# Electrochemical sensors for animal welfare

Ilaria Sorrentino, Claire Verplanck, Yohann R J Thomas Monday, Amélie Revaux

# Electrochemical Sensors for Animal Welfare

*Ilaria Sorrentino, Claire Verplanck and Yohann R.J. Thomas*

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Monday, September 11 2023

BIOMEDICAL SENSORS & DIAGNOSTICS I

# CEA : a public research organization

**4** *MAIN RESEARCH AREAS*

**Defense and national security**

**Nuclear and renewable energy**

**Fundamental research**

**Technology research**

CEA-Leti Grenoble

*CEA-Leti is one of the 3 technological research institutes of CEA*

# Our mission: transfer technology to industry

*Connect academic research directly to industrial R&D to catalyze innovation and shorten time-to-market for businesses.*

**COMMERCIALIZATION**

TRL 7-9

***Technology research***

Scaleup

TRL 3-7

Technology maturation

Proof of concept

**SCIENTIFIC IDEA**

TRL 1-3

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Derwent™
A Clarivate Analytics company

Since **1967**

**2,000** people

**Patents:**
- > 3,000 in portfolio
- 40**%** under license agreement

**Startups:**
- 68 created for 20 years (75% in activity)
- 3500 jobs created

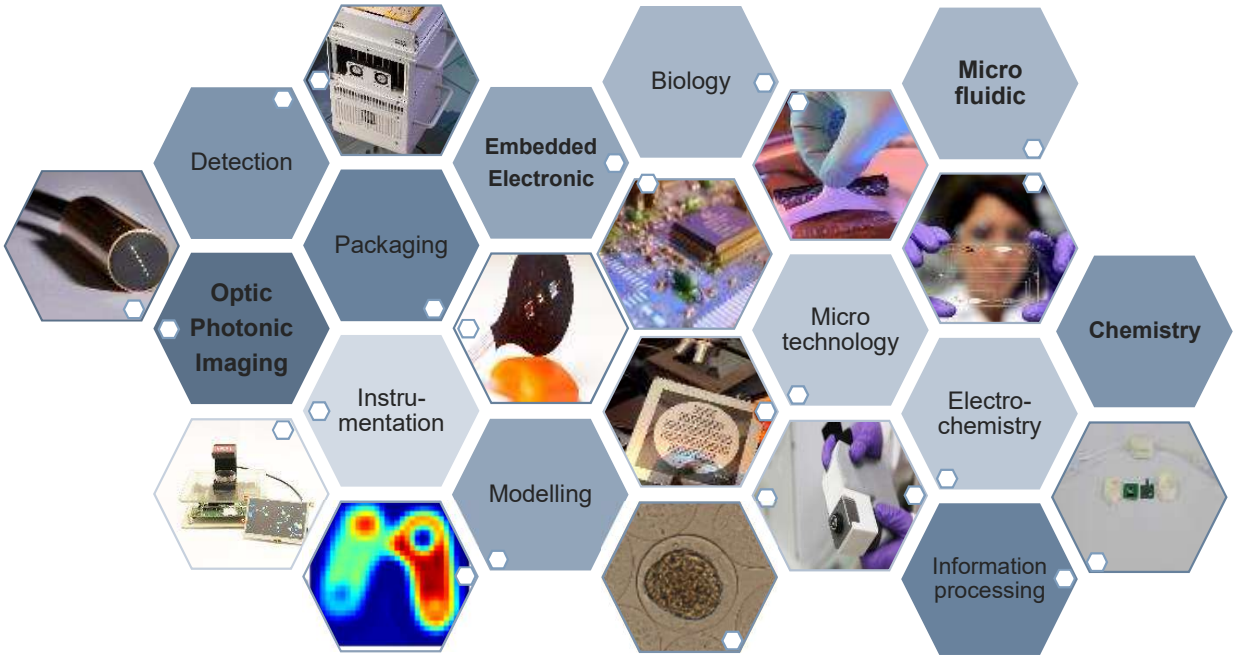
**Cleanrooms:**
- 500 state-of-the-art equipment in 200 & 300 m²
- 10 000 square meters cleanroom

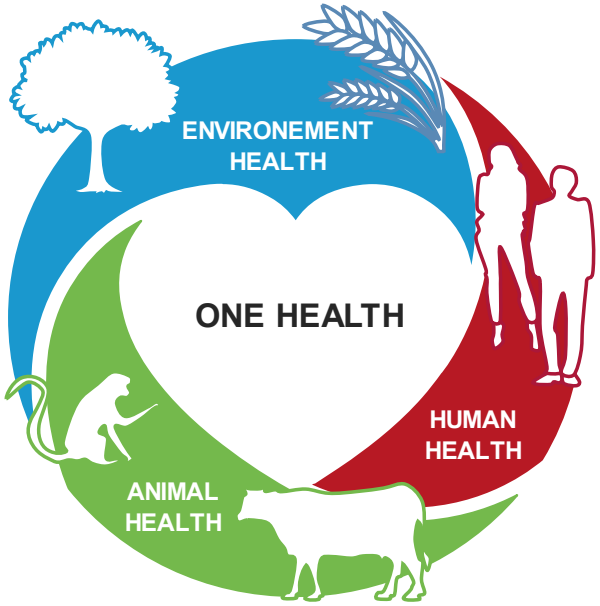
**Budget:**
- 315 M€
- 85% from R&D contracts

# Leti health
## Mission and Expertise

*Address societal needs in the fields of biology, healthcare, environment and security*

