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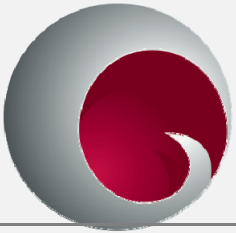
# Lactate Thick-Film Screen Printed Biosensor

**SAFEGARD:** Sensor Arrays for Environmental,  
Generic and Routine Detection



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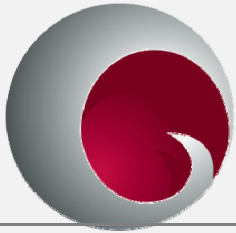
# Lactate Thick-Film Screen Printed Biosensor



# Why Lactate?

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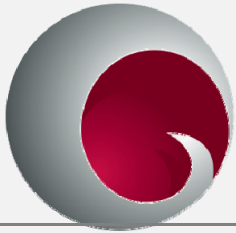
- early diagnosis of mastitis in dairy cows
- current enzymatic detection methods mainly use lactate oxidase or lactate dehydrogenase
- require lengthy and mechanically complex preparatory stages
- enzyme based sensors could provide rapid, simple alternative



# Requirements for Lactate Sensor

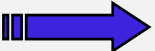
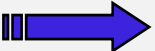
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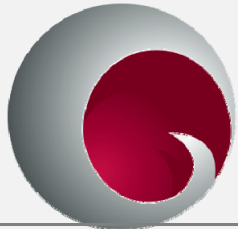
- simple, economical manufacturing process *e.g.* screen printing technology
- rapid, accurate measurements in highly humid environment with large temperature variations
- no sample pre-treatment or requirement for trained operators
- adaption to on-line analysis system



# Background to Lactate Sensor Development

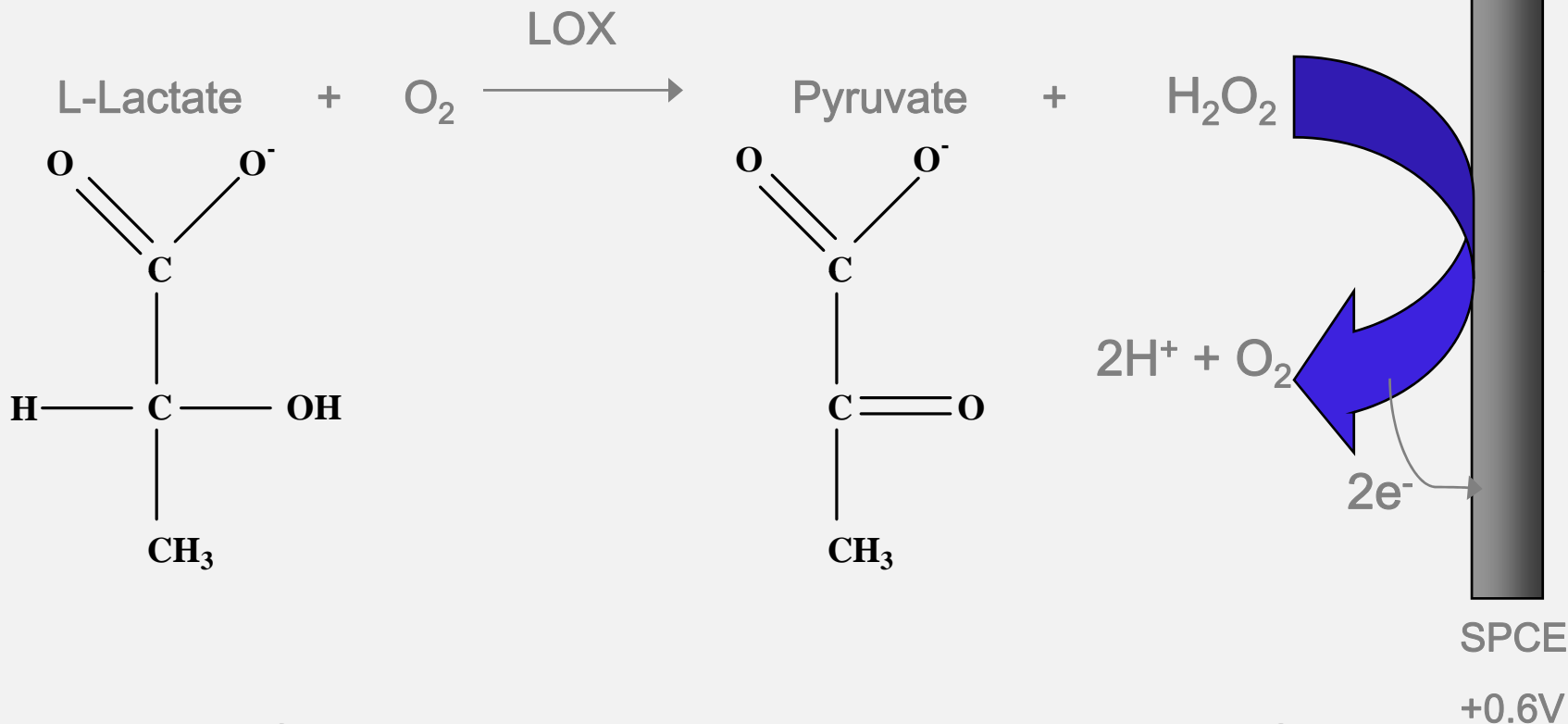
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- Hart *et al* (1996) - screen printed enzyme electrodes for lactate detection
- lactate oxidase / graphite mixture printed onto electroplated graphite pads
- based on electrochemical detection of hydrogen peroxide
- commercialisation  supplier of SPCEs  GEM / AET

