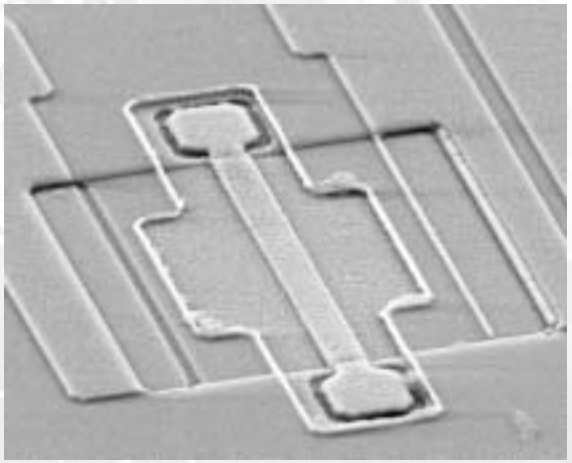
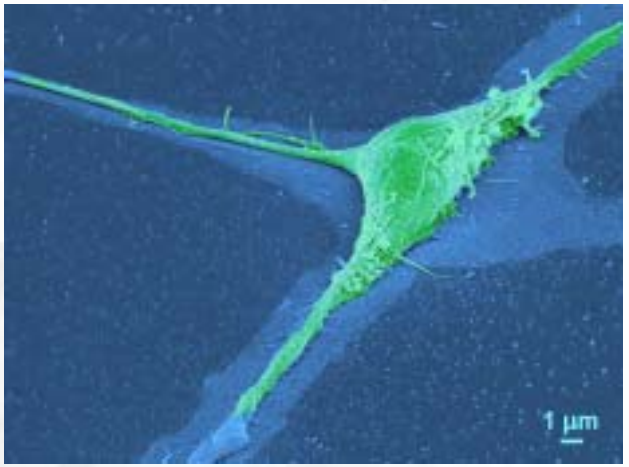
Towards a neuroelectronic sytem – interfacing neurons with microelectronics

Andreas Offenhäusser

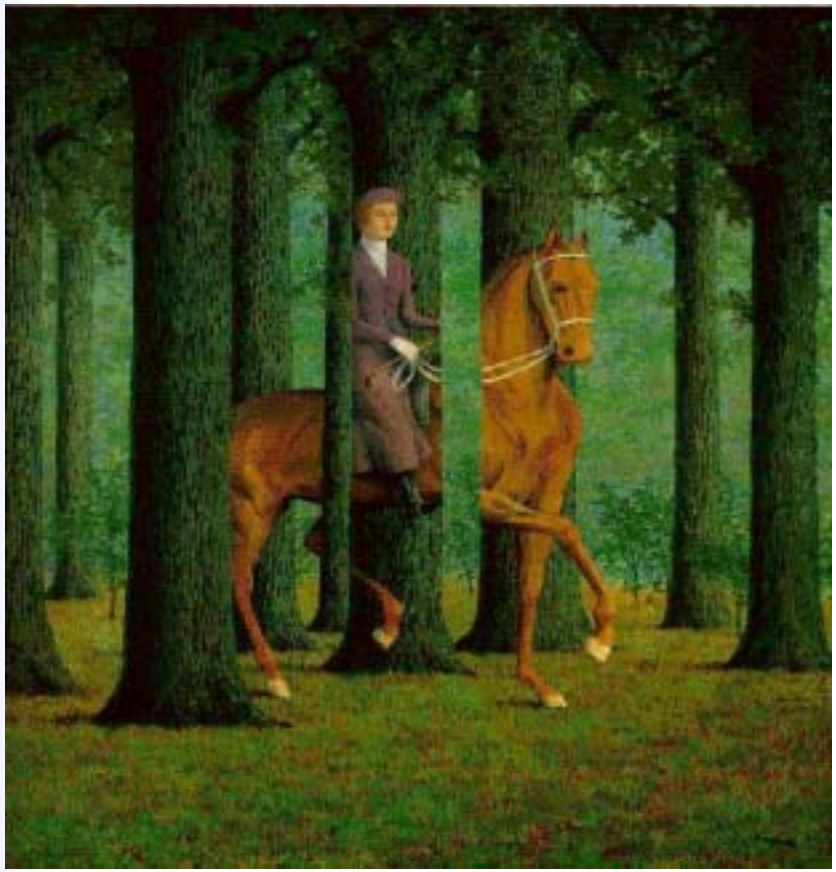
Institute for Thin Films and Interfaces (ISG-2), Forschungszentrum Jülic





Biological information systems

Pattern recognition

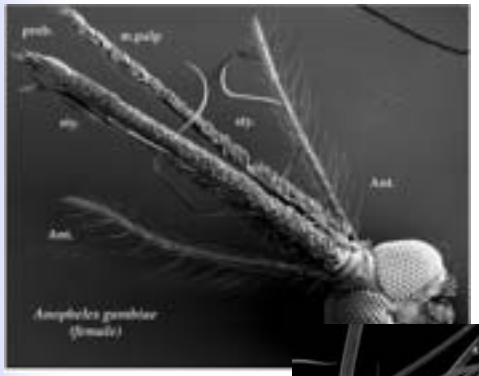


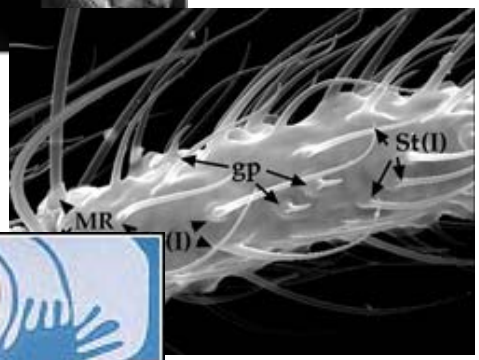
Magritte

Image recognition and processing

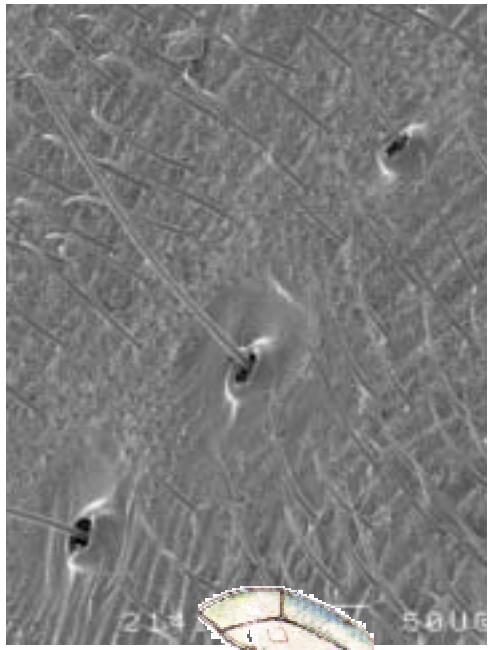
Biological sensing systems

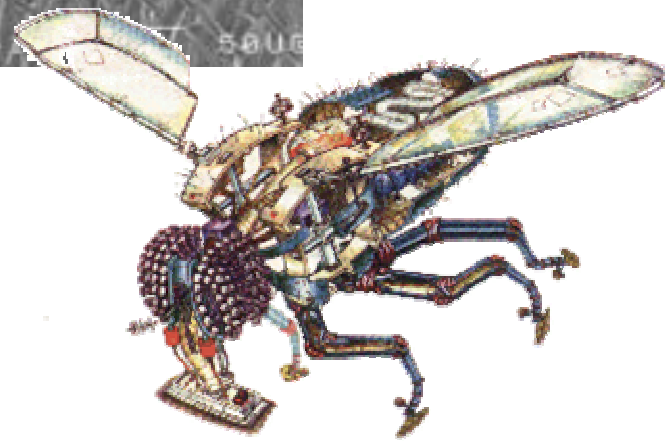
Olfaction



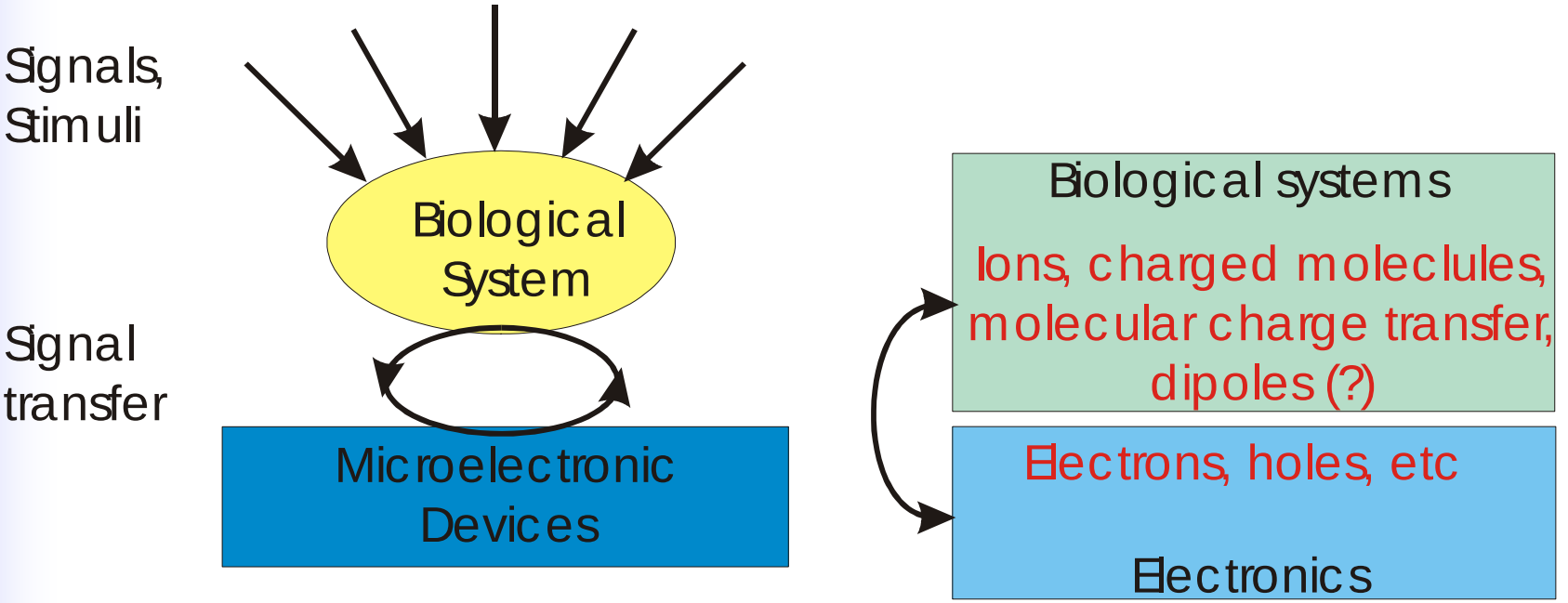


Mechanoreceptors



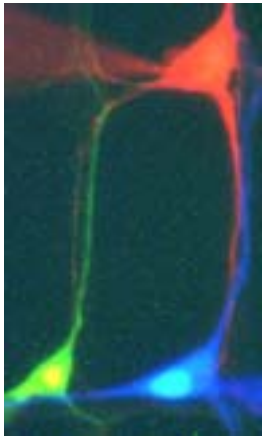


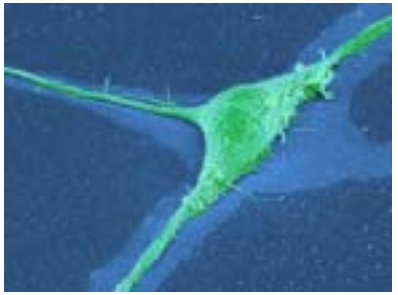
Interfacing Biology with Electronics

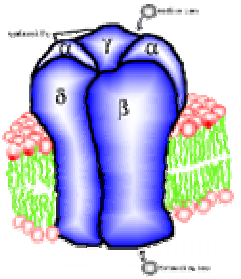


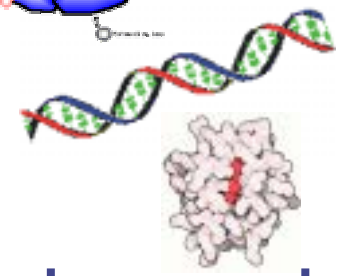
Biology meets Electronics

Structures









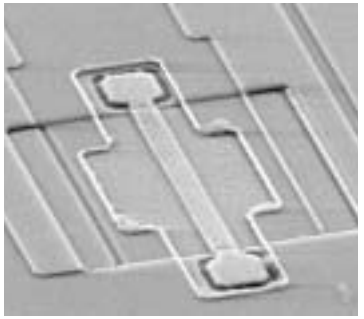
1 mm | 100 µm | 10 µm | 1 µm | 100 nm | 10 nm | 1 nm