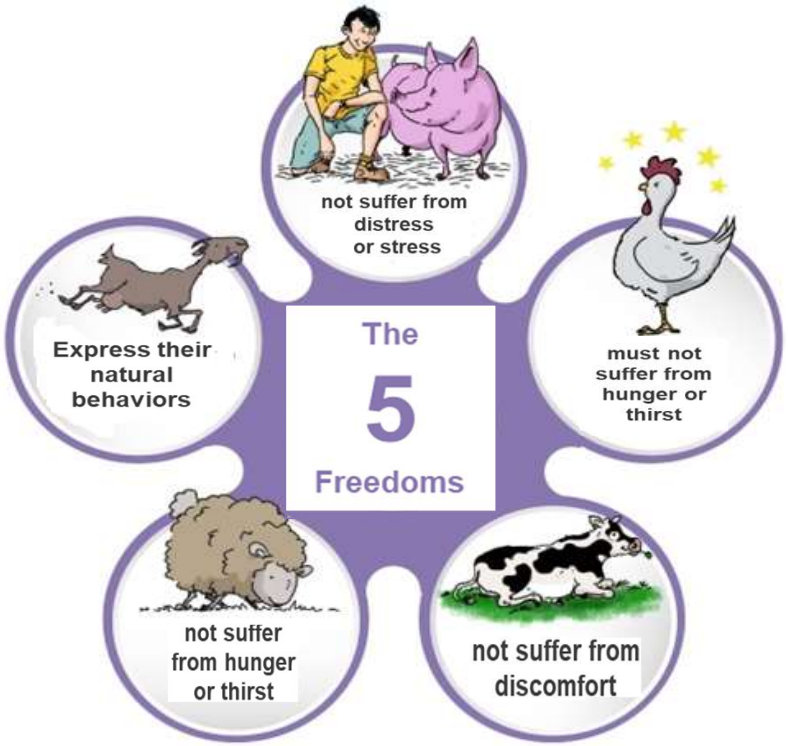
*Create & develop innovative solutions for our industrial partners*




*Multidisciplinary teams - One stop shop for completely integrated systems*

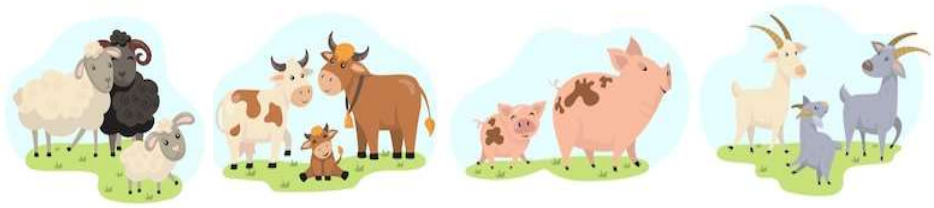
# WAIT4: Welfare Artificial Intelligence and new Technologies for Tracking key indicator Traits in animal facing challenges of the agro-ecological Transition




*Source: Website of the Chamber of Agriculture for Animal Welfare (France)*

To be proactive for **animal welfare** (AW), we need to understand their **point of view** and **needs**

**Project goal:** Increasing research-based capacities to quantify and characterize AW in the **agro-ecological (AE)** perspective and revising **farming practices** and **decision-making tools** by considering interactions between the animal and its environment

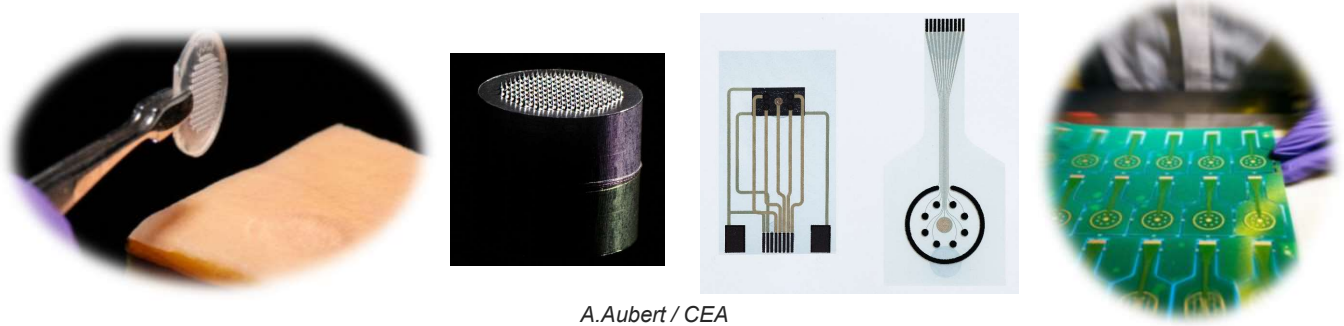


*https://bien-etre-animal.chambres-agriculture.fr*

# WAIT4: Welfare Artificial Intelligence and new Technologies for Tracking key indicator Traits in animal facing challenges of the agro-ecological Transition

- **WP1:** Analysis of interactions between the animal and its environment, using artificial intelligence
- **WP2:** Monitoring physiology for acute and mid-term adaptation to stress factors
- **WP3:** Determining the impact of agro-ecological transition on animal welfare
- **WP4:** Dissemination

**CEA Objective:** Coupling hydrogel microneedles (MN) to electrochemical sensors in order to develop minimally invasive connected devices for real-time monitoring in animals Interstitial Fluid (ISF)



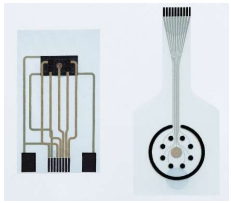
*A.Aubert / CEA*

# WAIT4: Welfare Artificial Intelligence and new Technologies for Tracking key indicator Traits in animal facing challenges of the agro-ecological Transition

## WP2- New sensors of animal physiology

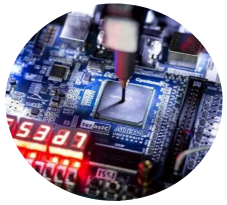
### Biomaterials

- Hydrogel formulation
- MN design
- Morphological and structural characterization



### Electrochemistry

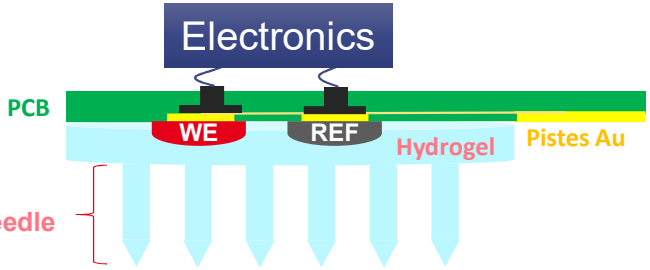
- Formulation ISE ($Na^+$, pH) and REF
- Electrodes Design
- Calibration test on simple model solutions, complex and ISF sampling



