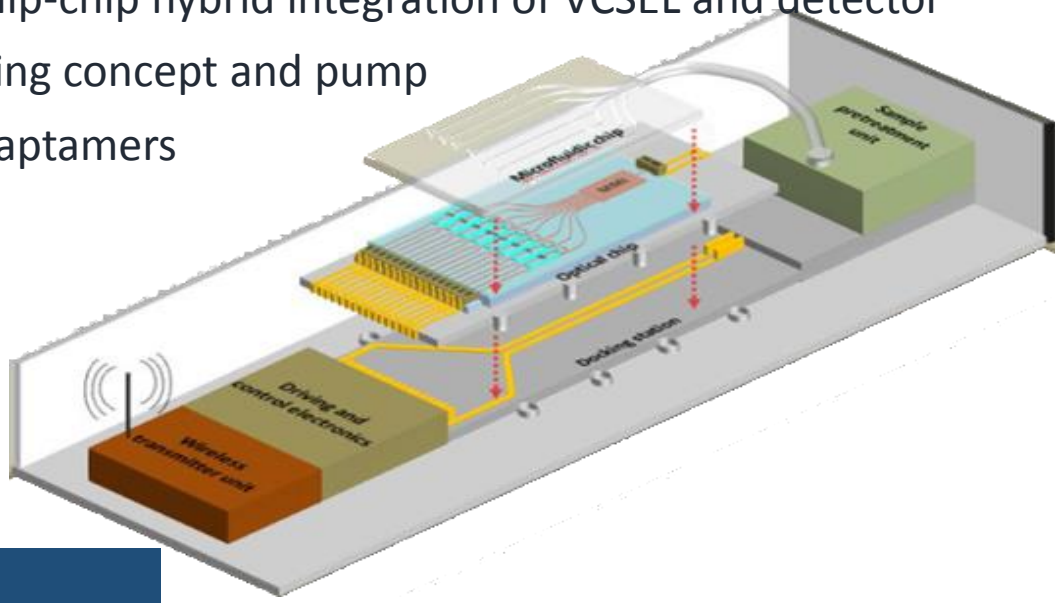
[1]ICCS/NTUA (GR), [2]LIONIX (NL), [3]CSEM (SW), [4]UPVD (FR), [5]BRFFA (GR), [6]Surfix (NL), [7]WU (NL), [8]IRTA (ES), [9]SAXION (NL), [10]COVAP (ES)

BIOFOS GOALS

Portable, cost-effective, multi-analyte biosensing system for on-site monitoring of food quality

- Photonics based on TriPleX platform and flip-chip hybrid integration of VCSEL and detector
- Microfluidic platform with innovative valving concept and pump
- Biological platform based on regenerable aptamers
- Stable and selective immobalization
- Electronics and user interfaces



Food system	Target analyte
Olive oil	Copper, phosmet
Nuts	Aflatoxin B1, ochratoxin A
Milk	Aflatoxin M1, penicillin, lactose

3 food systems – 7 targeted analytes

BIOFOS consortium (1)

10 partners from 5 European countries

Photonics	LioniX BV	The Netherlands
	ICCS/NTUA	Greece
	SAXION University	The Netherlands
Micro-fluidics	CSEM	Switzerland
	LioniX BV	The Netherlands
Biology	Université de Perpignan	France

BIOFOS consortium (2)

Biochemistry	Biomedical Research Foundation, Academy of Athens	Greece
	Surfix BV	The Netherlands
	ICCS/NTUA	Greece
	Wageningen University	The Netherlands
Electronics - Integration	ICCS/NTUA	Greece
	CSEM	Switzerland
End users	Institut de Recerca I technologia Agroalimentaries	Spain
	Sociedad Cooperativa Andaluza Ganadera del Valle de los Pedroches	Spain

BIOFOS: Stakeholders and needs (1)