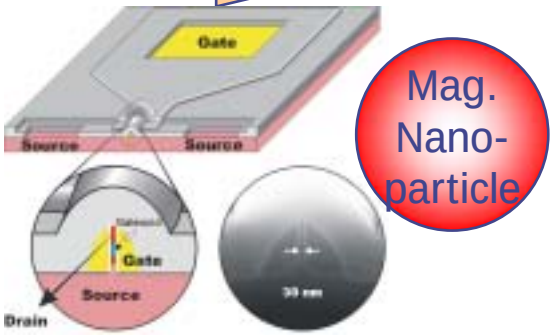
Optical lithography

Bottom-up

Micro- and nano contact printing

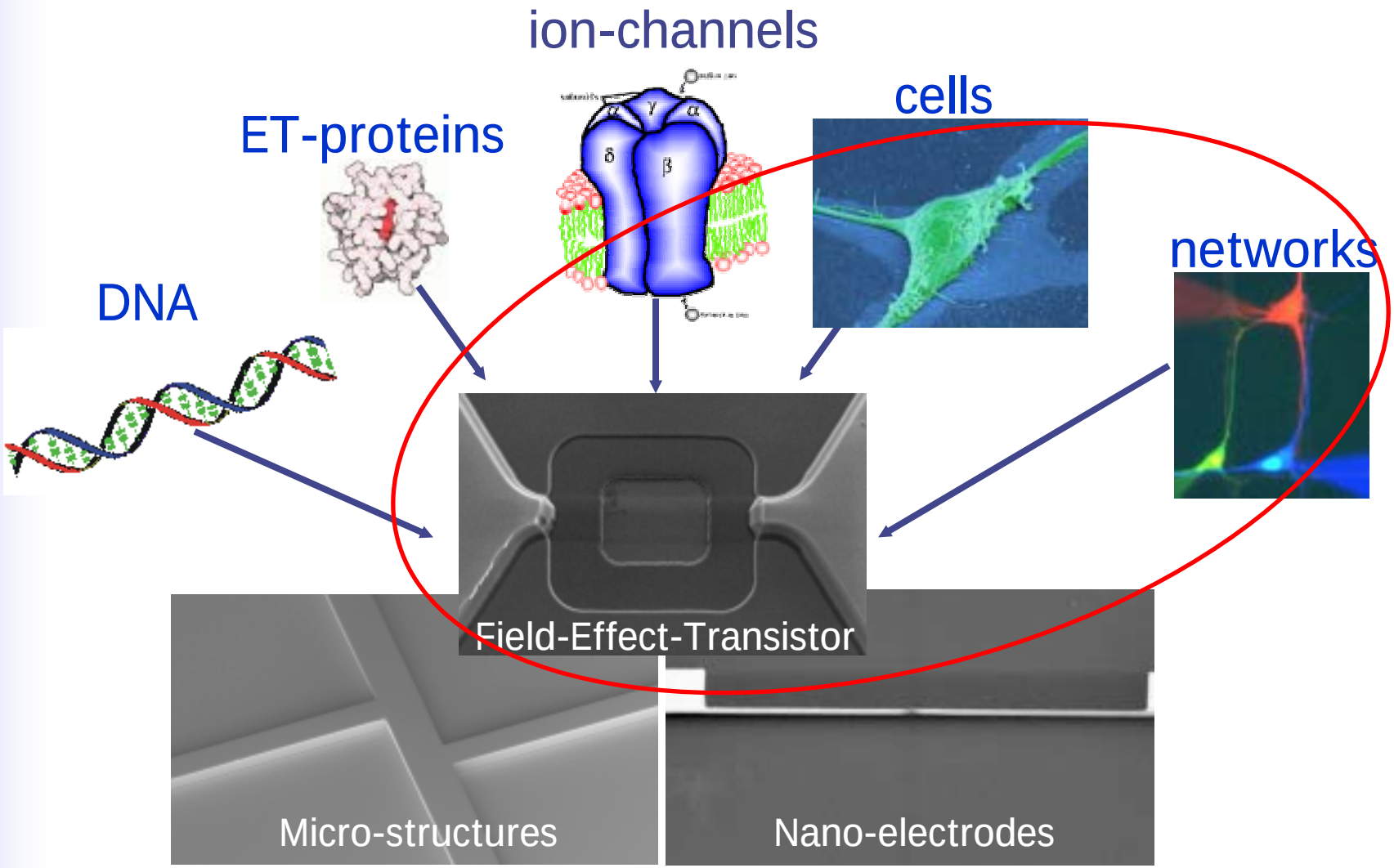
E-beam lithography





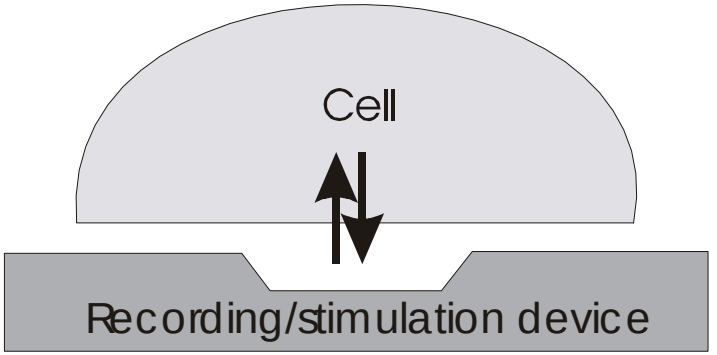
Mag. Nano-particle

Bioelectronic hybrids

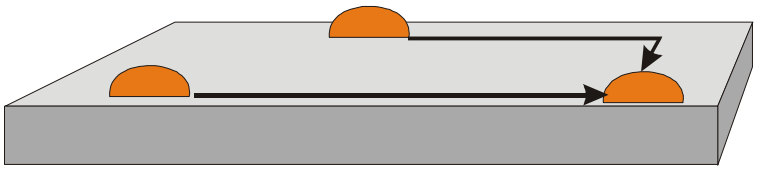


Bioelectronic hybrids

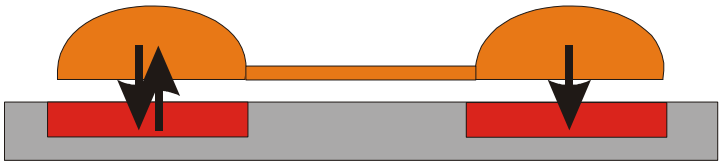
Bi-directional coupling:



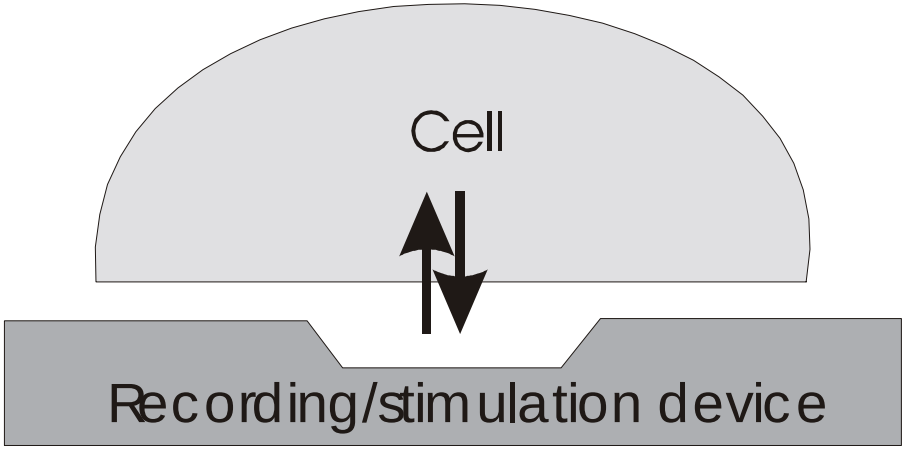
Defined networks:



Interfacing networks of neurons with microelectronic devices:



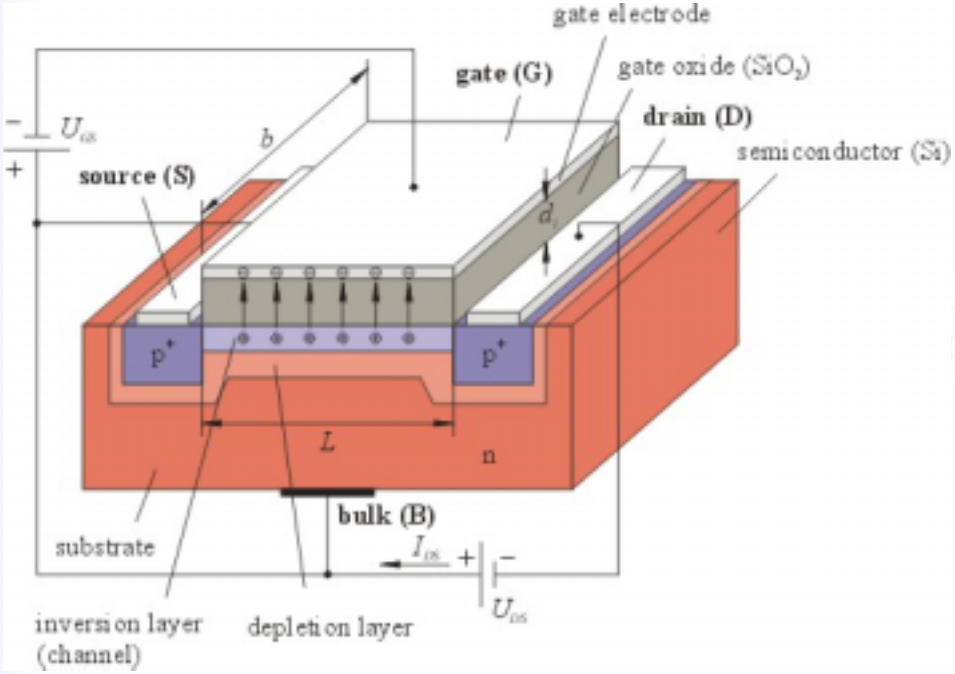
Cell-transistor coupling



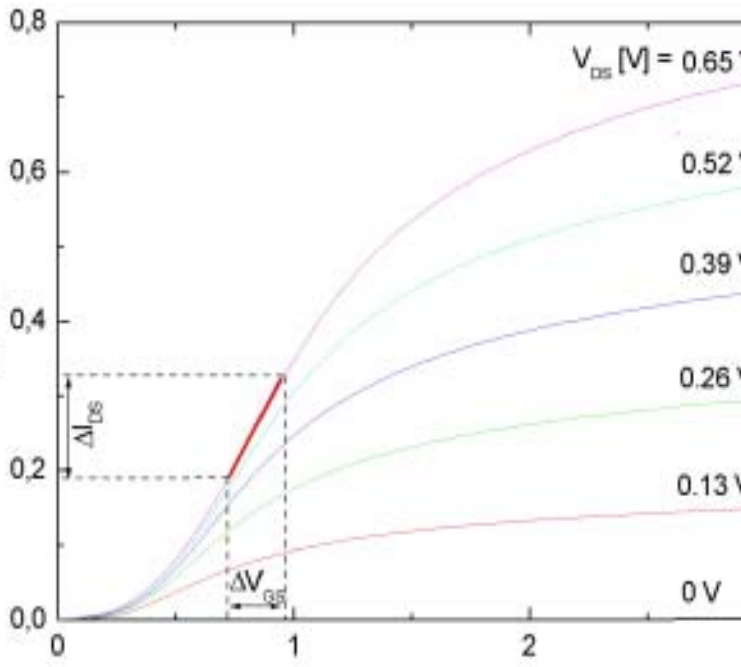
1) Recording of cellular signals using microelectronic devices

2) Stimulation cellular response using microelectrode

Field-Effect Transistor

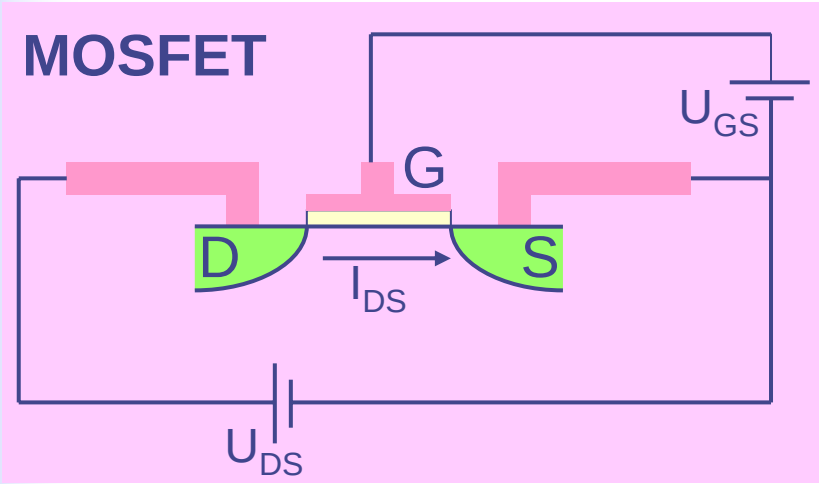


Transfer characteristics

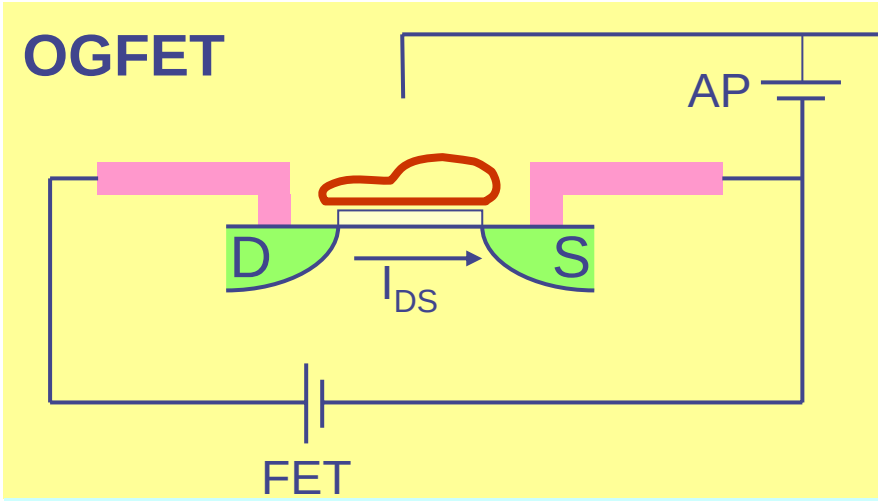


- *Gate potential modulates inversion channel between drain and source*
- *Changes in drain-source current are proportional to changes of the gate potential (linear region)*

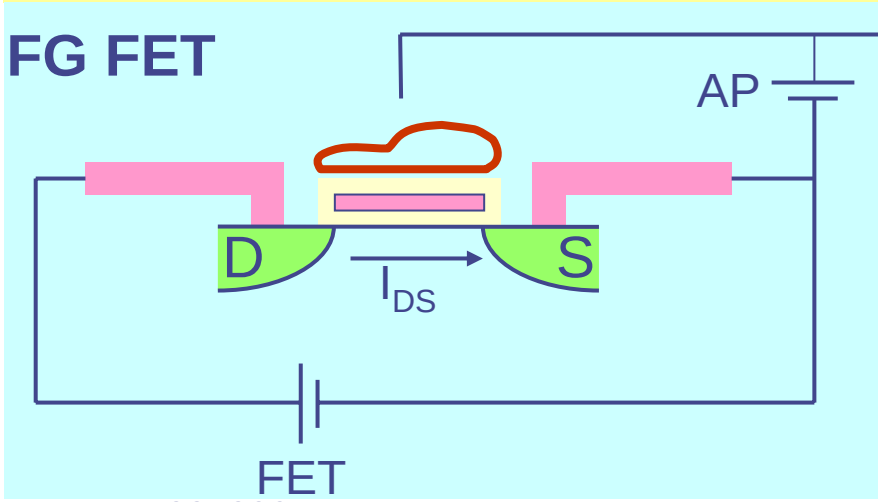
Concepts



Concept derived from the work of the Bergveld laboratory e.g. BERGVELD, P., WIERSMA, J., MEERTENS, H. (1976) IEEE Trans. Biomed. Eng. **23** 136-144



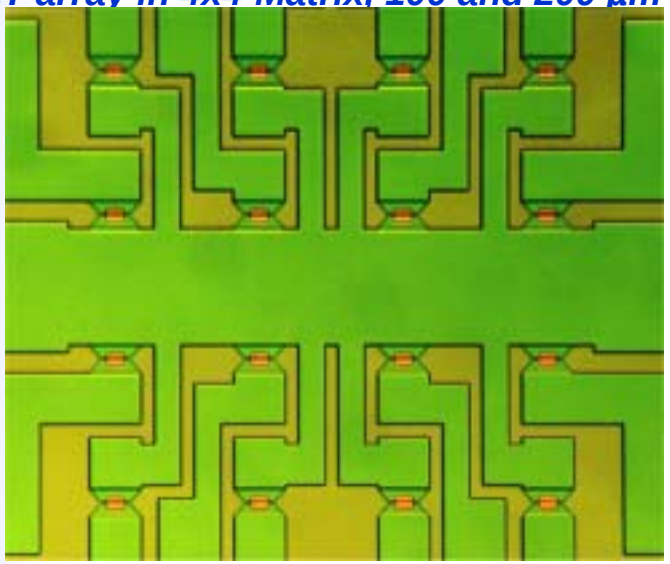
Offenhäusser A., Sprössler C., Matsuzawa M., Knoll W. (1997) Biosensors & Bioelectronics 12: 819-826.
Ingebrandt S. et al. (submitted)