### Electronics

- Set up communication protocol
- collect data
- send data

## Integration

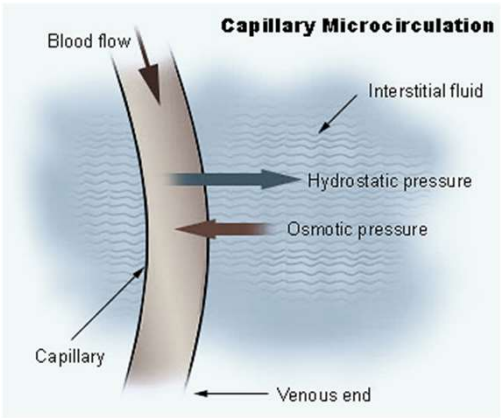



- ✓ Sampling ISF (MN)
- ✓ Measuring in real time (Electrochemical sensors)
- ✓ Reporting data (Electronics)

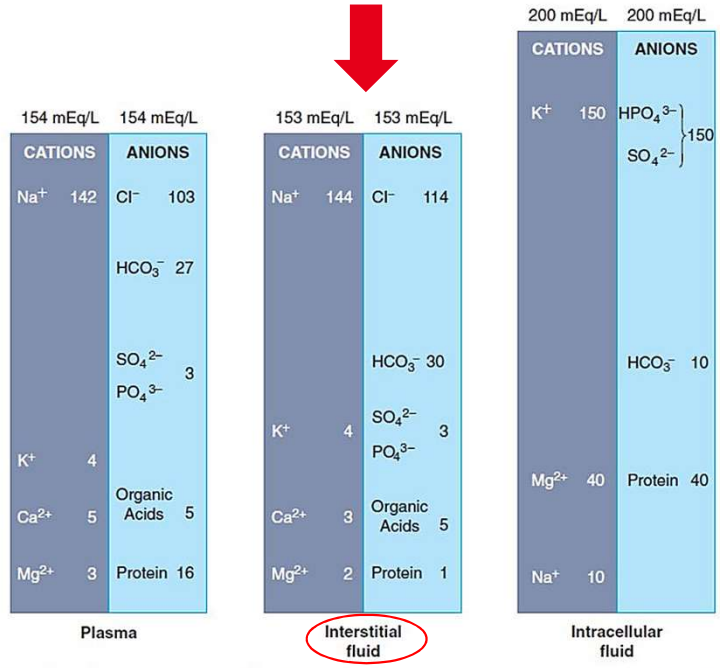
# WAIT4: Target matrix

Multi-parameter electrochemical platform for monitoring physiological variables of interest like potassium and pH in animals Interstitial Fluid (ISF)

ISF is a **complex fuid** containing amino acids, sugars, fatty acids, coenzymes, hormones, neurotransmitters, salts and waste products from the cells.




ISF is present between cells and represents around 80% of extracellular fluids (ECF).




Chemical composition of body fluid compartments. [Reproduced with permission from Brunicardi FC, Andersen DK, Billiar TR, et al (eds): *Schwartz's Principles of Surgery*, 10th ed. McGraw-Hill, Inc., 2015. Fig 3-2, p. 67.]

Source : Tintinallis Emergency Medicine A Comprehensive Study Guide_8th

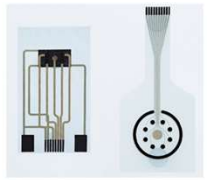
# WAIT4: Physiological variables of interest

**$Na^+$ and pH**: concentration range (sensitivity & accuracy)

- **pH**: variation between 7.35 and 7.50 in blood (in cows)
  Desired sensitivity range 0.03 to 0.05 pH point
  - ΔE over full range < 9 mV
  - Sensitivity range < 3 mV
- **$Na^+$** – cow and sow in plasma: 130-145 mmol/L
  Desired sensitivity range < 4-5 mmol/L
  - ΔE over full range < 3 mV
  - Sensitivity range < 1 mV

Very low values at the limit of possible detection

# Electrochemical sensors



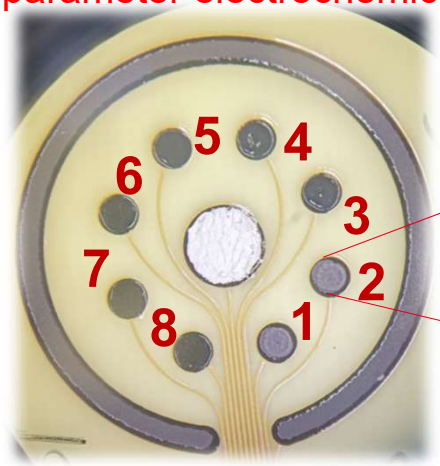
## ADVANTAGES

- **Multiparametric measurements with one platform**
  - **Ions:** $Na^+$, $K^+$, $Ca^{2+}$, $Mg^{2+}$, pH, $NH_4^+$, $NO_3^-$, $Cl^-$ …
  - **Metabolites:** glucose, lactate, NO…
  - **Conductivity**
- **Complex fluids**
  - **Biological:** sweat, blood, plasma, …
  - **Aqueous:** Drinking water, rivers, swimming pools, …
  - **Industrial**
- **Adaptable to different substrates**
  - **Materials:** Silicon, polymers, papers, fabrics, …
  - **Properties:** Flexible, stretchable, wearable, …
  - **Fabrication processes adapted, collective**
- **Other advantages**
  - **Continuous measurements**
  - **Miniaturization**
  - **Biocompatibility**
  - **Cytotoxicity**
  - **Platform adapted to the application**