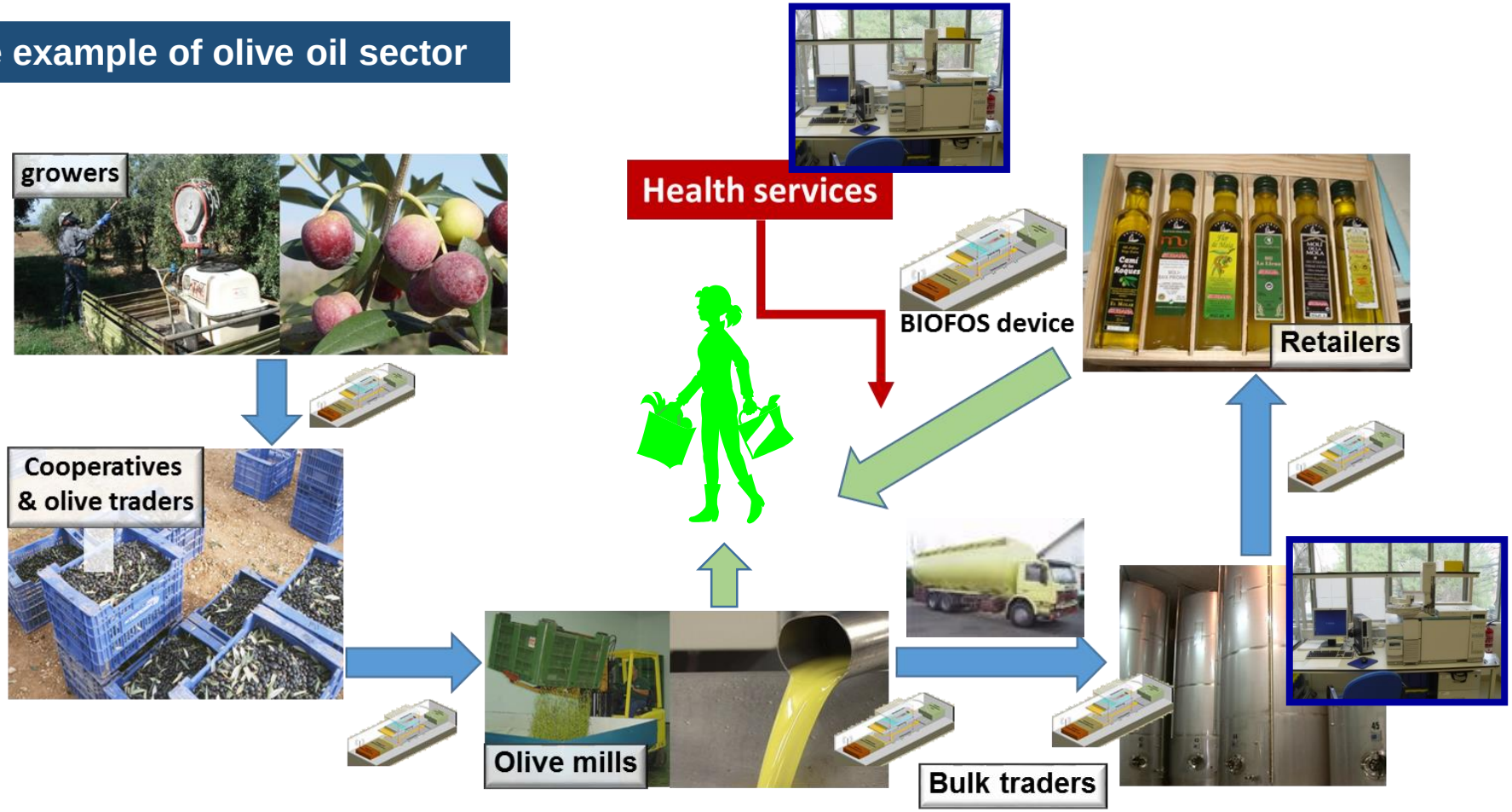
Food industry is the second largest sector in the manufacturing industry of EU, consisting of about *310,000 enterprises*, with *4.8 million employees* and **a large network of stakeholders** (production, process and storage of food products).

Stakeholders

Type/role	Examples	Interest/role
• **Farmers**	Land owners, farm workers, unions, associations	Potential victims, loss of production
• **Agricultural suppliers and services**	Seed suppliers, pesticide manufacturers, fertilizer manufacturers, transport companies	Risk management and risk reduction
• **Food distributors and processors**	Food wholesalers and retailers, transport companies	Potential victims, loss of part of the production
• **Health protection agencies (Regional/National)**	Public health institutions, food standards agencies, regional health boards and environmental health departments	Risk management and regulation; risk communication
• **Environmental protection agencies (Regional/National)**	Ministries of environment, environmental regulatory agencies, local authorities	Risk management and regulation
• **Large agencies (European/International)**	European Commission (Directorates for Agriculture, Environment, Health); WHO, FAO	Risk management and regulation; risk communication
• **Non-Governmental Organizations**	Pesticide action groups, organic farming groups, animal welfare groups	Risk communication; representatives of victims of exposure; lobbyists for action
• **Others**	1) Rural residents, 2) media, 3) scientists (epidemiologists, toxicologists, environmental scientists)	Potential victims of exposure; 2) risk communication, 3) risk analysis & communication