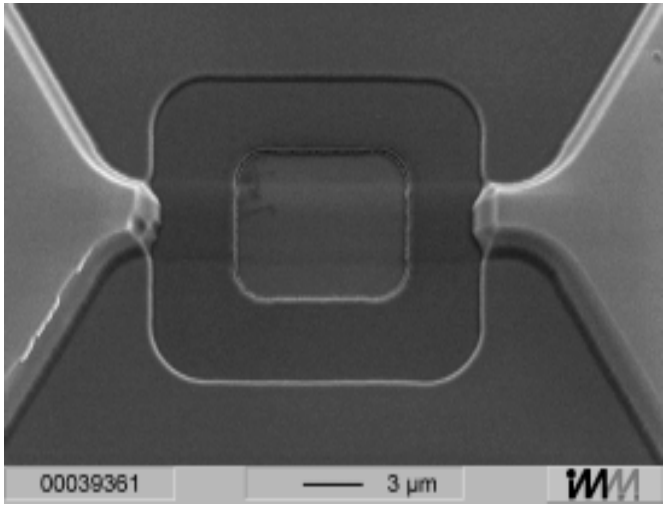
Offenhäusser A., Rühe J., Knoll W. (1995) J. Vac. Sci. Technol. A 13, 2606-2612.
Meyburg S. et al. (submitted)

Field-Effect Transistor

FET array in 4x4-Matrix, 100 and 200 µm distance

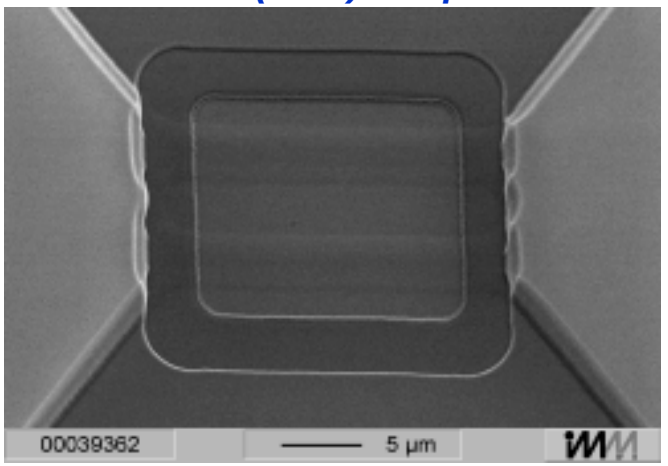


Individual Gate (SEM) 5x1.5 µm²



Encapsulated Chip

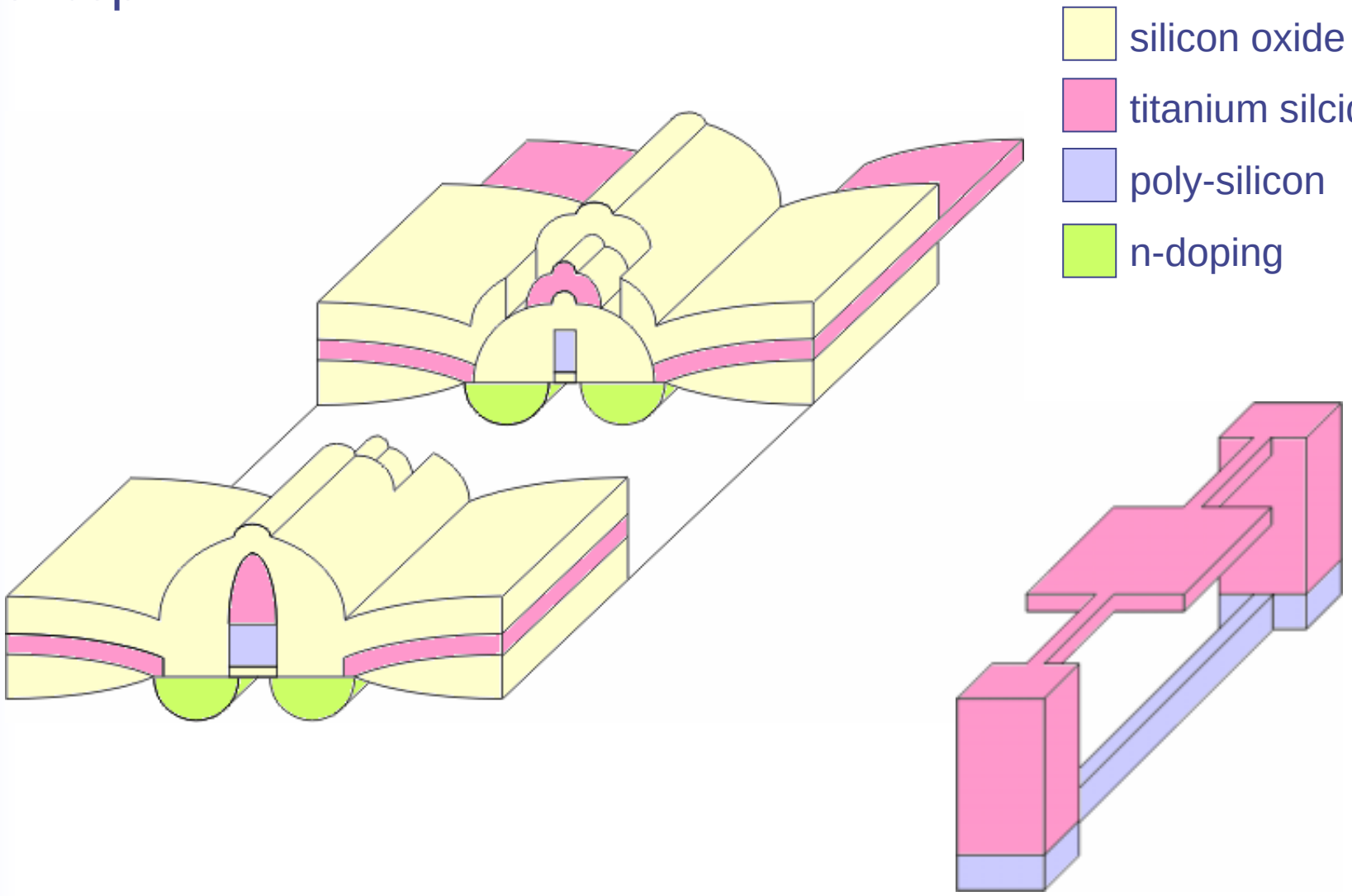
Double-Gate (SEM) 8x6 µm²



A. Offenhäusser et al., Biosens.&Bioelec.12 (1997) 819

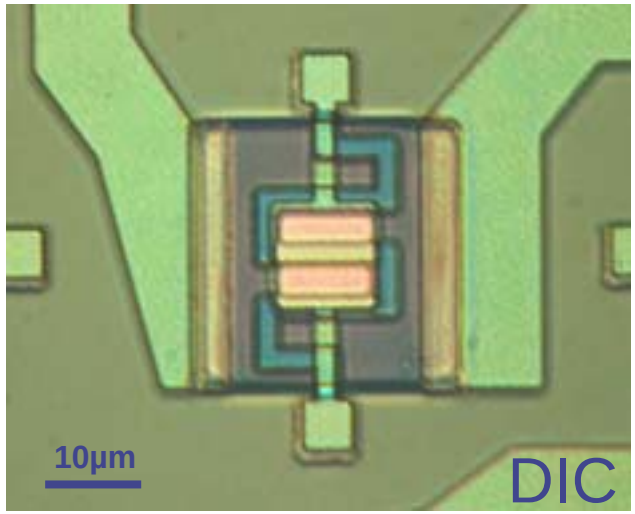
Floating-gate Field-Effect Transistors

Concept:

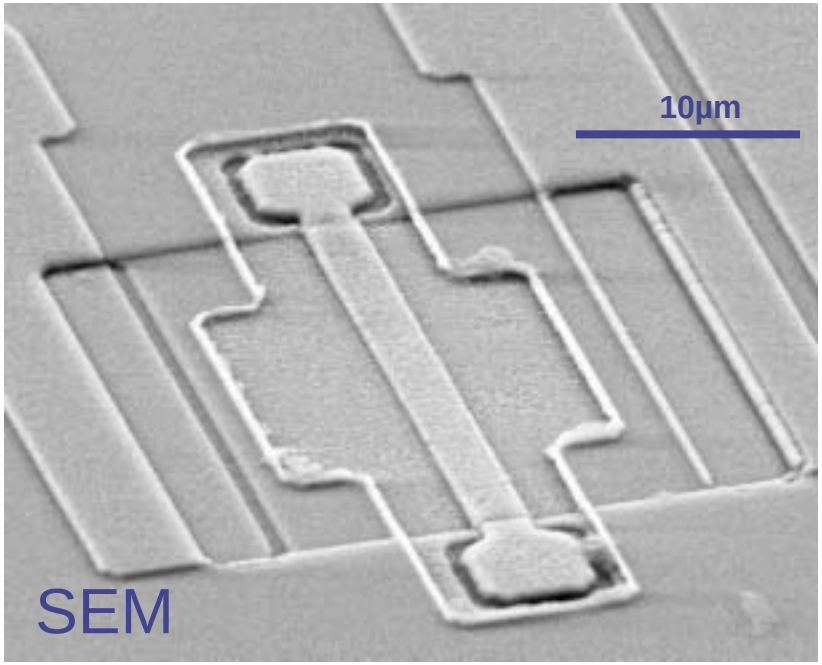


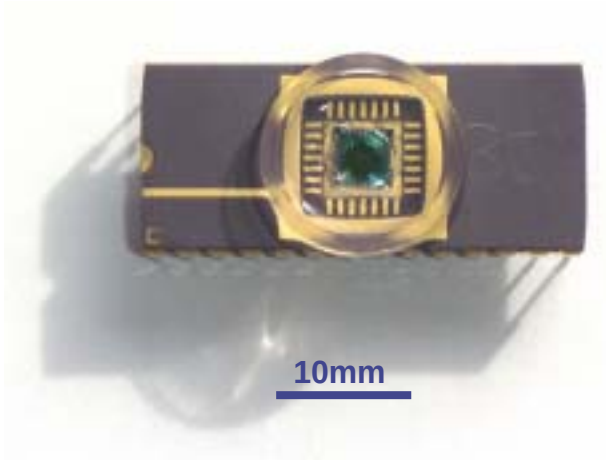
Floating-gate Field-Effect Transistors

Poly-Si sensor gate, MEA



Ti-Si sensor gate, LIN

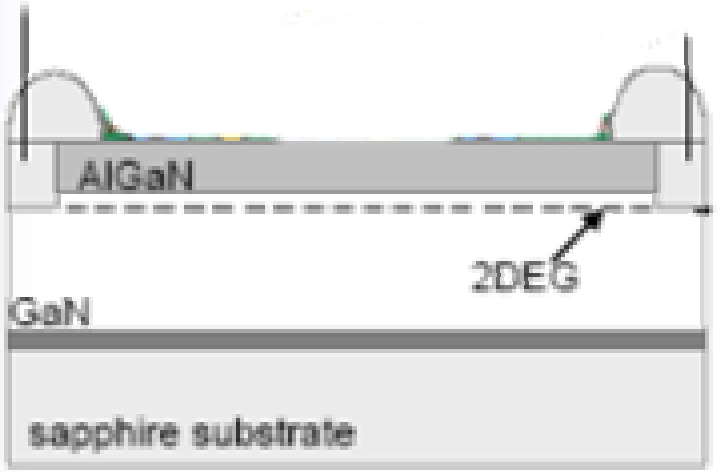


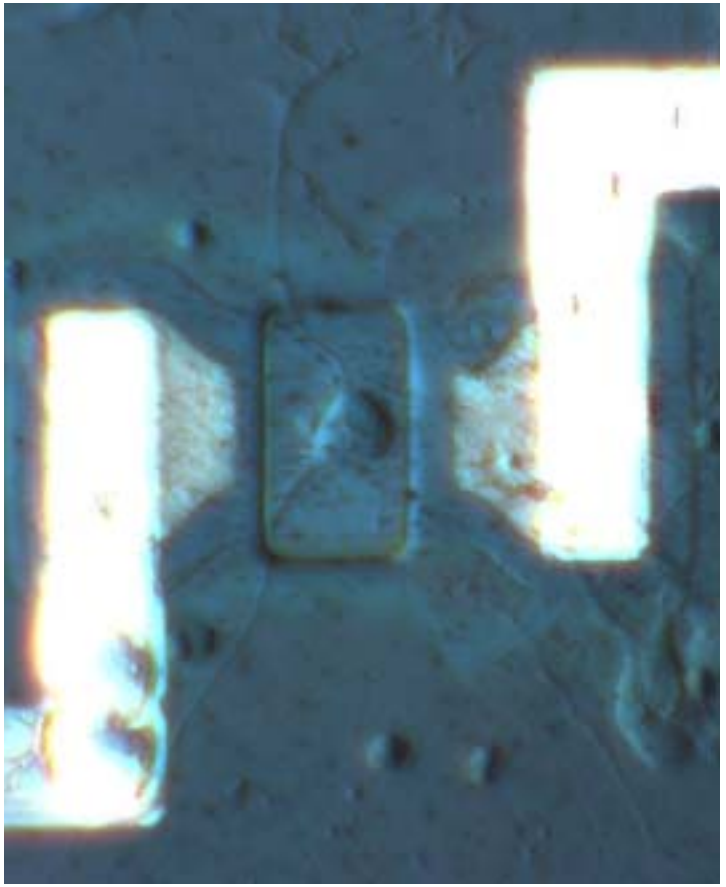


Source: S. Meyburg, J. Moers, M. Goryll, S. Ingebrandt

High Electron Mobility Transistors (HEMT)