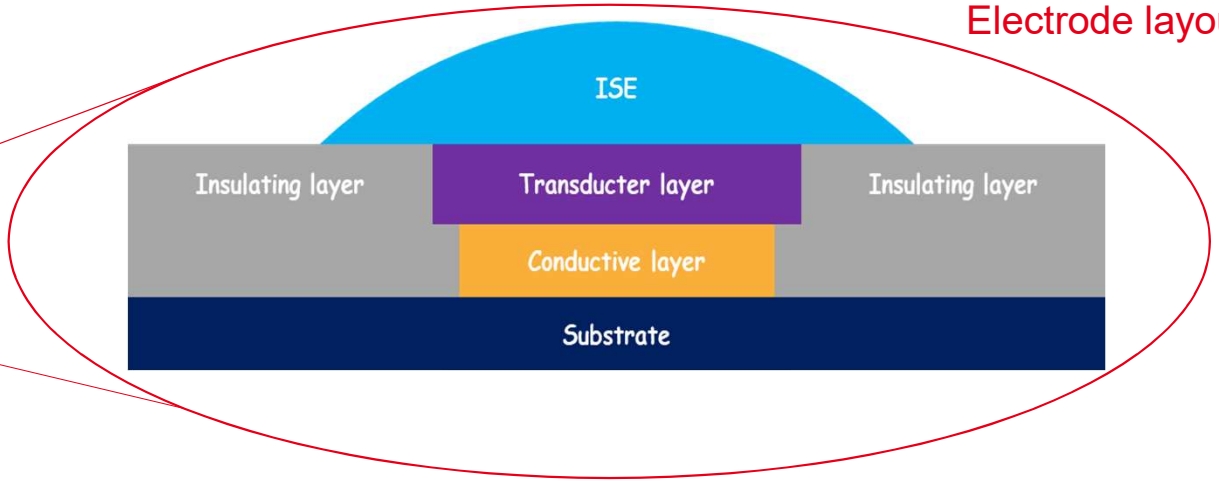
# WAIT4: Electrochemical Sensors

Multi-parameter electrochemical platform for monitoring physiological variables of interest like sodium, potassium and pH in animals Interstitial Fluid (ISF)

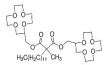
Multi-parameter electrochemical platform



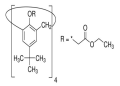

Electrode layout




- **$Na^+$:**
  - Ion Selective Electrode (ISE) :3 ionophores



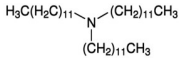
*Ionophore Na VI*



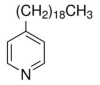
*Ionophore Na X*

*Ionophore Na IV*

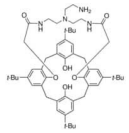
- **pH :**
  - Iridium Oxide: IrOx
  - ISE: 3 ionophores pH