BIOFOS: Stakeholders and needs (2)

The example of olive oil sector



Users and needs along the total of the value chain (from field/farm to commercial stores)

BIOFOS: Stakeholders and needs (3)

User Needs

Screening tool to detect contamination of food samples early in the production chain and prevent contamination of larger quantities

Sensitivity and overall efficiency similar to heavy analytical methods

Portability and **low cost per analysis**

Short time-to-result (from hours to minutes) and **simplicity** (setup, operation)

Multi-analyte detection

Distribution of *Questionnaires* - including actual options chosen for BIOFOS device and demands for final users requirements.

Olive oil sector (>60)	Nuts sector (>10)	Milk sector (>50)
▪ Large companies: producer/ exporter ▪ Non-profit organization: association of olive oil cooperatives/companies ▪ Small/medium cooperative companies ▪ Official laboratories for food analysis	▪ International foundation on nuts and dried fruits - International Nut Council ▪ Large and medium cooperative companies ▪ Official laboratories for food analysis	▪ Dairy farmers ▪ Providers of specialized analysis and measurements services to farmers ▪ Milk processing industries for the production of dairy products

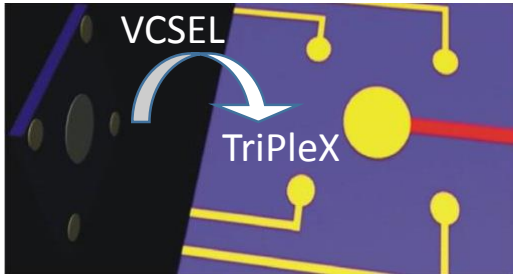
Needs confirmed ✓ **Interest for targeted analytes confirmed** ✓ **Options for interface qualified** ✓

Innovation process and technology translation (1)

BIOFOS approach: Turning the basic technology into manufacturable and low-cost products

Take lesson from optical communications and invest on hybrid photonic integration for:

ultra-compact, ultra low-cost photonic chips



Manufacturability

Cost

Combine the best passive platform for biosensing applications (Si_3N_4 /SiO_2) with cheap InP elements, i.e. VCSEL(aser) and photodiode-array

Take a second lesson from optical communications and develop sophisticated signal processing algorithms in order to

Achieve high performance with conventional resonators, lasers and driving circuits

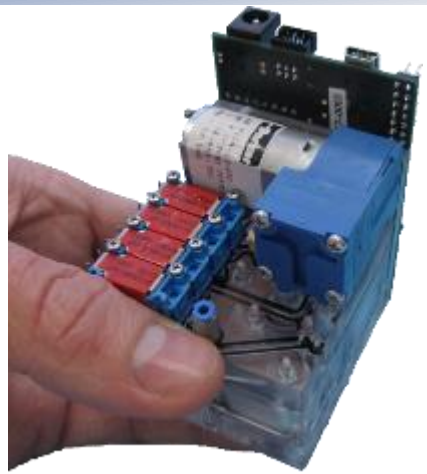
Manufacturability

Cost

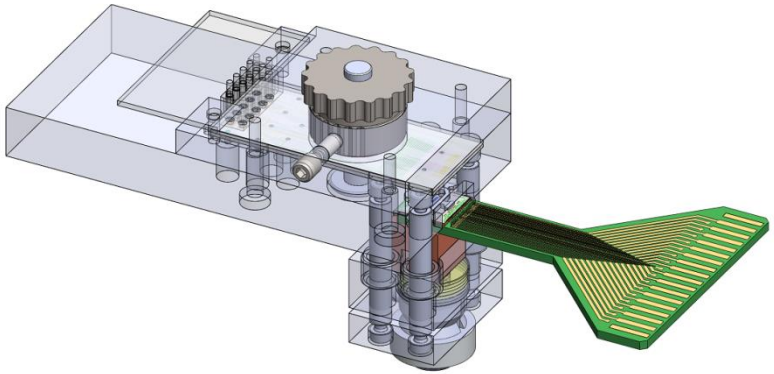
As coherent transmission has showed, algorithms are always cheaper than high-accuracy photonic and electronic hardware