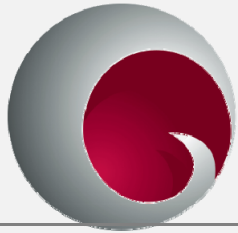


# Electrochemical Detection of Hydrogen Peroxide

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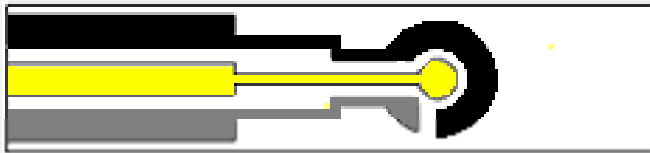
- oxidation of L-lactate to pyruvate and peroxide catalysed by LOX
- peroxide oxidation at SPCE polarised at +0.6V vs. Ag/AgCl
- electron flow at electrode surface proportional to L-lactate in sample



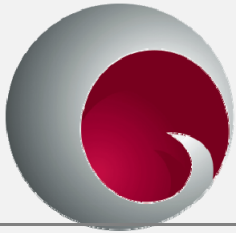
# Role of GEM in Sensor Development

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- electrode design
  - three electrode setup
  - working electrode potential more stable



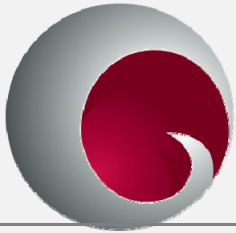
- batch manufacture simplified
  - cost reduction
  - high reproducibility
- smaller, well-defined geometric area



# Role of GEM in Sensor Development

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- ink formulations
  - mediated carbon
  - Ag/AgCl
  - dielectric
  
- manufacture of screen-printed base electrodes



# Role of AET in Sensor Development

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- enzyme formulations:
  - stability
  - immobilisation on electrode surface
  - optimisation of enzyme loading