*Hydrogen Ionophore I*



*Hydrogen Ionophore II*



*Hydrogen Ionophore V*

# Biosensor development - Experimental section

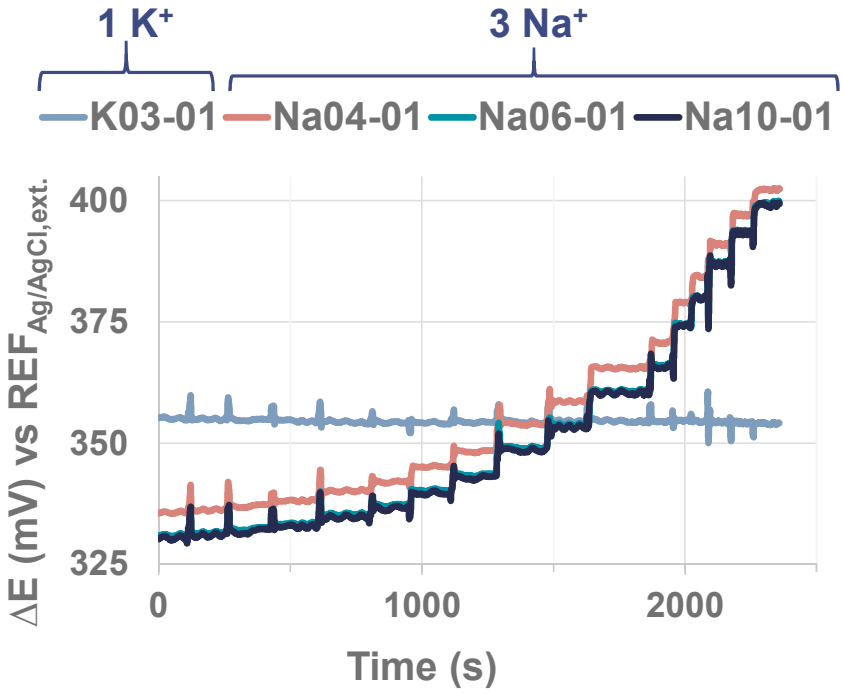
1. Sensing $Na^+$ 


2. Sensing pH 

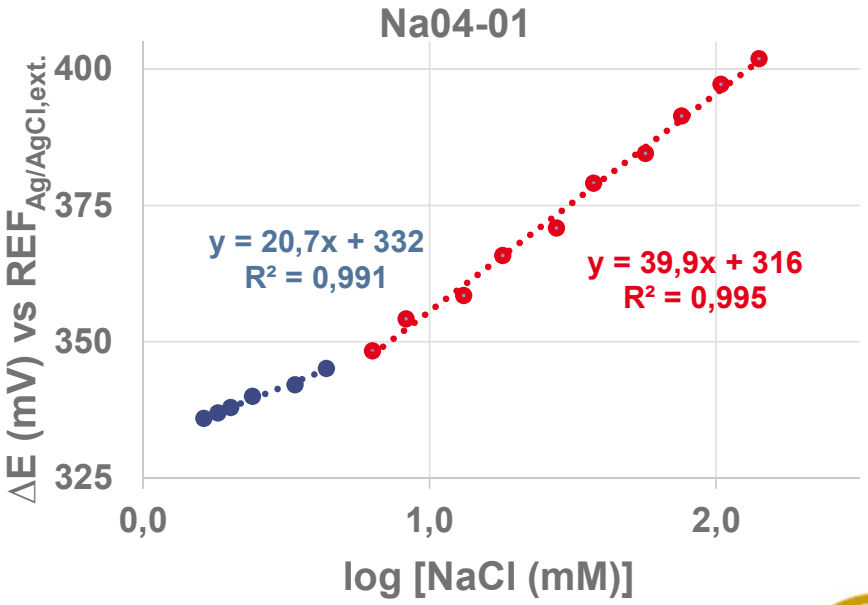

3. Multiparameter platform – Interference such as $K^+$ 

# 1. Sensing $Na^+$

Method of standard additions in complex buffer **pH = 7,4**



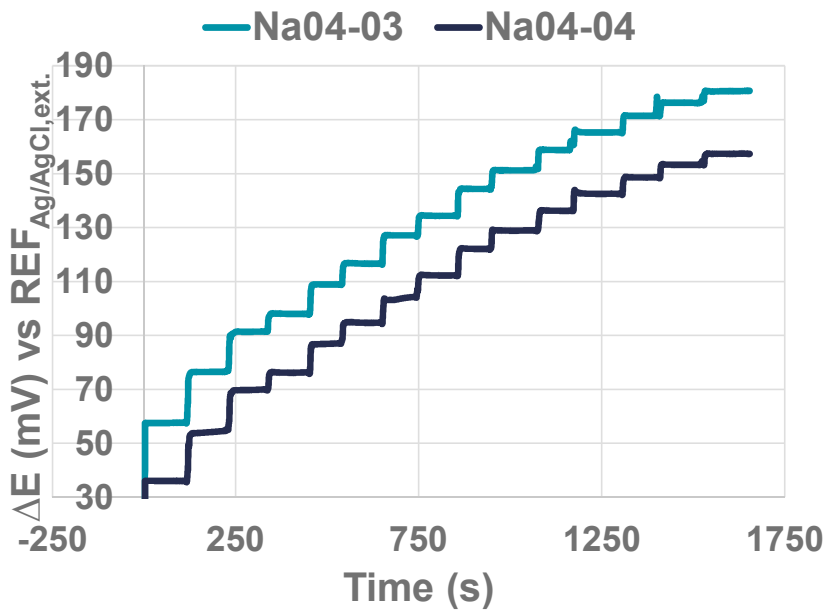

| ISE | Na04-01 |
|---|---|
| S (mV/dec) | 39,9 |
| $R^2$ | 0,995 |
| $E_0$ (mV) | 315,6 |

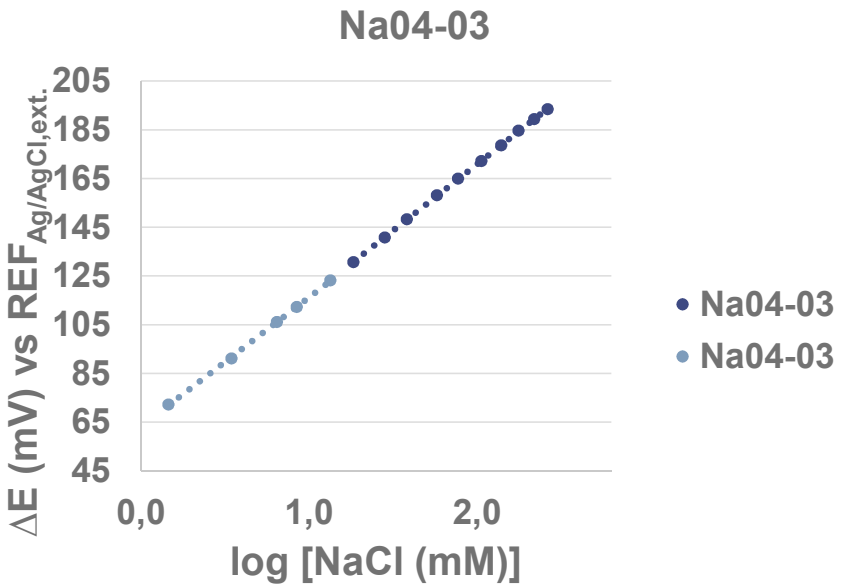



**Linearity between 6mM et 150mM**

# 1. Sensing $Na^+$ - New formulations

Method of standard additions in complex buffer **pH = 7,4**




| ISE | Na04-03 | Na04-04 |
|---|---|---|
| S (mV/dec) | 55,0 | 54,4 |
| $R^2$ | 0,999 | 0,999 |
| $E_0$ (mV) | 46,8 | 27,1 |

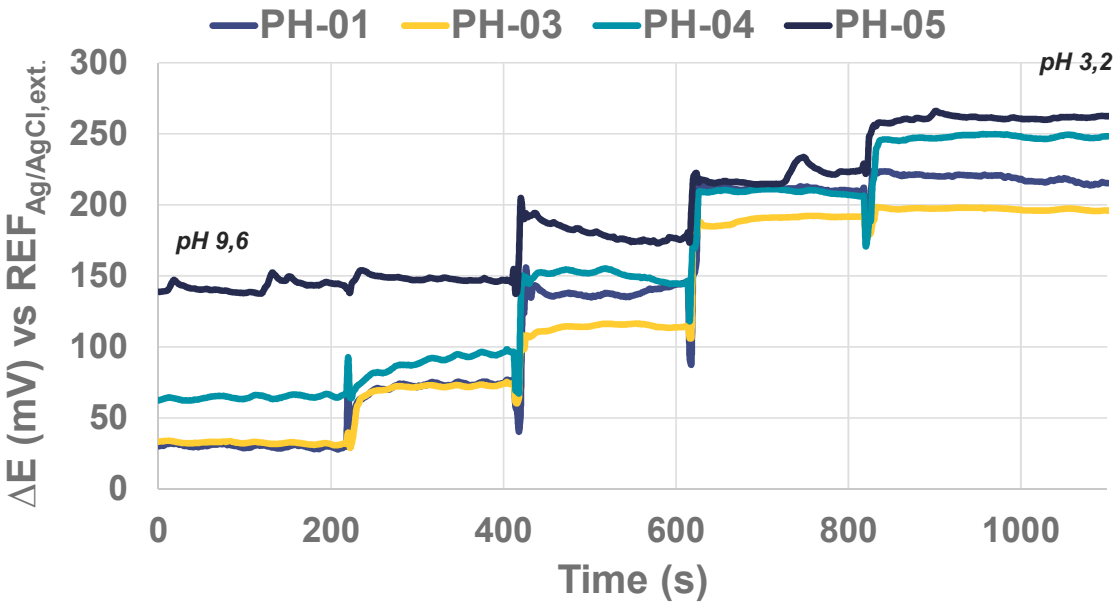
**linearity over the whole range (1.5mM-260mM)**

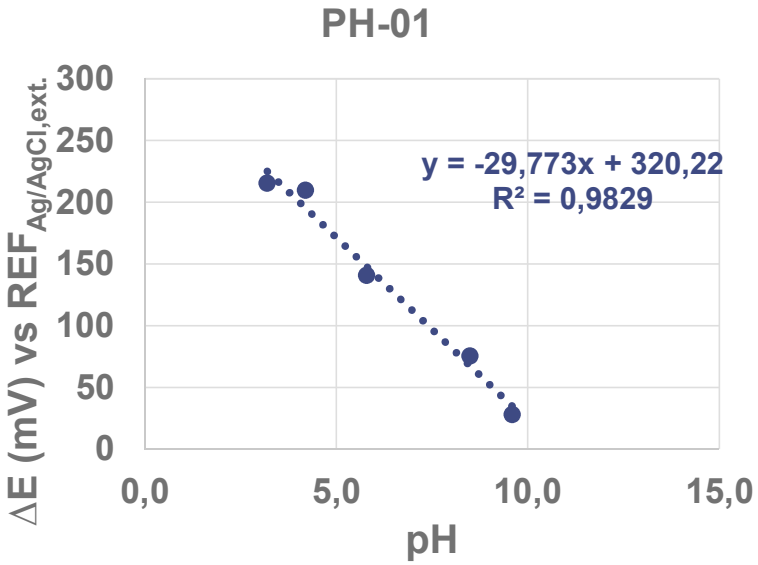
The new ISE $Na^+$ formulations show **improved sensitivity** and a **lower LOD**