Estimation of costs BIOFOS system

- Cost of instrument objective: EURO ~ 5,000
- Cost of (reusable) cartridge: objective 30x
- Cost of consumables (sample prep + detection)
- Cost per analysis objective: EURO ~ 0.50
- Calculations updated regularly



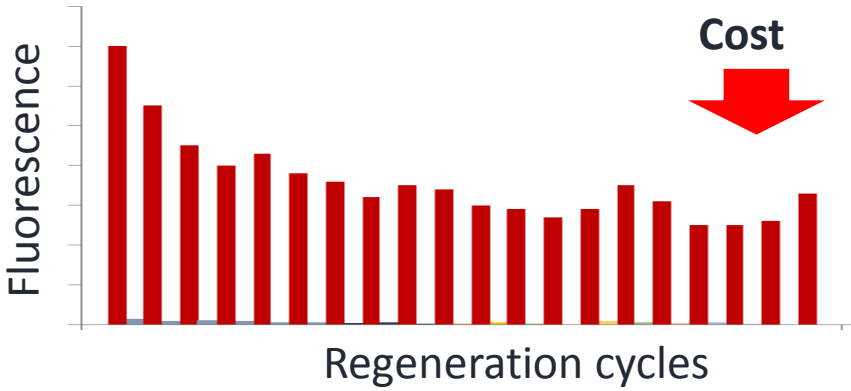
Cost per Test	Internal Volume	Composition, Type, Product No	Cost per Unit	Unit type	Nuts				Oil				Milk							
					Plan A		Plan B		Plan A		Plan B		Plan A1		Plan A2		Plan B Alla+Peni		Plan B Lactose	
					Filter		SPE		Insecticide		Instecticide		Captiva		Filter		SPE		SPE	
					Portion/ Test	Cost/ Test	Portion/ Test	Cost /Tes	Portion/ Test	Cost/ Test	Portion/ Test	Cost/ Test	Portion /Test	Cost/ Test	Portion /Test	Cost/ Test	Portion/ Test	Cost/ Test	Portion/ Test	Cost/ Test
Consumbables																				
Sample																				
Nuts		20g																		
Olive oil		3g																		
Milk	0.005																			
Offline Processes																				
MeOH		SigmaAldrich 34860-4x2.5L CHROMASOLV®, for HPLC, ≥99.9%	24.5	Liter	0.08	1.96	0.08	2	0.01	0.25		0	0.003	0.07	0.005	0.12		0		0
H2O			0.2	Liter	0.02	0	0.02	0		0		0		0		0		0		0
n-hexane		SigmaAldrich 270504-4x2.5L CHROMASOLV®, for HPLC, ≥95%	49.7	Liter	0.05	2.49	0.05	2.5	0.001	0.05	0.005	0.25		0		0		0		0
Binding buffer		PBS1x pH 7.4 +1mM MgCl2	0.281	Liter		0		0		0		0		0	0.045	0.01		0		0
ACN		SigmaAldrich 271004-2L Acetonitrile anhydrous, 99.8%	86	Liter		0		0		0	0.011	0.95		0		0		0	0.01	0.86
HCL 3N		33050-U SUPELCO	500	Liter		0		0		0		0		0		0	######	0.04		0
Syringe			3	Piece	1	3	1	3	1	3	1	3	1	3	1	3	1	3	1	3
Container			0.5	Piece	1	0.5	1	0.5	1	0.5	1	0.5	1	0.5	1	0.5	1	0.5	1	0.5
Transfer syringe 1ml			3	Piece	1	3	1	3	1	3	1	3	1	3	1	3	1	3	1	3
Pretreatment Process						0		0		0		0		0		0		0		0
H2O			0.2	Liter	0.0135	0	0.0135	0		0		0		0		0	0.001	0	0.0001	0
ACN		SigmaAldrich 271004-2L Acetonitrile anhydrous, 99.8%	86	Liter		0		0		0	0.011	0.95		0		0	0.001	0.09	0.002	0.17
Cleaning Agent - for milk		Fairy	1.513	Liter		0		0		0		0	0.0113	0.02	0.0113	0.02	0.0113	0.02	0.0113	0.02