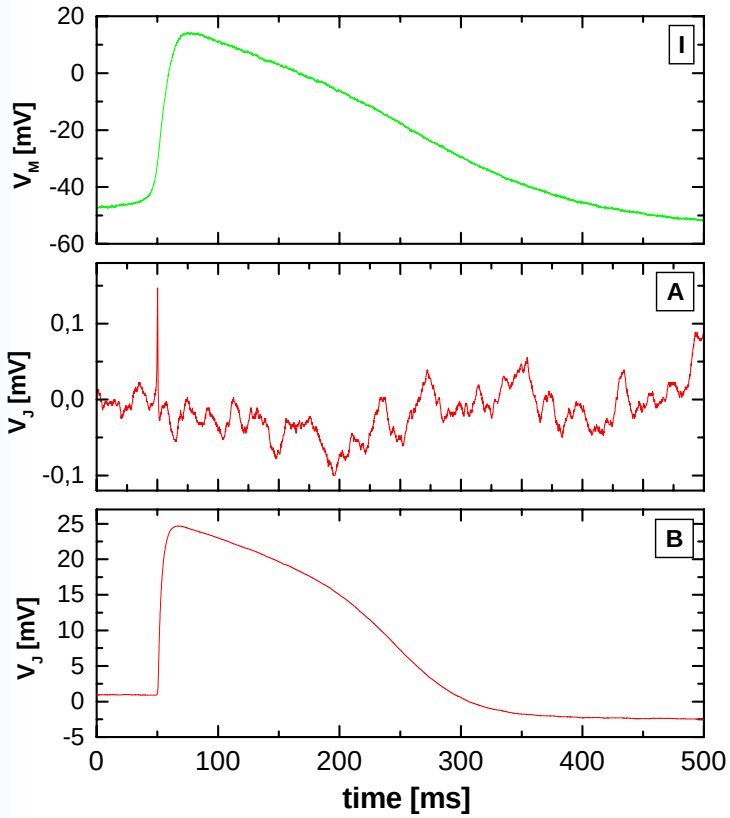
AlGaN/GaN HEMT-structure





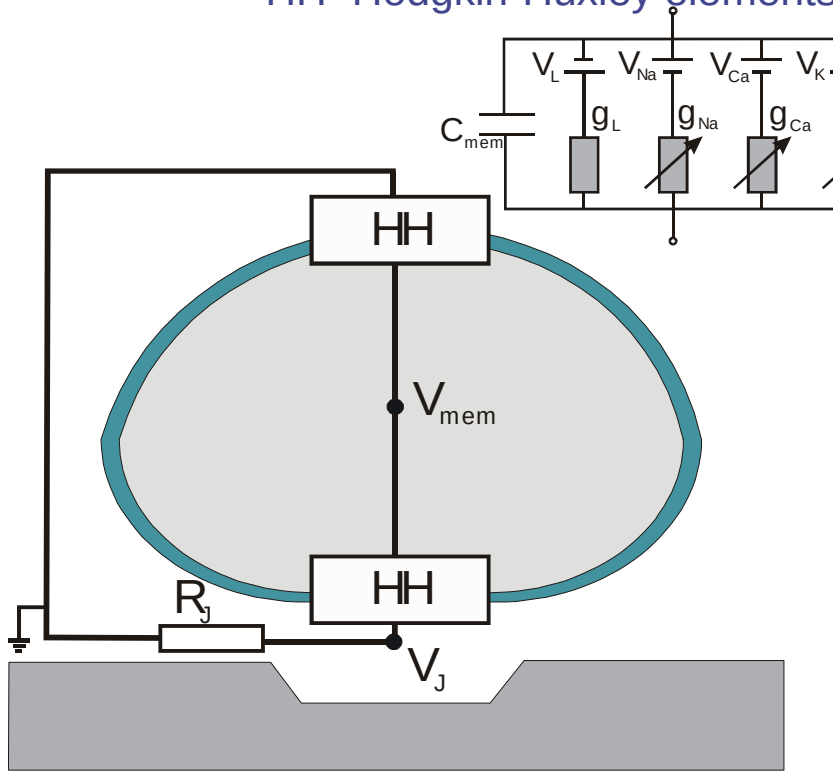
Source: Georg Steinhoff, Martin Eickhoff, Martin Stutzmann (Walter-Schottky-Institute, TU Munich)
Sven Ingebrandt, Günter Wrobel, Peter Jaworka, Michel Marso, Hans Lüth(ISG, FZ Jülich)

Cell-Transistor Coupling



C. Sprössler et al., Phys. Rev. E 60 (1999) 2171-2176

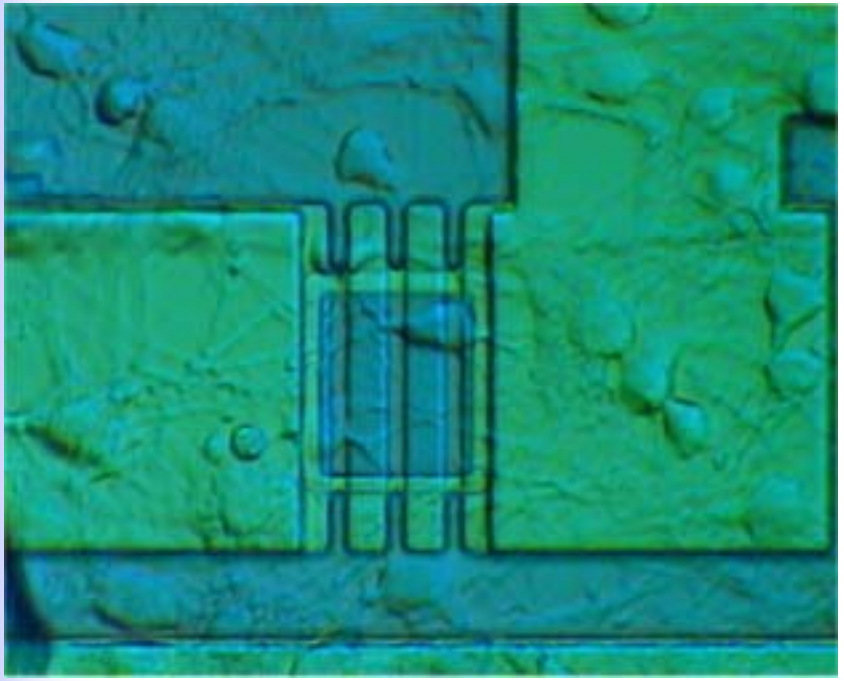
HH=Hodgkin-Huxley elements

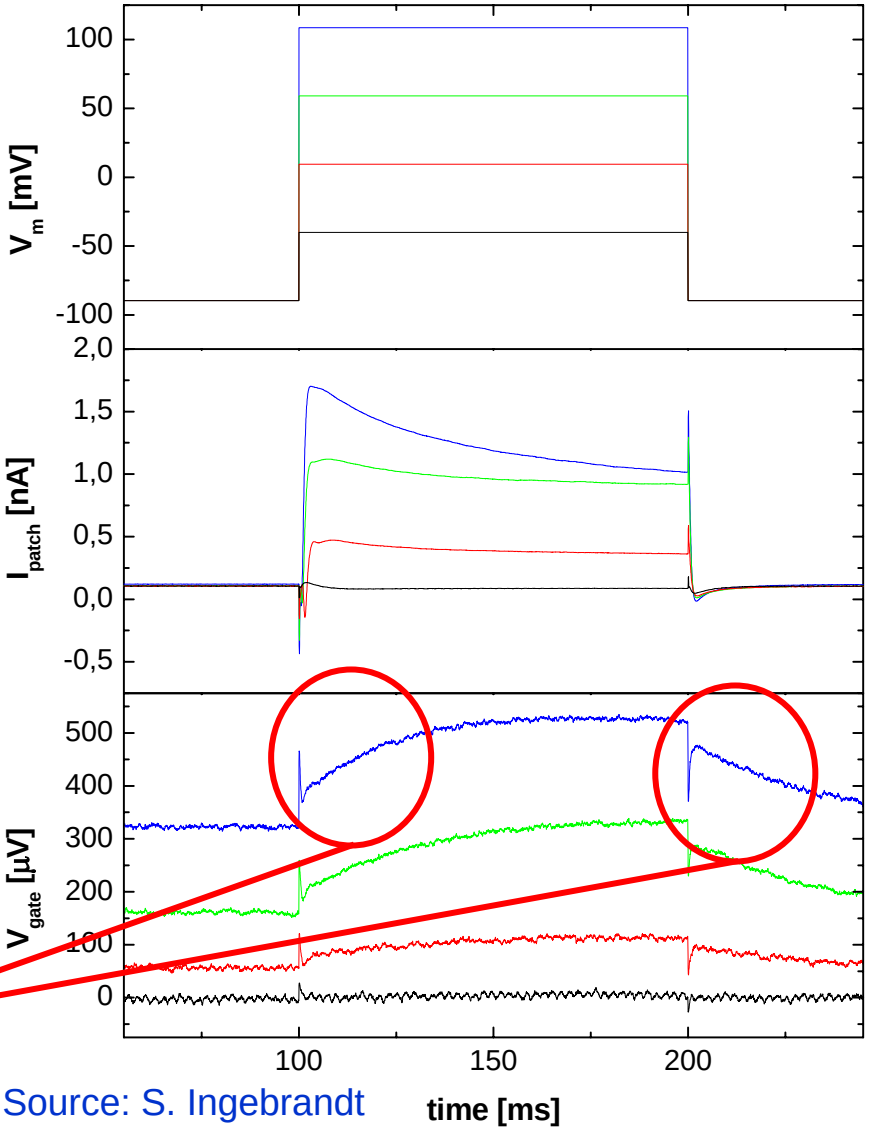


W.G. Regehr et al., J. Neurosci. Meth. 30, 91-106 (198
S. Vassanelli & P. Fromherz, Appl.Phys.A 66, 549-
(1998)
C. Sprössler et al., Phys. Rev. E 60, 2171-2176 (1999

Neuron-Transistor Coupling

Brain stem neurons (rat)





differences

Signals are not averaged !

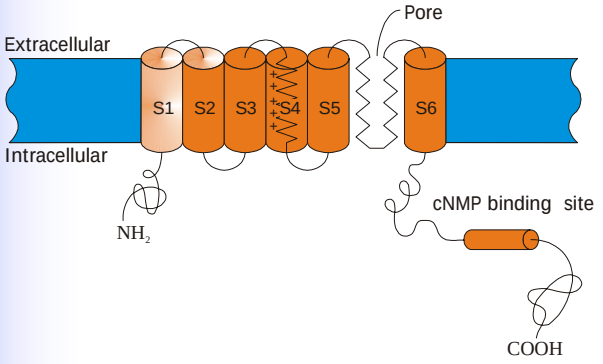
Source: S. Ingebrandt

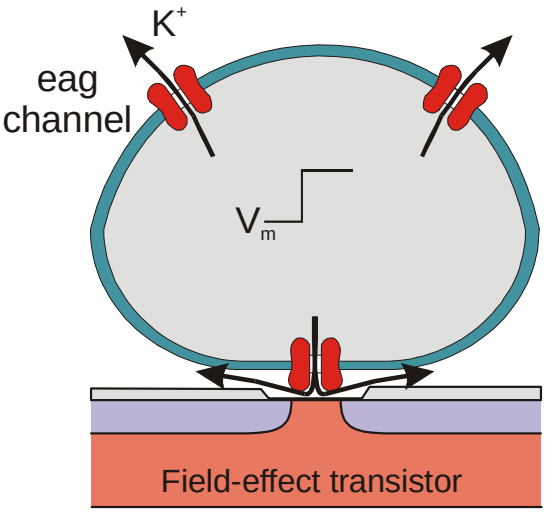
http://www.fz-juelich.de/isg/ISG2

Cell-transistor-hybrids

Genetically engineered cells (HEK293):

Bovine ether a go go (beag1)
K^+-channel (Frings et al (1998))



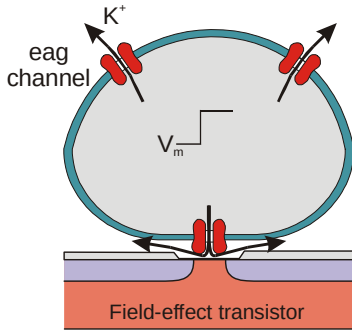


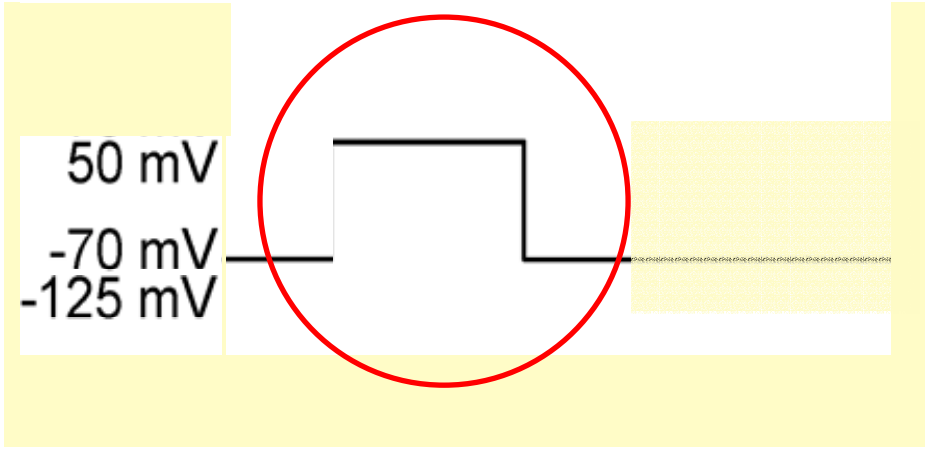
- outward rectifying potassium channel
- from the retina of bovine
- involved in control of cellular resting potential and regulation of action potentials
- channel activation is controllable

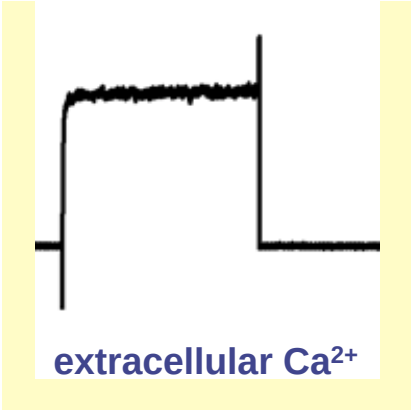
Cell-transistor-hybrids

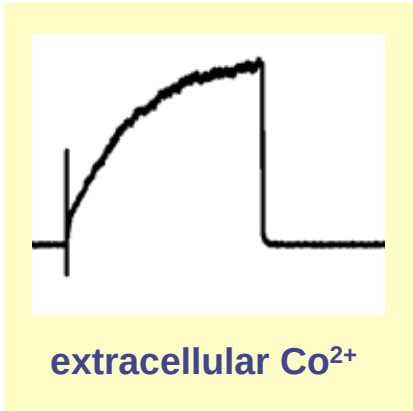
Genetically engineered cells (HEK293):

Bovine ether a go go (beag1)