# 2. Sensing pH

Method of standard additions in complex buffer

Range pH:**9-8-5-4-3 (+Aj Lactic acid)**



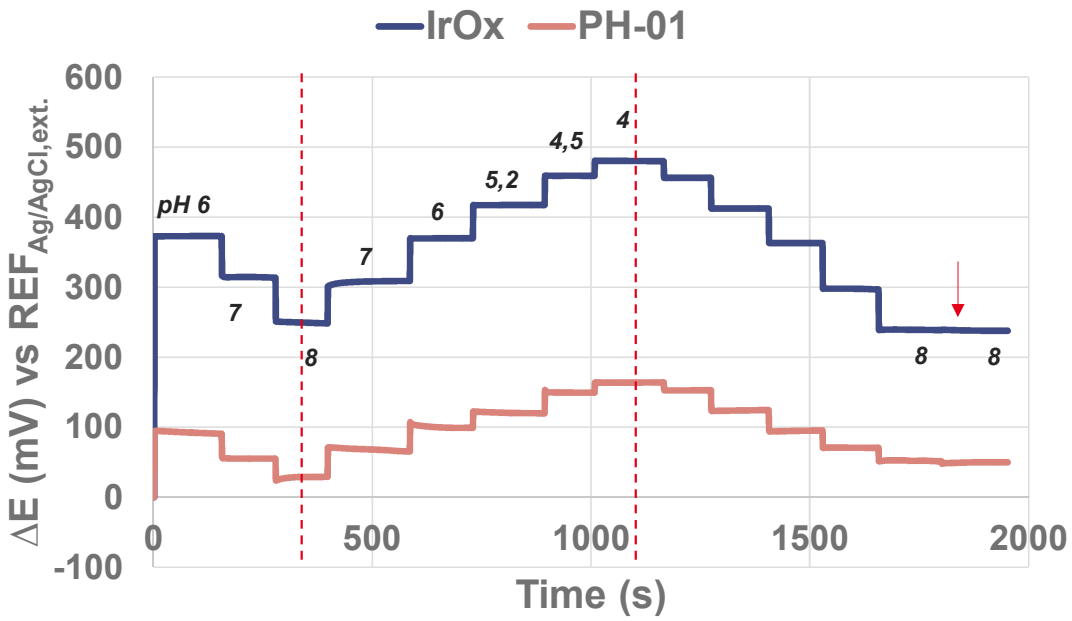



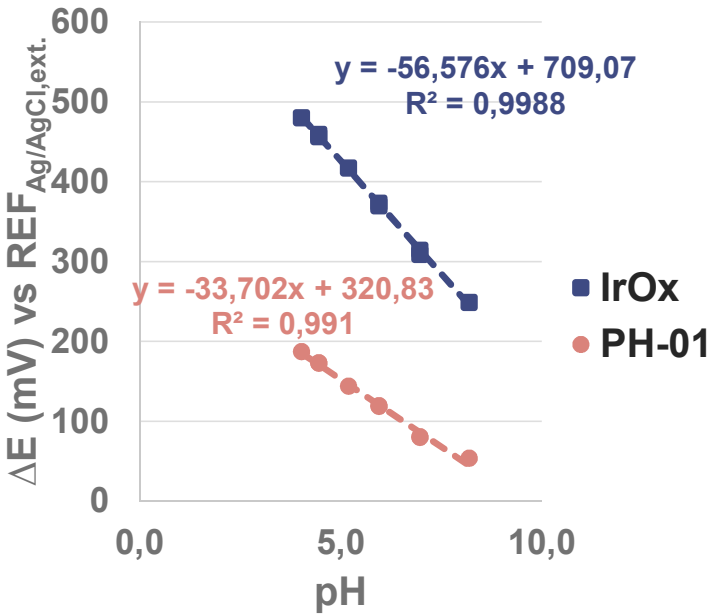

| ISE | PH-01 |
|---|---|
| S (mV/dec) | -29,8 |
| $R^2$ | 0,983 |
| $E_0$ (mV) | 320,2 |

**Not yet as sensitive as expected, needs to be optimised**

# 2. Sensing pH

Method of standard additions in complex buffer  Range pH:4->8 **(+Aj Lactic acid)**



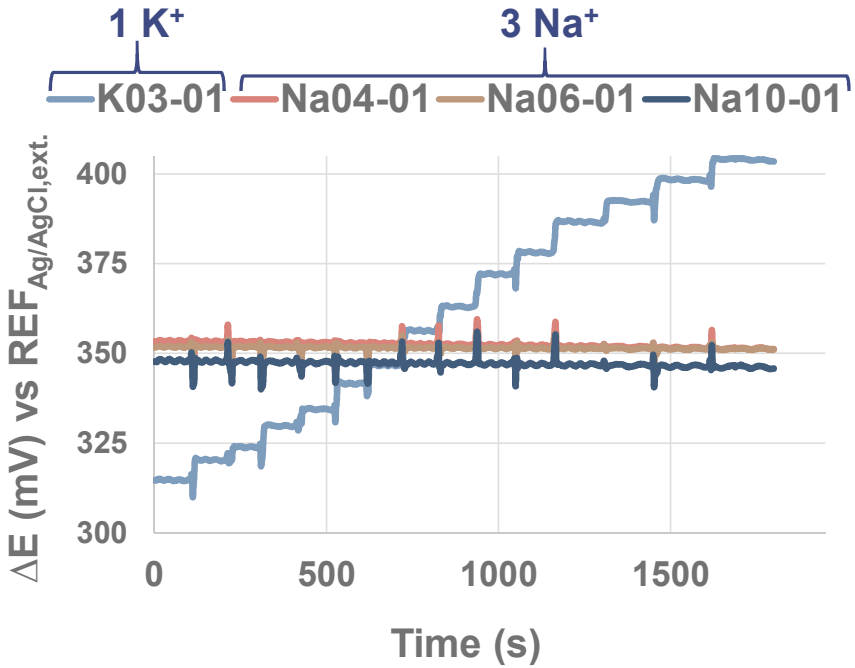

| ISE | PH-01 | IrOx |
|---|---|---|
| S (mV/dec) | -33,7 | -56,6 |
| $R^2$ | 0,991 | 0,999 |
| $E_0$ (mV) | 320,2 | 709,4 |