Rinse Agent - for milk		MSc rinse concentrate	0.76	Liter		0		0		0		0	0.0113	0.01	0.0113	0.01	0.0113	0.01	0.0113	0.01
NaOH 0.1M				Liter		0	?	###		0		0		0		0	0.001	0		0
Solvent 5				Liter		0	?	###		0		0		0		0		0		0
Solvent 6				Liter		0	?	###		0		0		0		0		0		0
Filter PTFE			1.9	Piece	1	1.9		0	1	1.9	1	1.9		0	1	1.9		0		0
Captiva - for milk			2.7	Piece		0		0		0		0	1	2.7		0		0		0
SPE - for milk		Bond Elut Plexa	2.3	Piece		0		0		0		0		0		0	1	2.3		0
SPE - for milk		Elut NH2	2.13	Piece		0		0		0		0		0		0		0	1	2.13
SPE - for nuts			2.5	Piece		0	1	2.5		0		0		0		0		0		0
Detection Process																				
Binding Buffer		PBS1x pH 7.4 +1mM MgCl2	0.281	Liter	0.0011	0	0.0011	0	0.0011	0	0.0011	0	0.001	0	0.001	0	0.0011	0	0.0011	0
Standard Buffer		Phosphate buffer 0.1M containing 5mM of MgCL2	2.16	Liter	0.0001	0	0.0001	0	0.0001	0	0.0001	0	1E-04	0	1E-04	0	0.0001	0	0.0001	0
Regeneration Solution		MeOH+T10E1	1.51	Liter	0.0002	0	0.0002	0	0.0002	0	0.0002	0	2E-04	0	2E-04	0	0.0002	0	0.0002	0
Wash Solution		10mM NaCl+ 5mM Tris pH 7.4	0.111	Liter	0.001	0	0.001	0	0.001	0	0.001	0	0.001	0	0.001	0	0.001	0	0.001	0
Bio-Sensor Chip																				
PCB		Custom made	1.2	Piece	0.0333	0.04	0.0333	0	0.0333	0.04	0.0333	0.04	0.0333	0.04	0.0333	0.04	0.0333	0.04	0.0333	0.04
Chip		for 1M chips	20	Piece	0.0333	0.67	0.0333	0.7	0.0333	0.67	0.0333	0.67	0.0333	0.67	0.0333	0.67	0.0333	0.67	0.0333	0.67
Coating																				
Antifouling layer			2	Piece	0.0333	0.07	0.0333	0.1	0.0333	0.07	0.0333	0.07	0.0333	0.07	0.0333	0.07	0.0333	0.07	0.0333	0.07
Anchoring layer		homogenouse anchoring layer	3.3	Piece	0.0333	0.11	0.0333	0.1	0.0333	0.11	0.0333	0.11	0.0333	0.11	0.0333	0.11	0.0333	0.11	0.0333	0.11
Aptamers			0.1	Piece	0.0333	0	0.0333	0	0.0333	0	0.0333	0	0.0333	0	0.0333	0	0.0333	0	0.0333	0
Laser immobilisation			2	Piece	0.0333	0.07	0.0333	0.1	0.0333	0.07	0.0333	0.07	0.0333	0.07	0.0333	0.07	0.0333	0.07	0.0333	0.07
Silane functionalization and immobilisation			0.3	Piece	0.0333	0.01	0.0333	0	0.0333	0.01	0.0333	0.01	0.0333	0.01	0.0333	0.01	0.0333	0.01	0.0333	0.01
Total Cost Consumables/Test						**14**		**14**		**9.7**		**12**		**10**		**9.5**		**9.9**		**11**