# Role of AET in Sensor Development

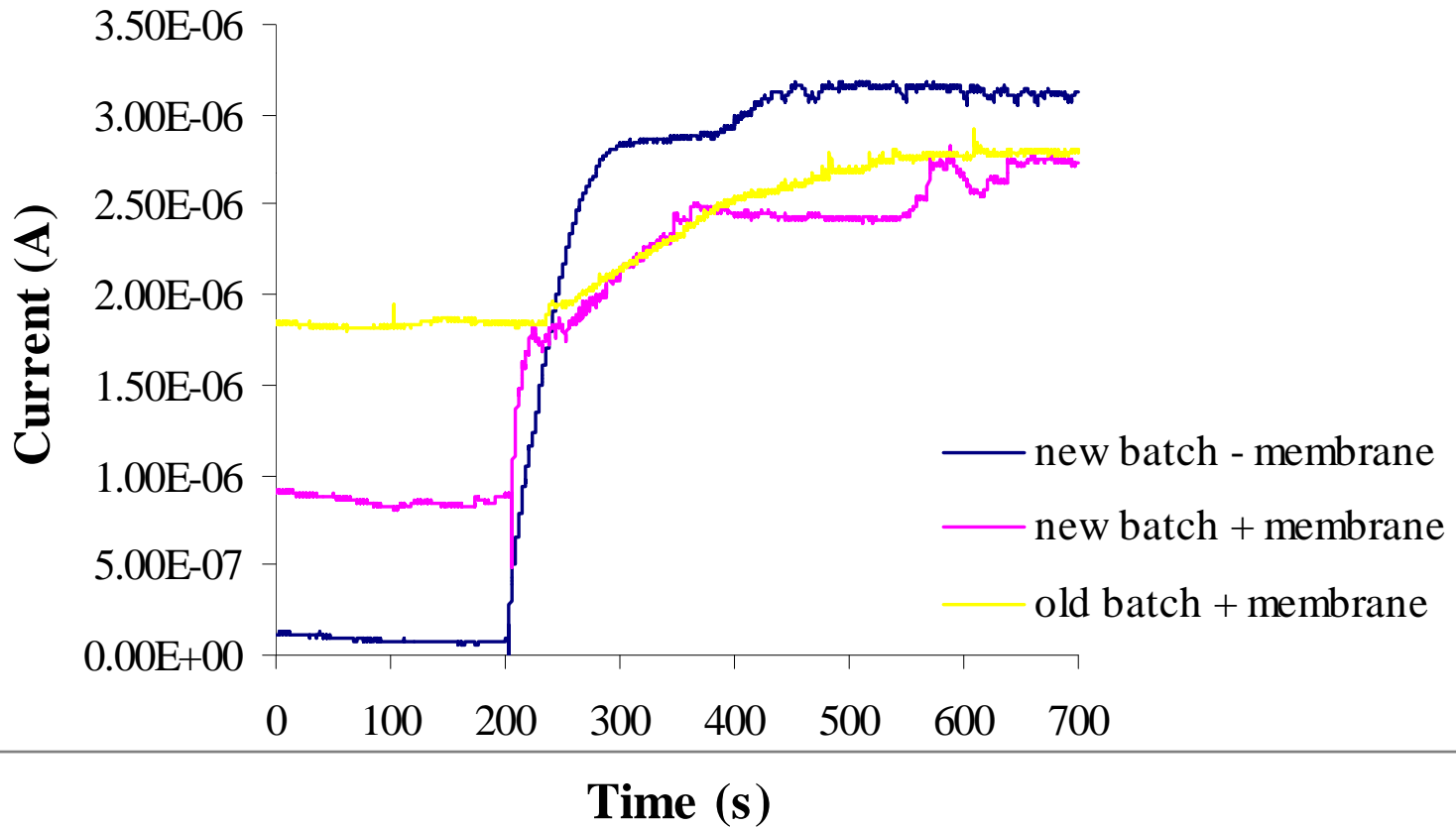
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- outer membrane – enzyme protection / diffusion limitations
  
- electrochemical measurement
  - working potential
  - signal-to-noise ratio



# Sensor Response with 1mM Lactate

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# Development

- optimisation of enzyme loading on sensor surface
- effect of temperature on enzyme performance
- evaluate different membrane formulations / methods of application
- short-term stability trials
- initiation of long-term stability trials
- evaluate requirement of inner-membrane



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# SAFEGARD: Sensor Arrays for Environmental, Generic and Routine Detection



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# Project Overview

- EU-funded as part of Fifth Framework “Quality of Life and Management of Living Resources” Program
- AET / GEM sub-contractors
- GEM supplier of screen-printed transducers



# Role of AET in SAFEGARD

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- enzyme stabilisation
  - mutant cholinesterases for higher sensitivity and specificity
  
- development of derivatised surfaces
  - towards specific immobilisation of enzymes on different electrode surfaces



# Importance of OP Detection

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- extensively used in modern agricultural techniques
- control of insect infestation and damage
- excess of active ingredients used as large amounts are washed off/absorbed onto surfaces contacted
- contamination and severe toxicological effects, particularly to CNS, arise as a result
- EU directives in force for monitoring / measurement