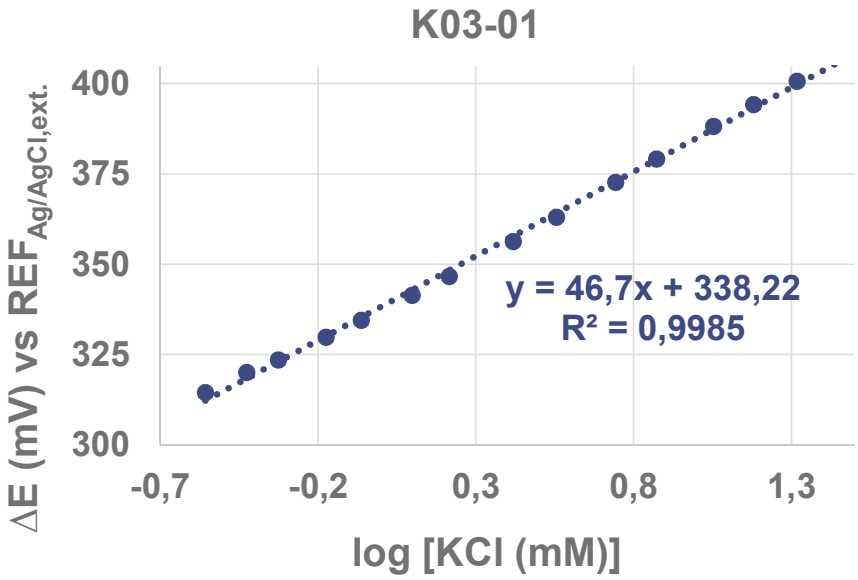
Comparing pH sensors IrOx and ISE (PH-01)

**IrOx** shows **higher sensitivity** and **stability** than ISE under the same conditions.

# 3. Interference - Sensing K+

Method of standard additions in complex buffer **pH = 7,4**

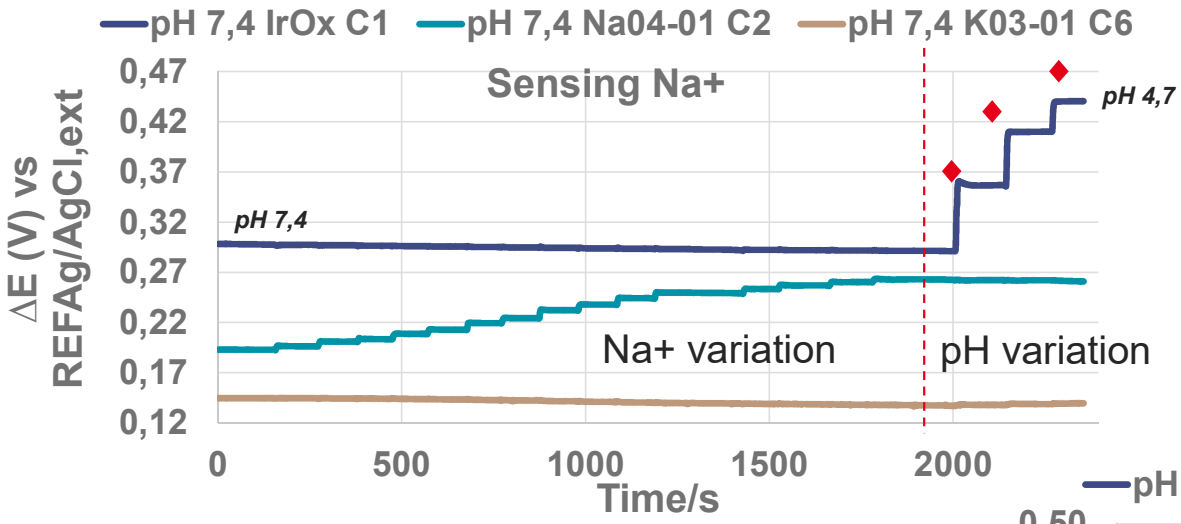


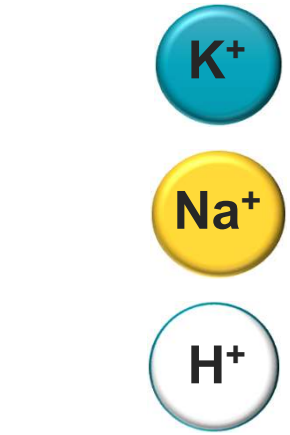




| ISE | K03-01 |
|---|---|
| S (mV/dec) | 46,7 |
| $R^2$ | 0,998 |
| $E_0$ (mV) | 338,2 |