Innovation process and technology translation (2)

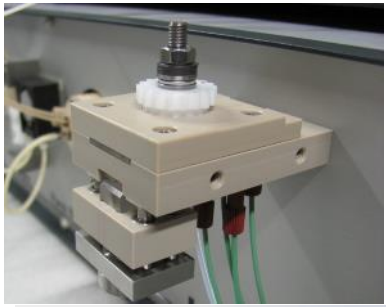
Use of advanced stable and selective immobilization methods for and aptamer deposition methods

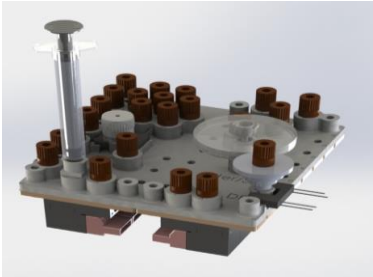
Regeneration of bio-recognition elements for re-use of the biosensor (30x)

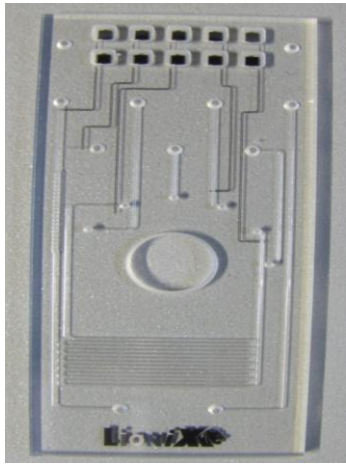


Innovative microfluidic technology for:

- **Management of liquid processing**
- **Miniaturization of pretreatment unit**
- **Effective interface with the photonic chip**







Manufacturability

Cost

Innovation process and technology translation (3)

Hybrid photonic integration of InP components on TriPleX platform	LionIX, ICCS, SAXION
Signal processing algorithms and techniques, and user interfaces	csem, ICCS
Immobilization and regeneration of biorecognition elements on-chip	surfix, BRFAA, ICCS, WAGENINGEN, UPVD
Microfluidic technologies for the pretreatment unit and the microfluidic chip	LionIX, csem
Validation in laboratory and in relevant industrial environment	IRTA, COVAP

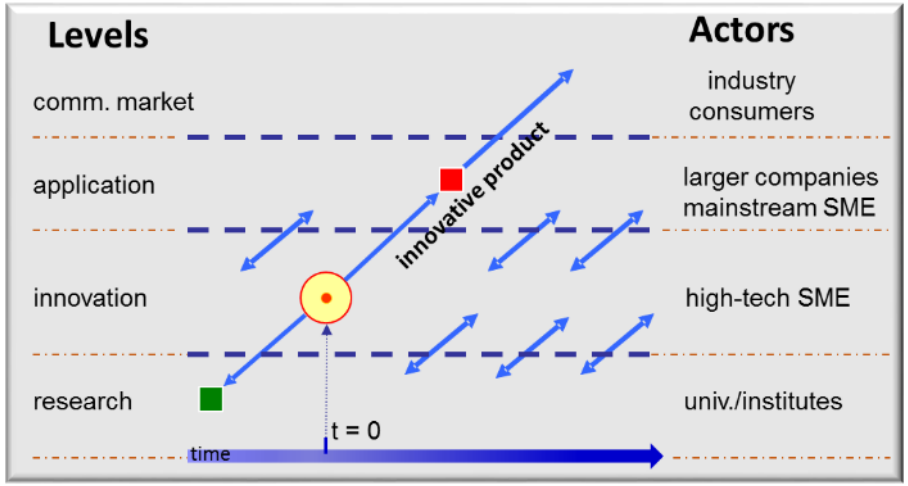
Lead

Support

Validated photonics, signal processing, aptamer immobilization/regeneration and microfluidics to be ready and available at the end of the project

TRL 5-6 at the end

Road to exploitation (1)



- **Research-to-application:** not a linear **trajectory**
- **Innovation focus on enabling technology level**
- **R&D:**
  - **development of concepts and building blocks**
- **Product:**
  - **Focus on market & requirements**
  - **Combination of applicable concepts and building blocks**

Levels	Actors	BioFQs	Candidates
Market	Industry/consumers	COVAP, IRTA	FrieslandCampina (NL}, Barry-Calebaud (Sp}, other
Application	Technology user	**Not present**	FOSS, AlfaLaval, joint-venture?, Lateral Flow Test companies, other
Innovation	High Tech SMEs and industrial institutes	LioniX, CSEM, Surfix	Cambio (UK), Eurogentec (B) (aptamers), Scienion (spotting, G), possible spin-out(s) from ICCS/NTUA, other
Research	Universities / institutes	ICCS/NTUA, UPVD, BRFAA, WU, Saxion	Research/institutes in

Road to exploitation (2)

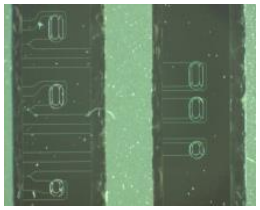
A technology user with direct access to the market is not present in the consortium