## Current Research Trends

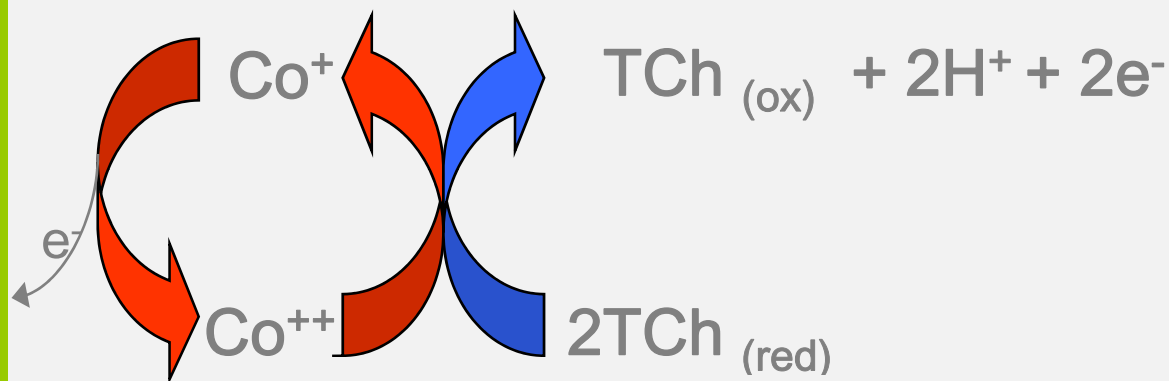
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- potentiometric/amperometric techniques largely employed
- based on utilisation of inhibition enzymes, usually cholinesterases, particularly acetylcholinesterase
- enzyme activity measurements before and after exposure to pesticides ⇒ difference in values directly related to pesticide concentration in sample
- allows for quantitative, sensitive detection systems



# Thiocholine Oxidation at CoPC-Modified SPCE

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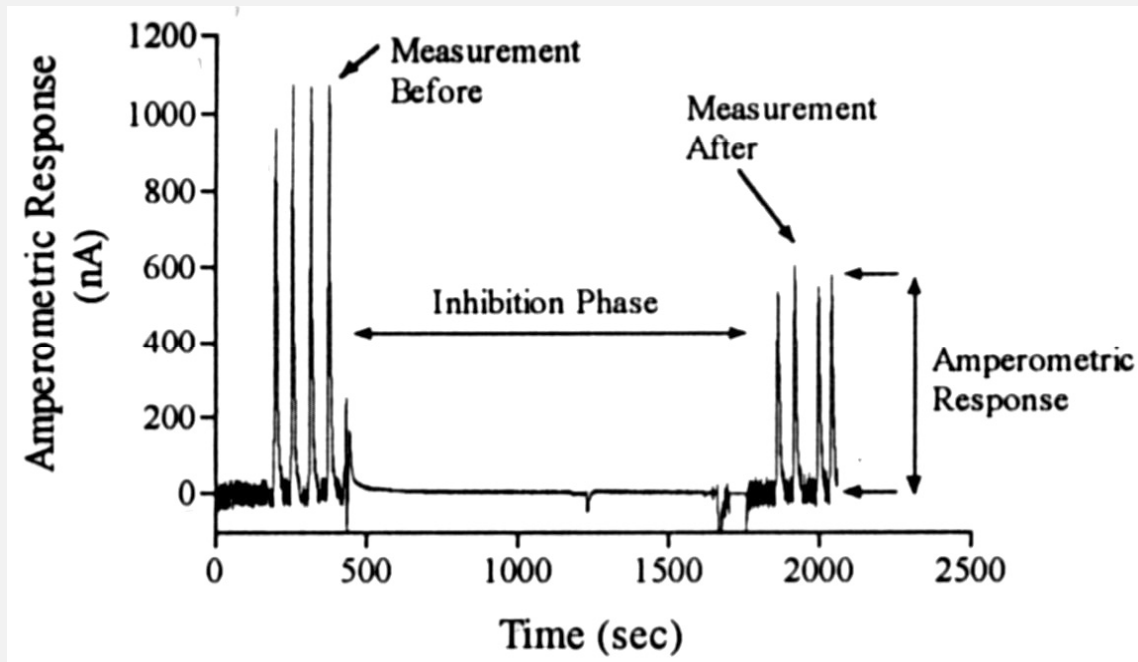


- Co electrocatalyses oxidation of TCh at low working potential (+100mV)
- reduction in enzyme activity after exposure to OP decreases production of TCh(ox) - hence lower signal



# Typical Amperometric Response

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(J.J.Rippeth, Univ. Leeds, 1999)

- measurements performed within flow-injection system
- decrease in amperometric response upon injection of pesticide sample



## Conclusions

- GEM and AET involved in collaborative EU and commercial projects
- GEM – manufacturer and supplier of screen-printing materials and electrodes
- AET – supplier of stable enzyme / protein systems
- synergy between GEM and AET – focus towards development of commercially viable biosensor systems



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