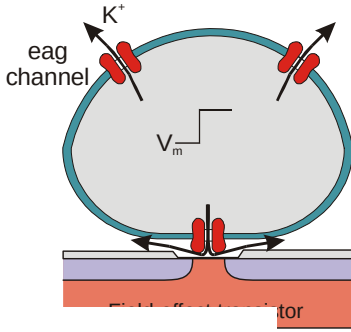




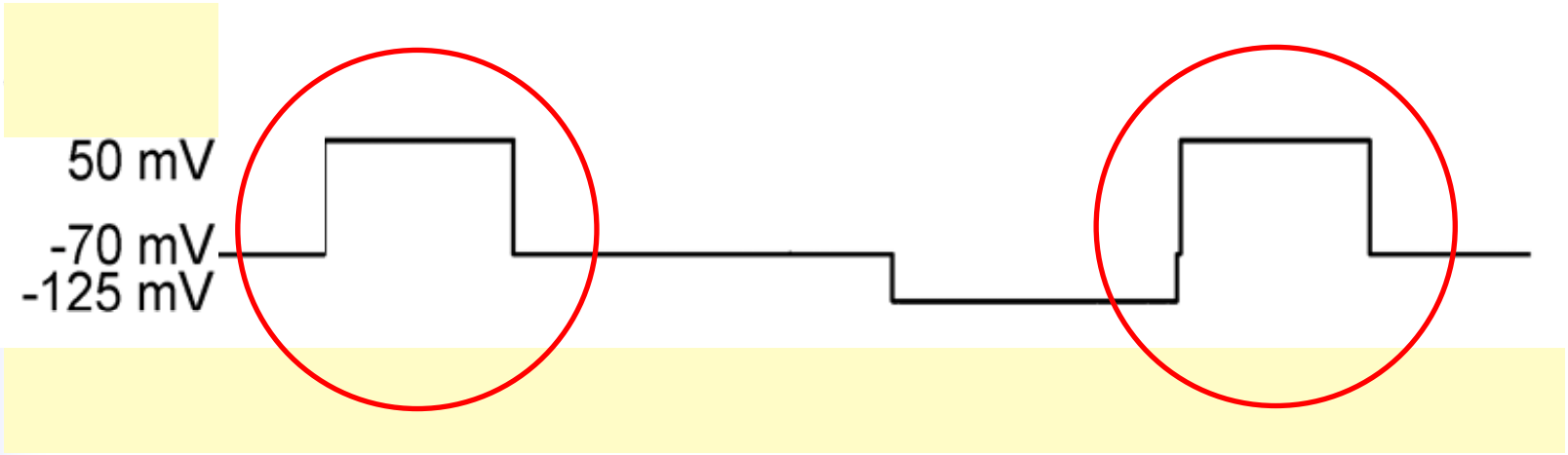
Source: G. Wrobel

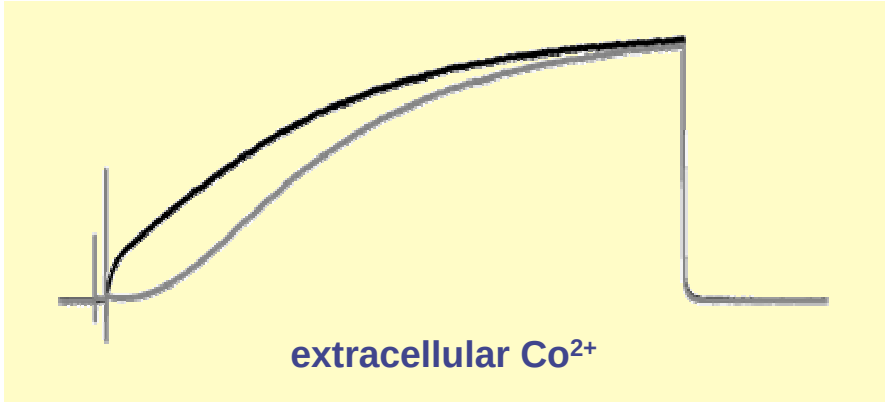
Cell-transistor-hybrids

Genetically engineered cells (HEK293):



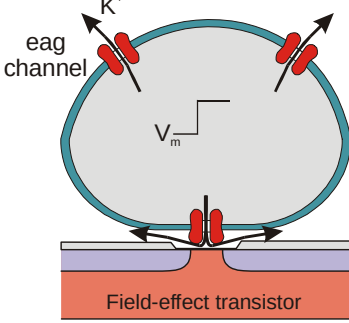
Channel-activation is dependent on stimulation pulses



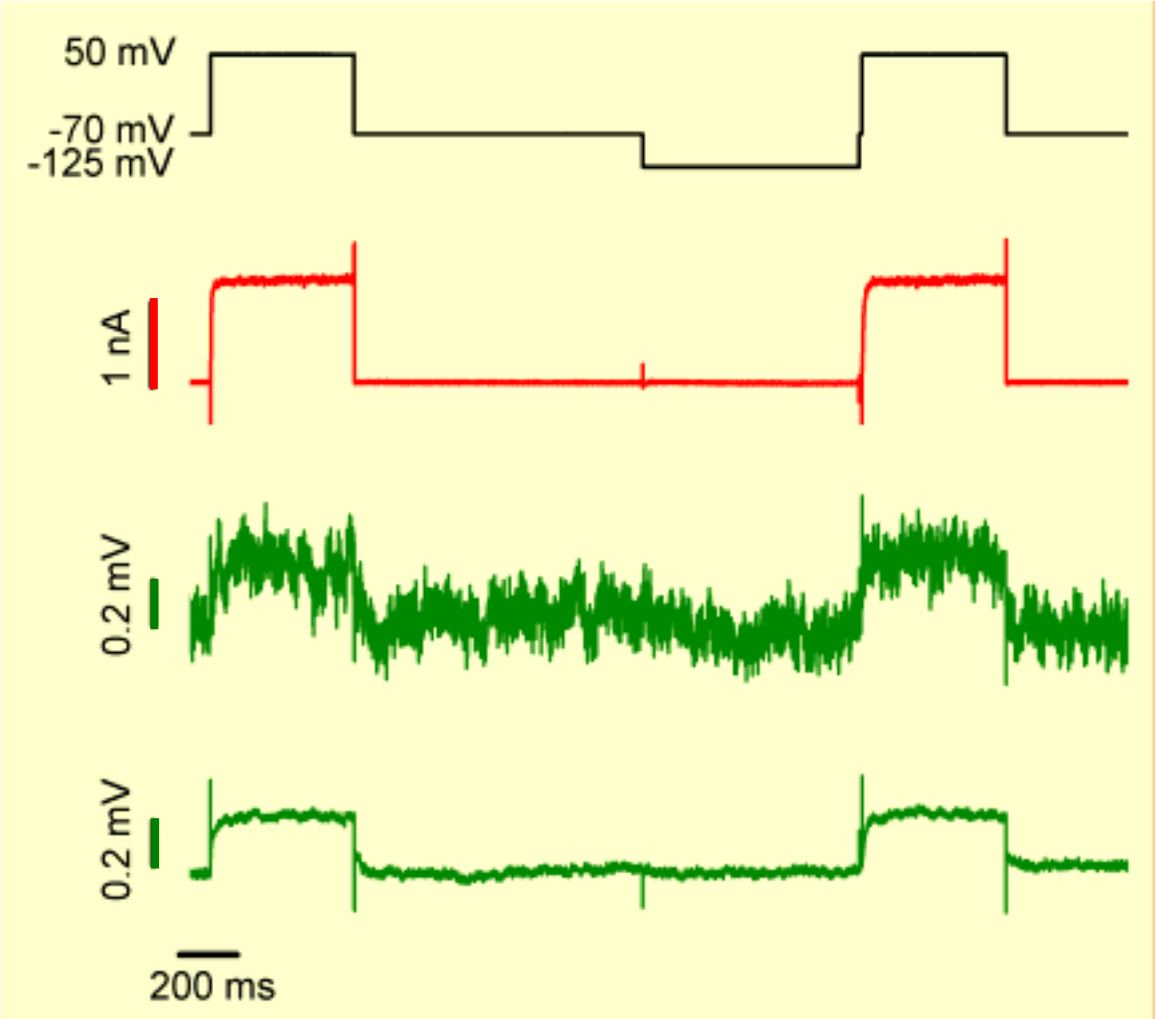


Source: G. Wrobel

Cell-transistor-hybrids



Extracellular Ca^{2+} solution / p-channel FET



stimulation protocol

membrane current

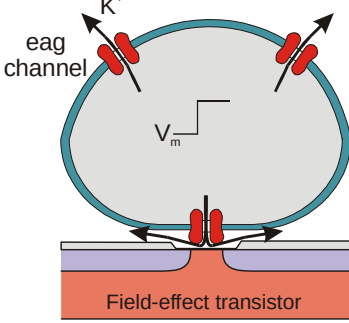
FET-recording

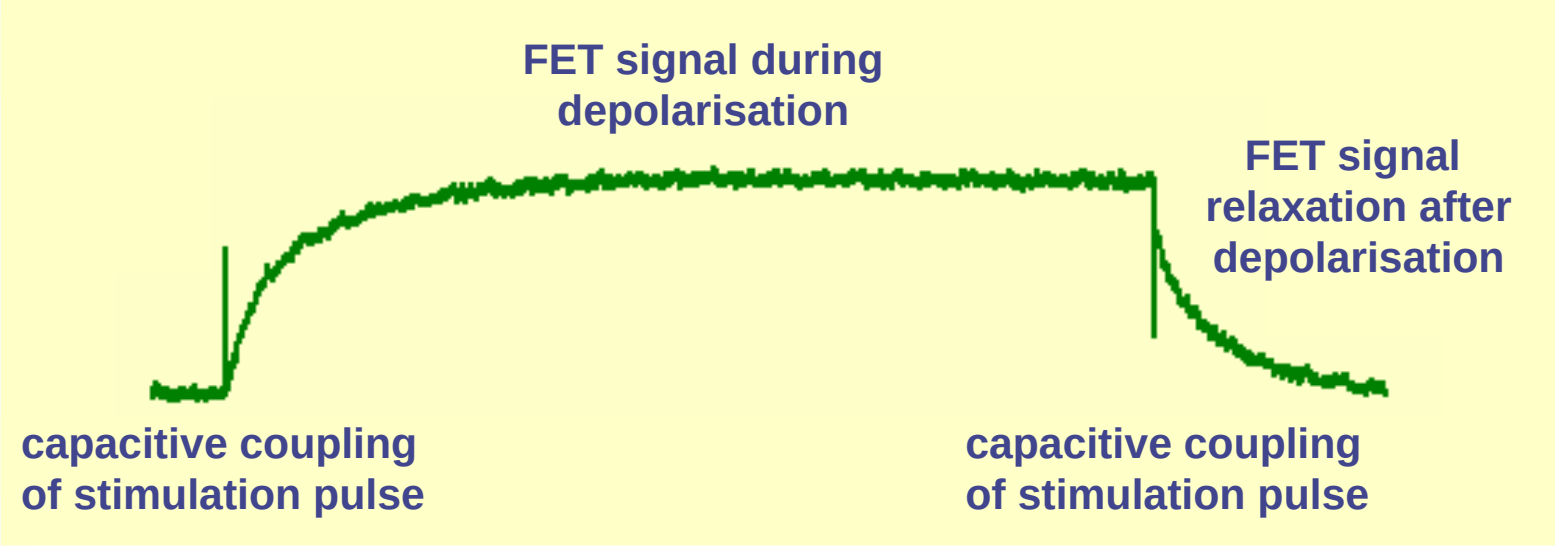
averaged (n = 50)
FET-recording

Source: G. Wrobel

Cell-transistor-hybrids

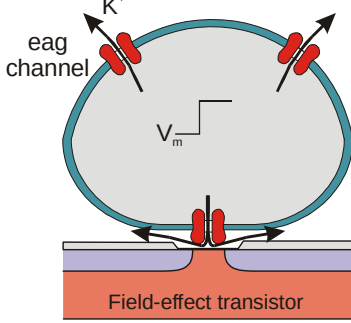
Signal shape



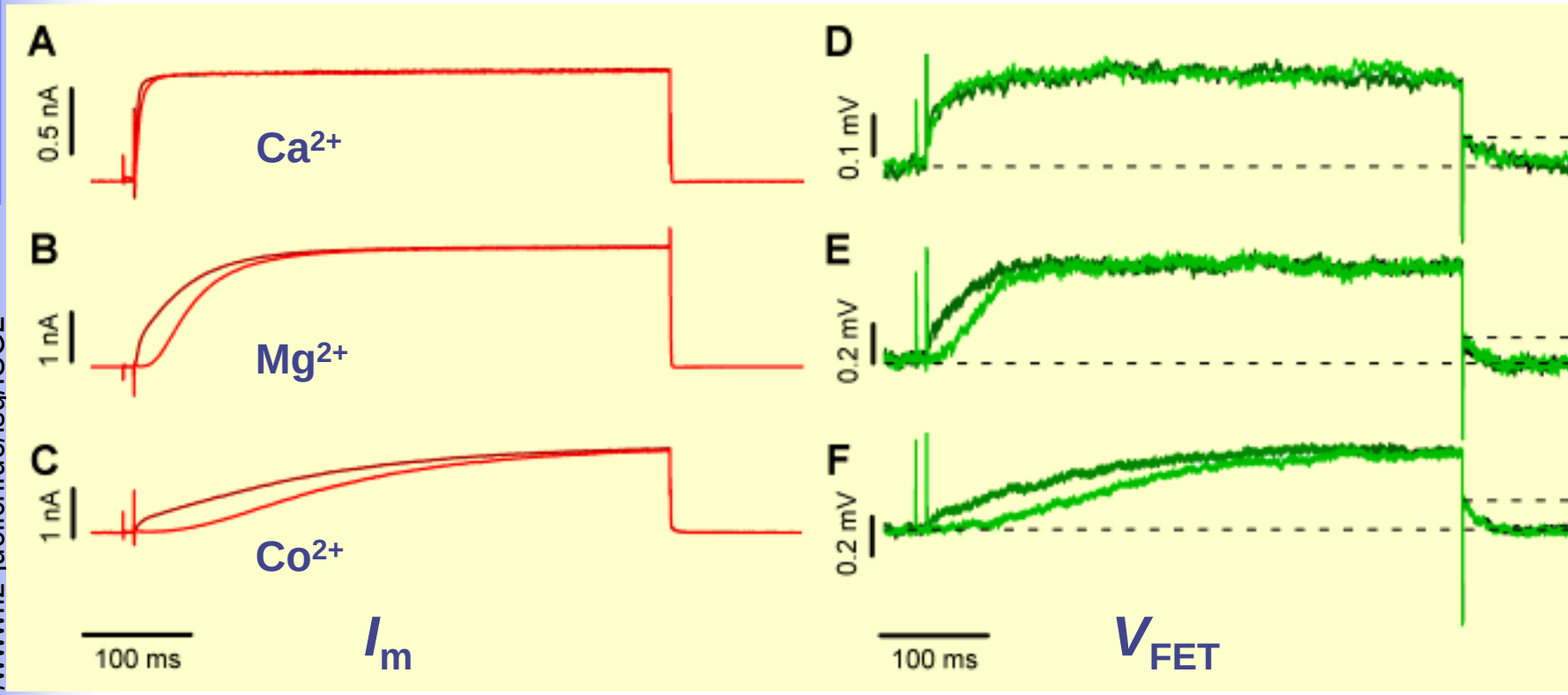


Source: G. Wrobel

Cell-transistor-hybrids



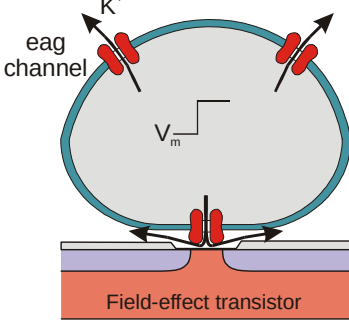
Comparison of whole-cell membrane current and p-channel FET signals