What are the opportunities:

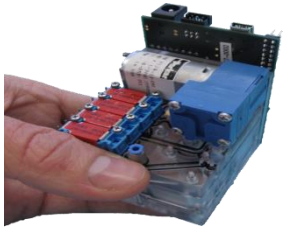
- Direct exploitation by setting up a joint-venture: very risky, venture capital required
- Licensing or partnership with existing companies outside consortium
- Funding: Fast Track to Innovation, Innovation Action (2017), Access To Finance (SME)

Intellectual property

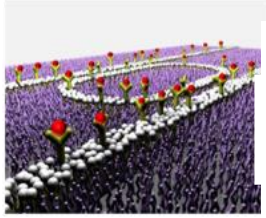
- TriPleX (LioniX)

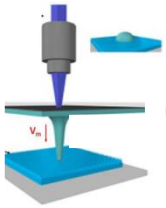


- Microfluidics (CSEM)



- Immobilzation (Surfix, ICCS/NTUA)





- Further IP can be generated in regeneration, processing, user interfaces/services
- Exploitation/licensing agreements depending on business case(s)

Manufacturability

- Prototype system units by consortium partners
- Product in collaboration:
  - instrumentation: industrial company (OEM or OEM supplier)
  - with new, complementary (SME) partners :

  e.g. Cambio (UK), Eurogentec (B) (aptamers), Scienion (spotting, G), spin-out(s) ICCS/NTUA, ...

Road to exploitation (3)

Exploitation of individual partner results, including spin-off applications

LioniX : TriPleX MRR and microfluidic valving concept

- Applications outside food, e.g. Companion Diagnostics, Drug Screening platform, Bioprocessing control
- In-line refractometry sensor

CSEM: microfluidic pump

- Applications outside food

Surfix and ICCS/NTUA: immobilization of recognition sites

- Applications outside food, e.g. Companion Diagnostics, Drug Screening platform, Bioprocessing control

Competition

- Many ventures are focussing on medical (genetic) diagnostics
  - USA - venture capital, government funding: mBioDiagnostics (Neogen), Seattle Sensors, BioDetection, …
- Many generic platforms – conversion from medical to food, genetic to chemical
- Lateral Flow Test companies – collaboration with generic platform supplier (Neogen)

Roadmap / exploitation trajectory

Prototype development
Business case I, II, ...
Innovation Action, SME Instr.

- Start-up or find industrial partner
- Involve users from the beginning

YEAR0: 1-2 MEURO

Product development
Define specific business case
Venture Capital/private, FTI,

- Continuous field tests at users'
- Find commercial partners for other business cases

YEAR2/3: 2-3 MEURO

Commercial exploitation
Business case I, II, ...
Venture Capital/private, ...

YEAR4/5: 3-4 MEURO

Road to exploitation (4)

Industrial Advisory Board:

- Prof. Dr. Dominik Rabus, ex-Bürkert (D) & Festo (D), measurement and control systems for liquids and gases, Technology Portfolio Manager
- Patric Salomon, enablingMNT (D) offering services in micro/nanotechnology, Partner, Partner in EU FoodMicroSystems project
- Dr. Antonio Logrieco, Inst. Of Sciences of Food Production, CNR-ISPA (It), a centre of excellence to improve safety and quality of agro-food products, Director; Coordinator of EU MycroRed project (2009-13)
- Dr. Willem Haasnoot, RIKILT (NL) National Reference Lab, Senior Researcher, dept. Assay Development
- Marcel de Vreeze, NEN (NL), non-profit NL Standardization Agency, Consultant, Dept. AgroFood & Consumers
- Ids Klompmaker, FrieslandCampina (NL), multinational dairy company, Coordinators Sensors and Process Technology
- Dr. Katerina Mastovska, Covance Laboratories (USA), commercial analysis and consultancy services, Associate Scientific Director, Nutritional Chemistry and Food Safety
- Dr. Belen Ripoll, Quality Manager, Unió Corporació Alimentària, a big cooperative in Spain for nuts, olive oil, wine and fruits

- Demonstrator system for PoN must-have!
- CEN/Standards: start at national level (NEN) → dissemination tool
- Multi-analyte!
- Antibiotics/pesticides: (too) many!?
- Spin-off: nutrients, e.g. Vit. B12

Contact

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