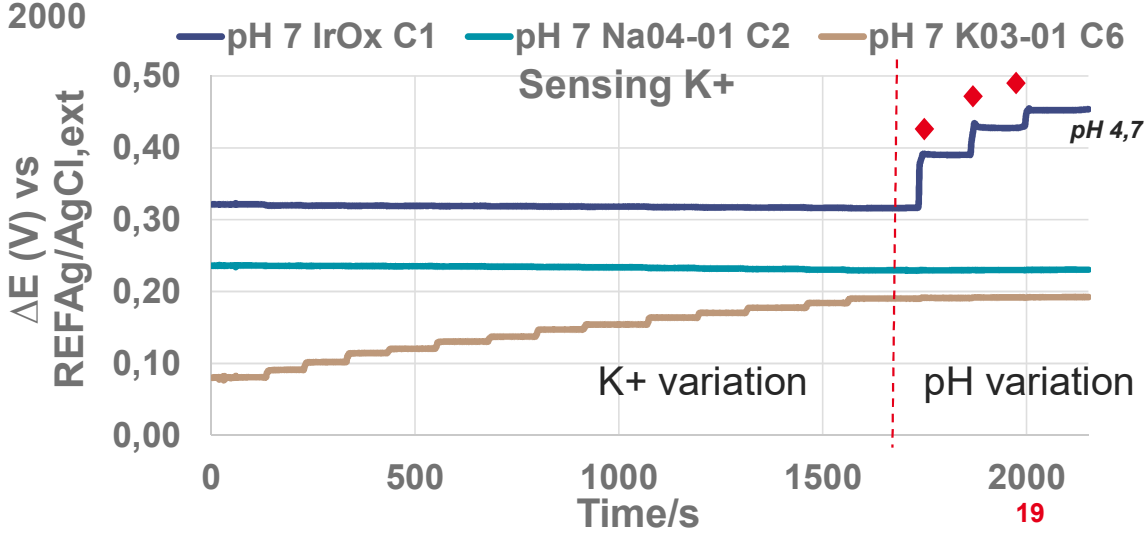
**Linearity between 0,2mM et 28mM**

# 2. Interference - Multiparameter platform



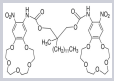

In multisensor experiments, **no interference was observed** between the detection of pH/$K^+$/$Na^+$



# Conclusions

In this study, we demonstrate that applying **our design** and **our fabrication strategy** of this platform enables the detection of **multiple parameters** involved in the **animal stress**

Multi-parameter electrochemical platform validation

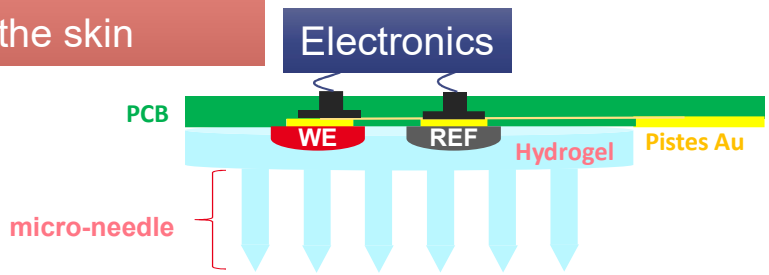
| Capteur | pH | $K^+$ | $Na^+$ |
|---|---|---|---|
| Formulation | IrOx |  *Ionophore K III* |  *Ionophore Na IV* |

These are the first promising results of **potentiometric electrochemical detection** of Na+, pH and K+ in **artificial ISFs**

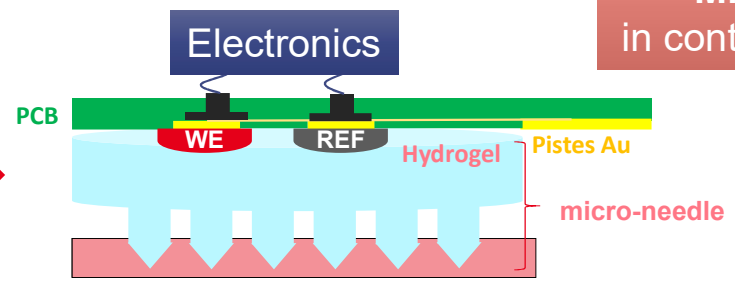
# Future perspectives / challenges

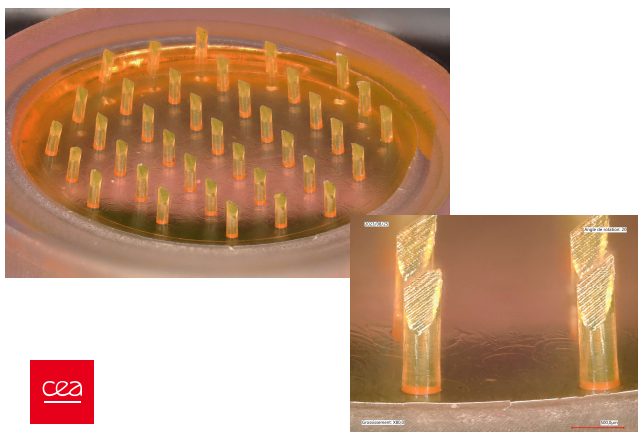

The next step will be the **integration** of **electrochemical platform** with the **micro-needle patches** to perform measurements of physiological parameters **minimally invasive** for the animals

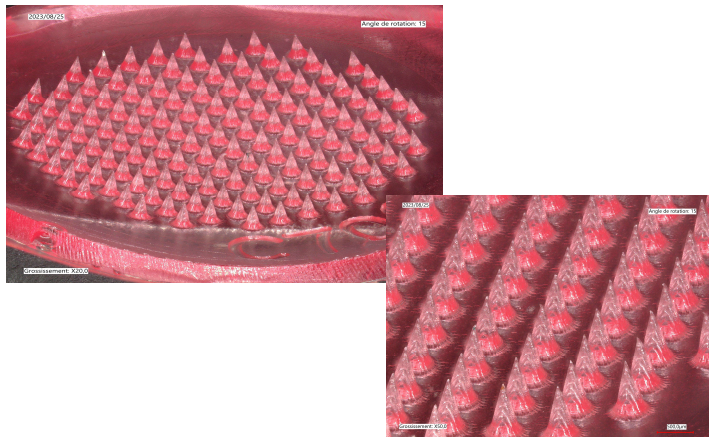
Insertion of the **dry MN patch** in/onto the skin

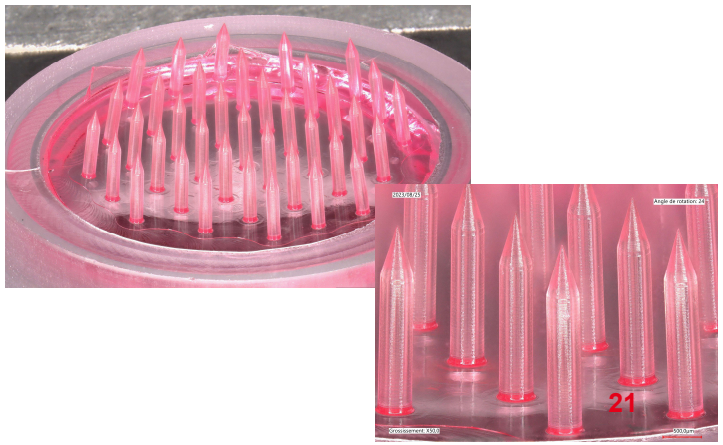



**Swelling** of the **MN patch** in contact with ISF








**Research Group**
**LMCD**

*Merci*

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**WAIT4 institutional consortium**

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# Q&A


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