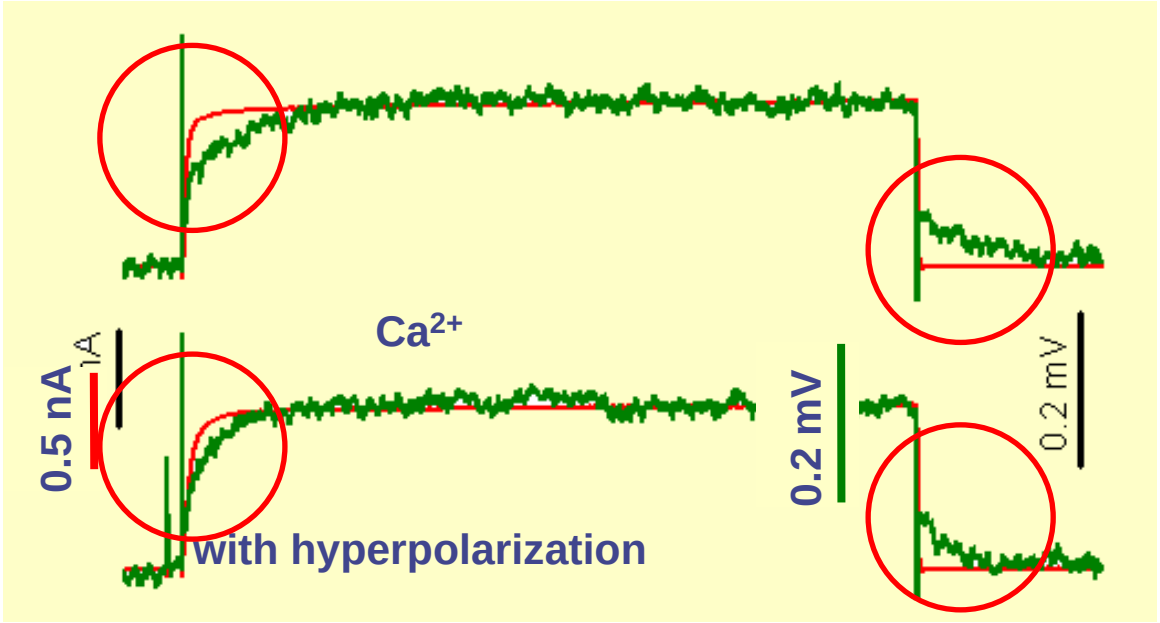


Source: G. Wrobel

Cell-transistor-hybrids

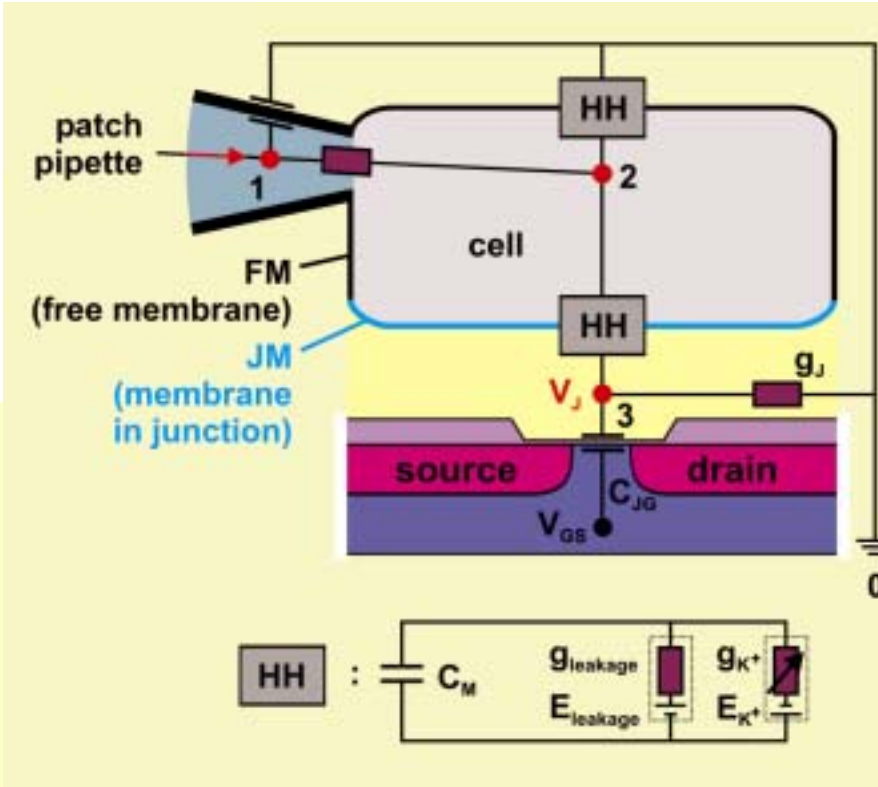
Time course of membrane current and FET signals (p-channel FET)

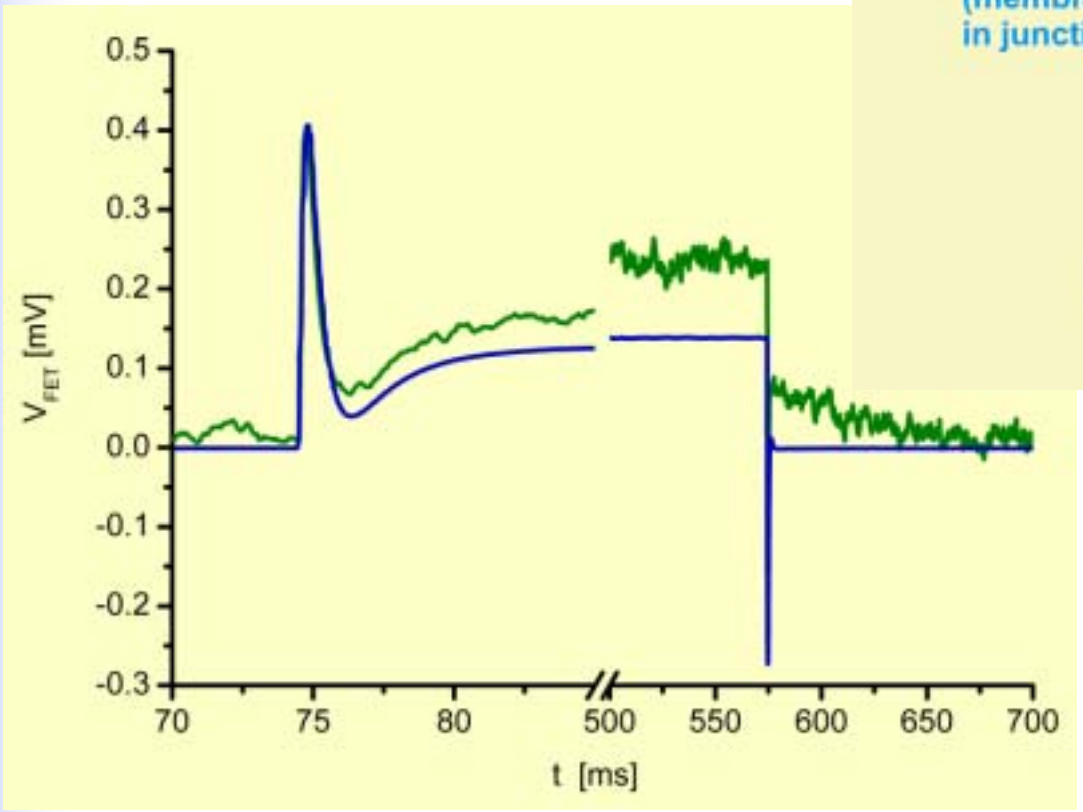




Source: G. Wrobel

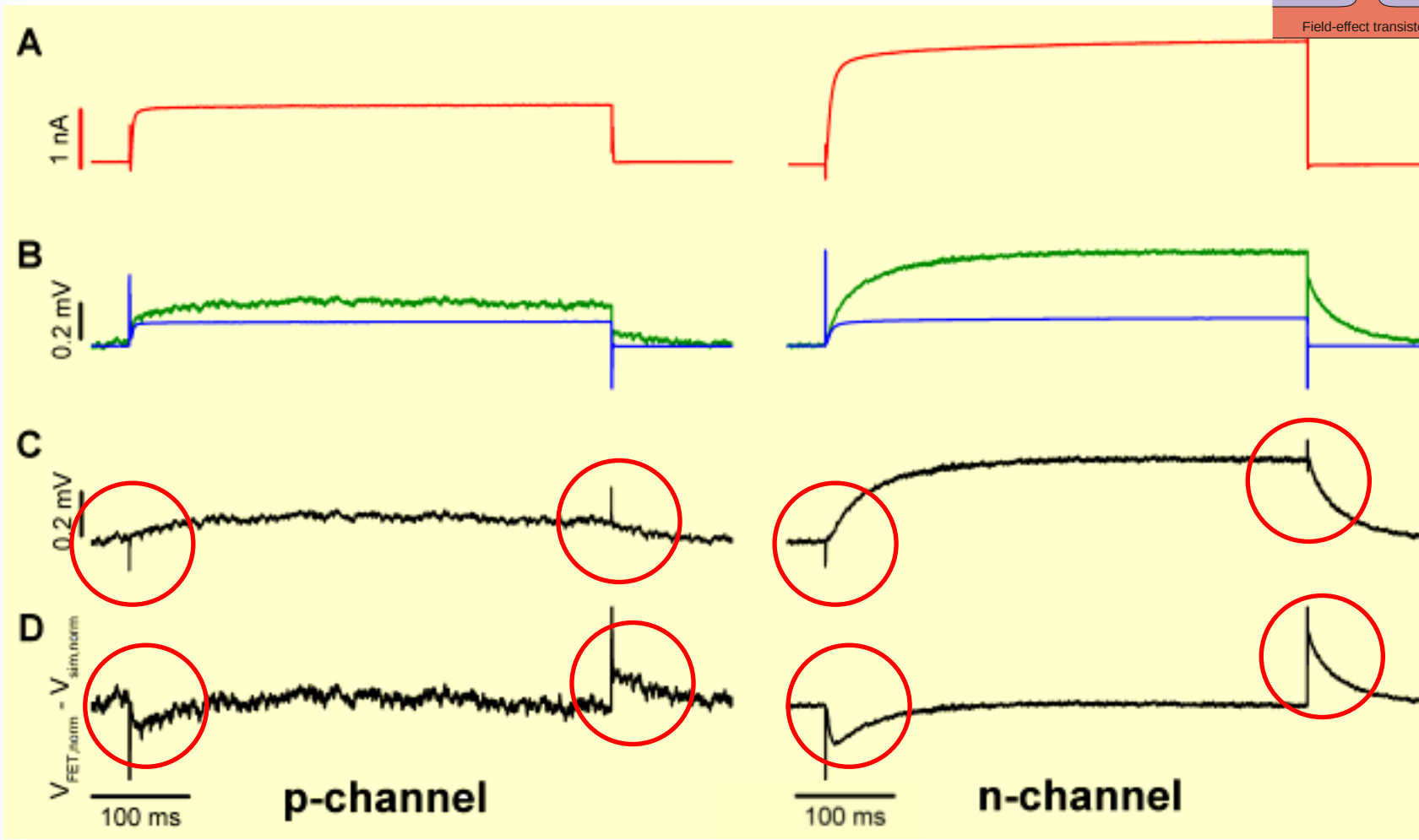
Signal simulation- Point Contact Model

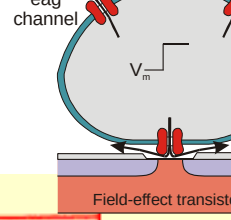




Source: G. Wrobel

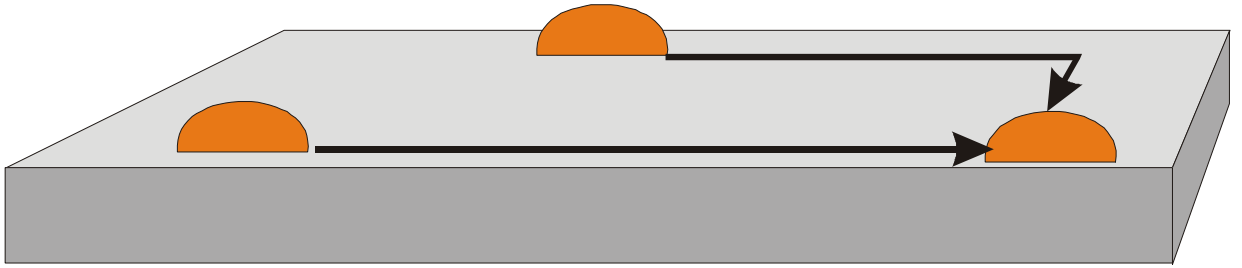
p- and n-channel devices





Source: G. Wrobel

Defined networks of neurons

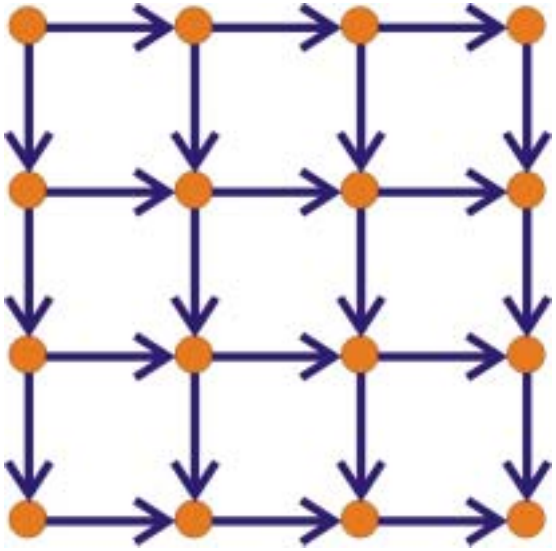


1) To design and create in vitro neural circuits

2) To study of neural information processing and transfer

Neuronal information processing

Information processing and transfer:



Goal:

To design and create in vitro neural circuits with which to further the study of neural information processing and transfer

Requirements:

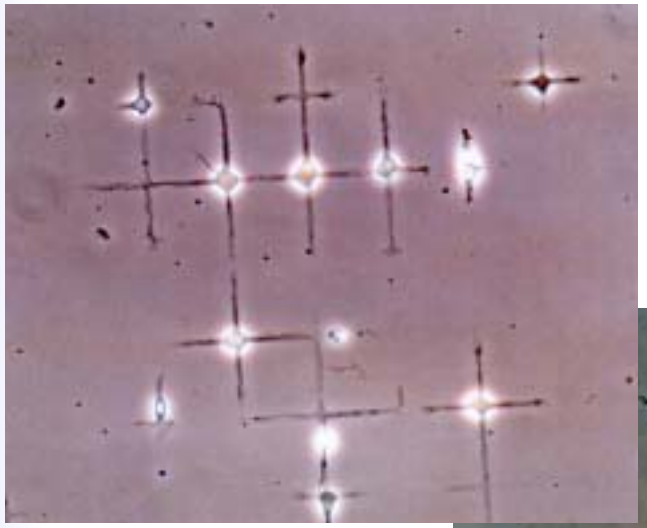
Cellular Lithography:

- Patterned substrates
  - patterning the chemistry of substrates
  - growing cells in patterns
- Substrate topography
  - growing cells in patterns

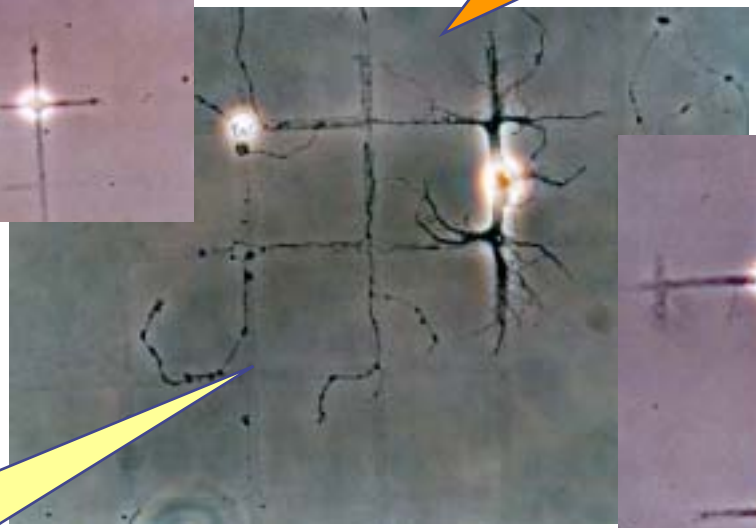
http://www.fz-juelich.de/isg/ISG2

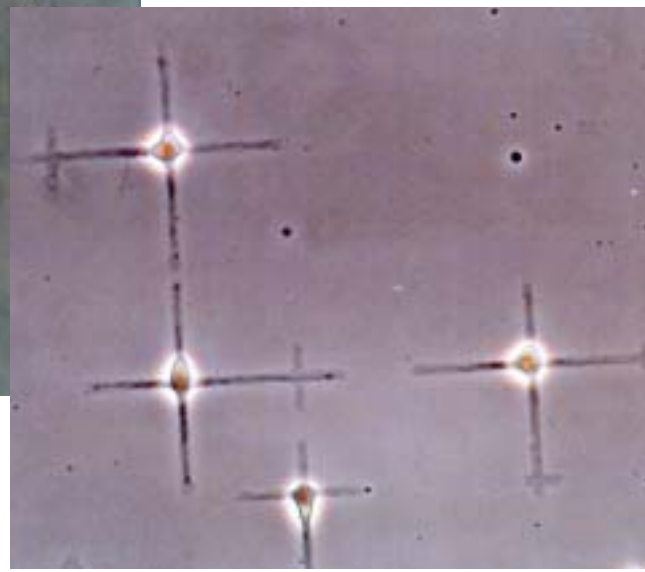
Cell patterning

Cellular lithography



Backgrounds of
cell repulsive material



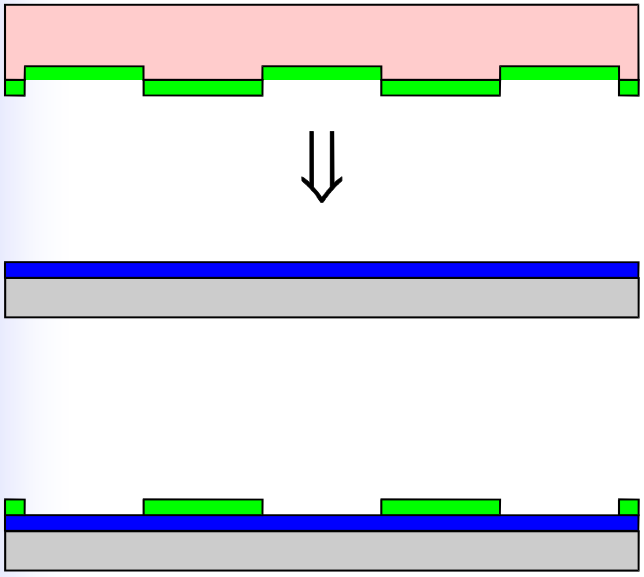


Grid patterns of
cell friendly material

M. Scholl, et al, J. Neurosci. Meth. (2000) *104*, 65-75

Microcontact Printing

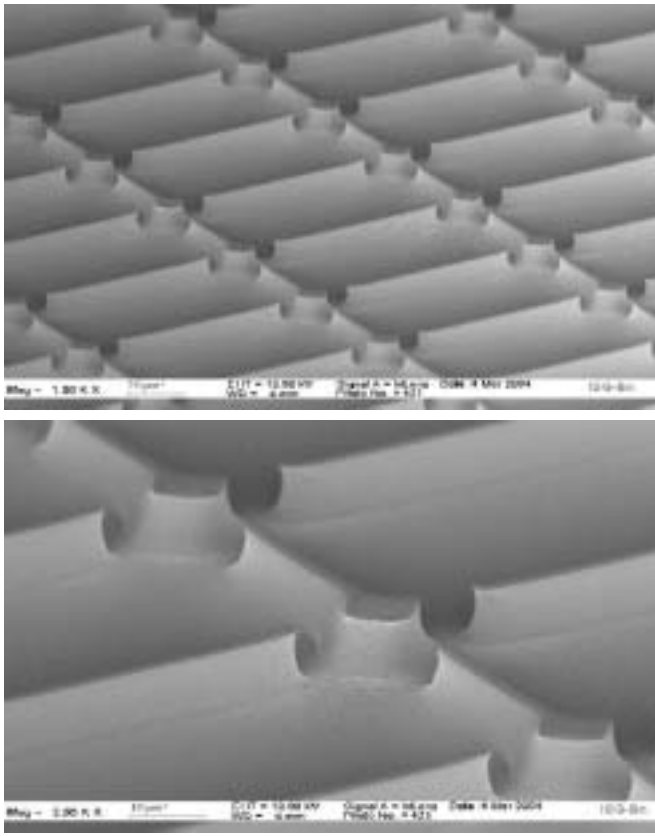
‘Soft’ (biomolecular) lithography



PDMS-stamp with `ink´

Substrate with surface modification

Microstamp protein



Source: S. Schäfer, G. Wrobel

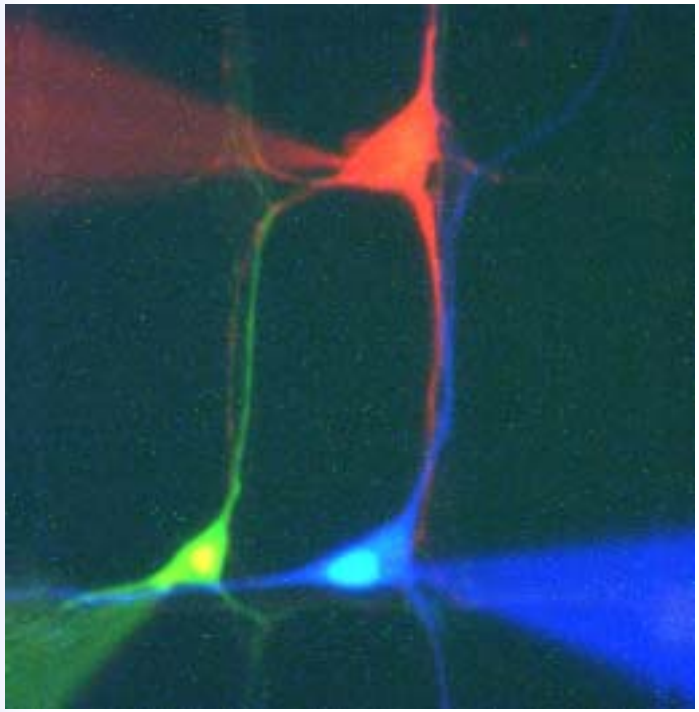
Stamp concept derived from the work of the Whitesides laboratory, e.g. Singhvi, R., Kumar, A., Lopez, GP, Stephanopoulos, GN, Wang, DIC, Whitesides, GM, and Ingber, DE (1994). Engineering cell shape and function Science 264, 696-698.

Neuronal networks

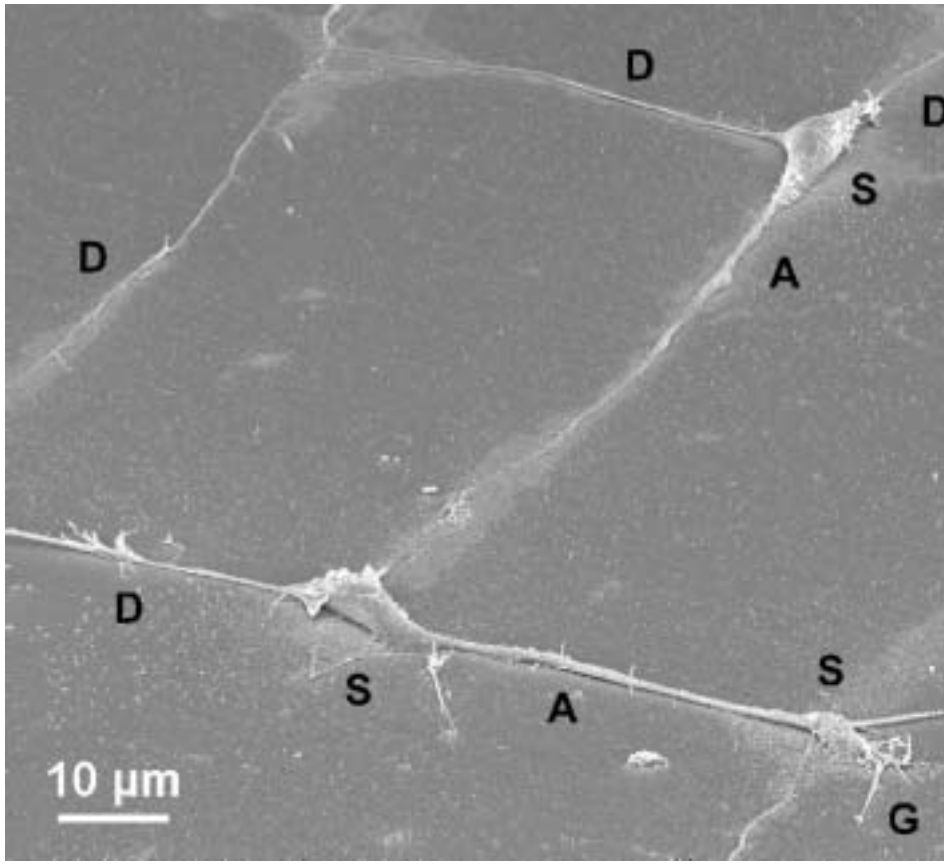
Morphology

Polar tracers can be used to visualize cellular connections:

Sulforhodamine, Cascade Blue, Lucifer Yellow



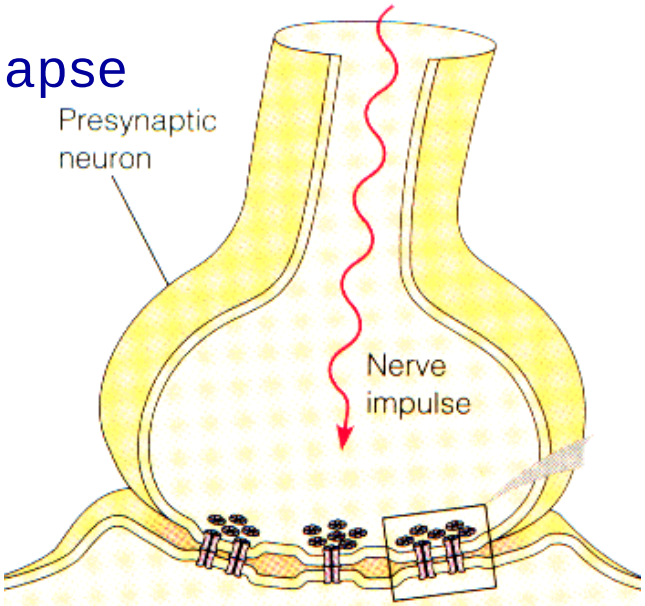
Combined fluorescent images of all three channels



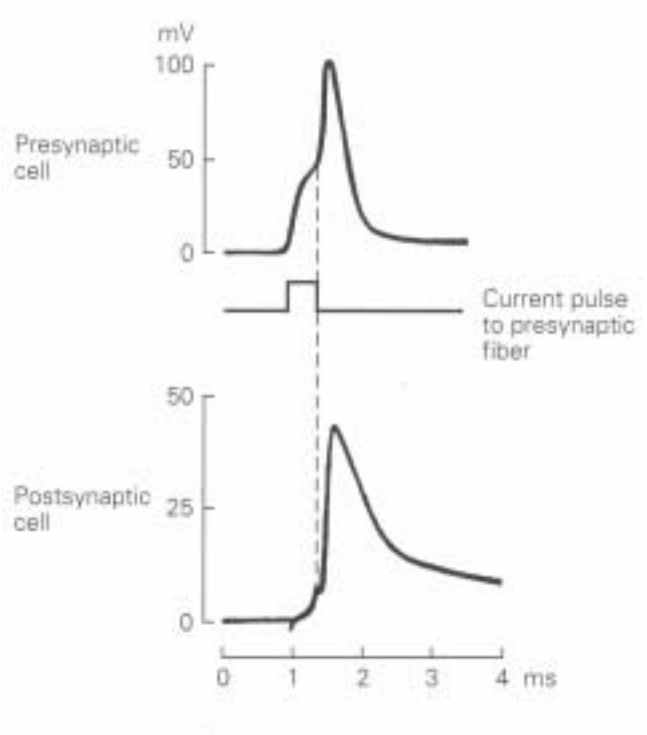
A. Vogt et al, (submitted)

Synaptic transmission in networks of neurons

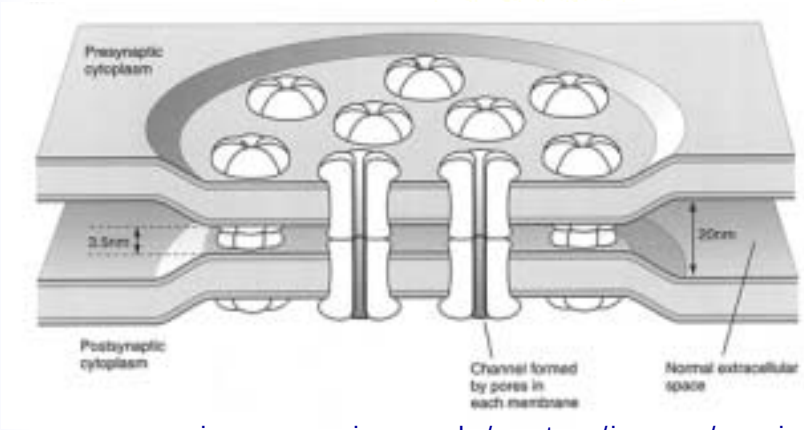
Electrical synapse



bio.winona.msus.edu/berg/illust/synapse1.gif



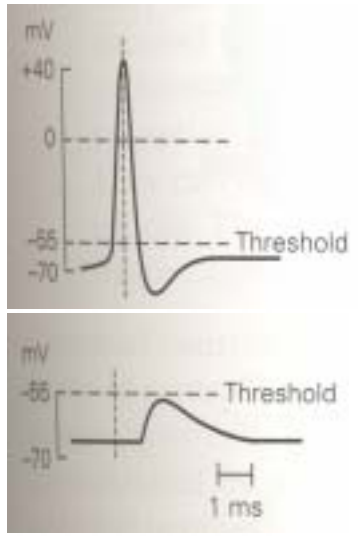
Principals of Neural Science, Kandel et al, 2000



www.consciousness.arizona.edu/qantum/images/gap.jpg

Synaptic transmission in networks of neurons

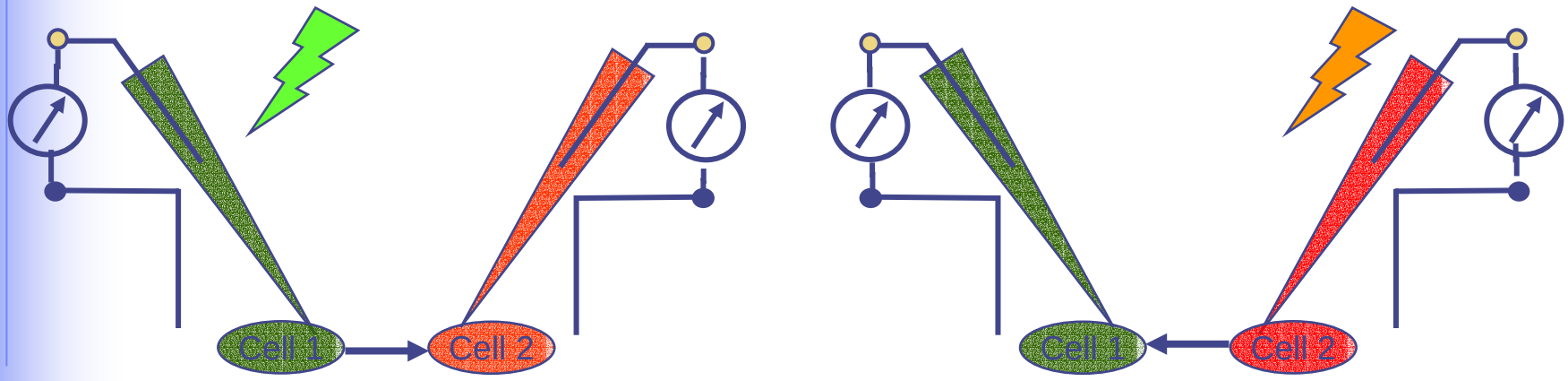
Chemical synapse



Principals of Neural Science, Kandel et al, 2000

bio.winona.msus.edu/berg/illust/synapse2.gif

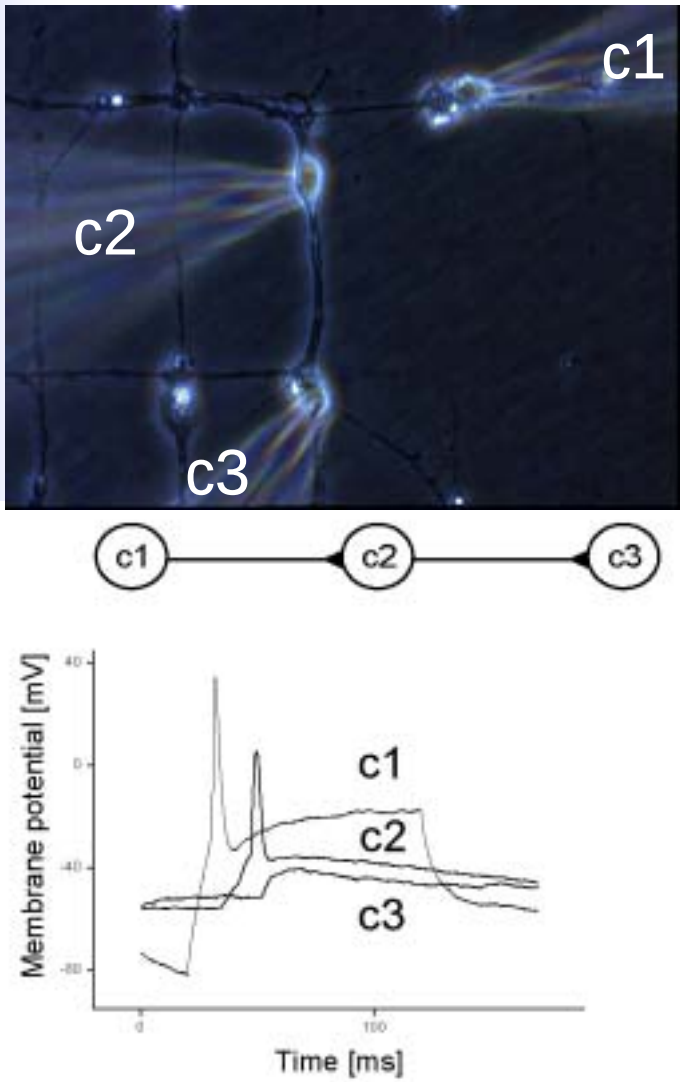
Experimental setup

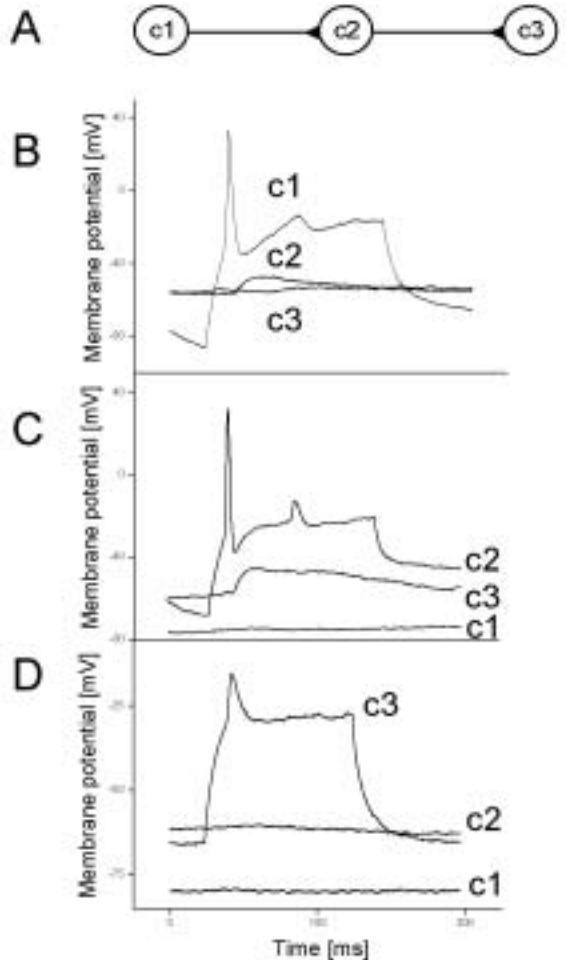


Neuronal networks

Chemical synapses

Cortical neurons on ECM-gel after 13 days in culture

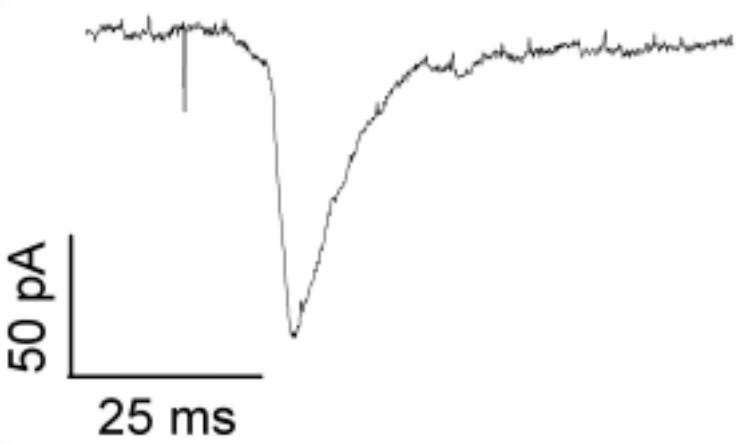




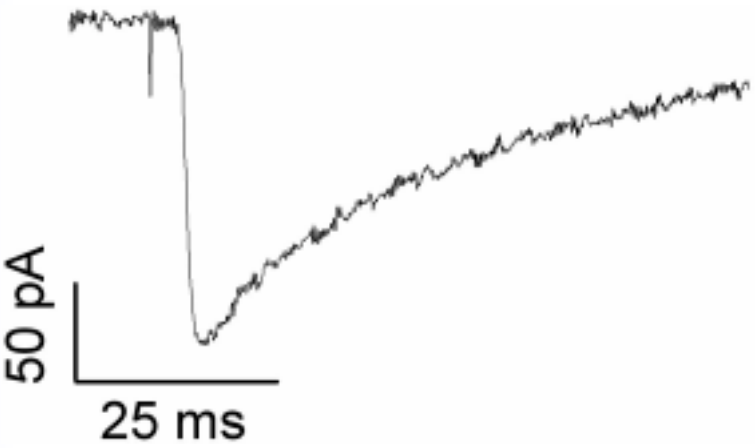
A. Vogt et al, Biotech. Progress (2003) 19, 1562-1568

Synaptic plasticity in networks of neurons

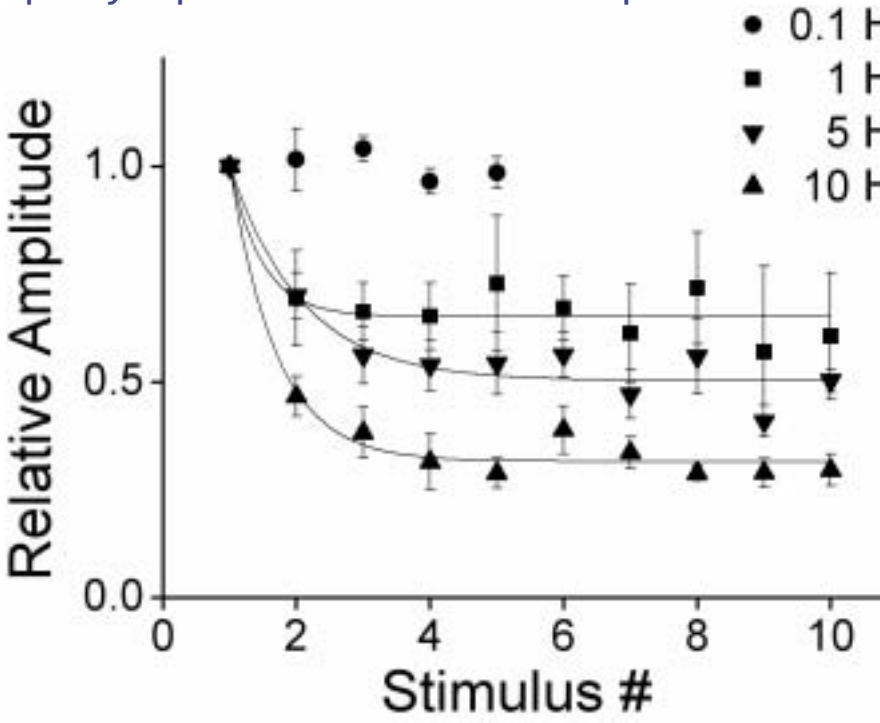
Excitatory synapses



Inhibitory synapses



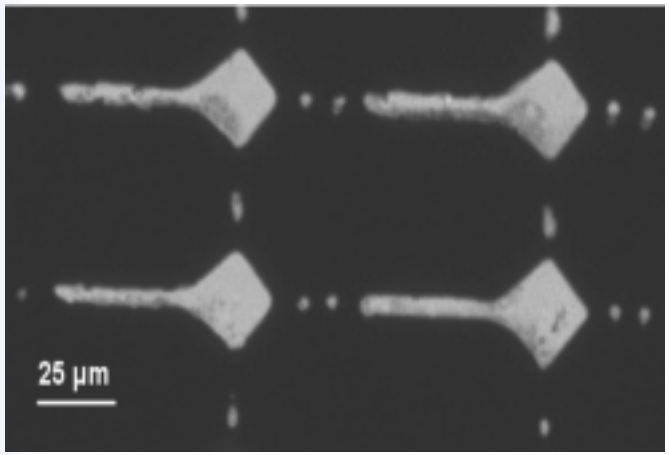
Inhibitory synapse challenged with presynaptic APs at different frequencies



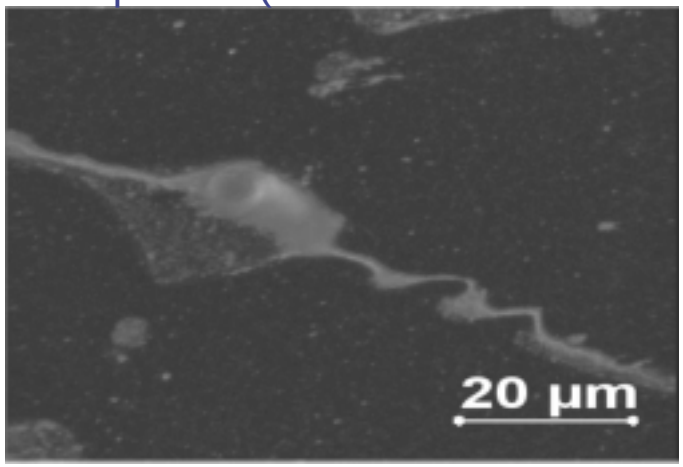
A. Vogt et al, (submitted)

Polarity in networks of neurons

Polarity inducing pattern (???)



Cortical neuron (12 DIV) on an interrupted micropattern (after mechanical stress)



	Gap size	Correctly oriented synapses [#]	Incorrectly oriented synapses [#]	Correctly oriented synapses [%]
A	1 µm	10	16	38.5
B	2 µm	3	5	37.5
C	5 µm	4	11	26.6
	Sum	17	32	35

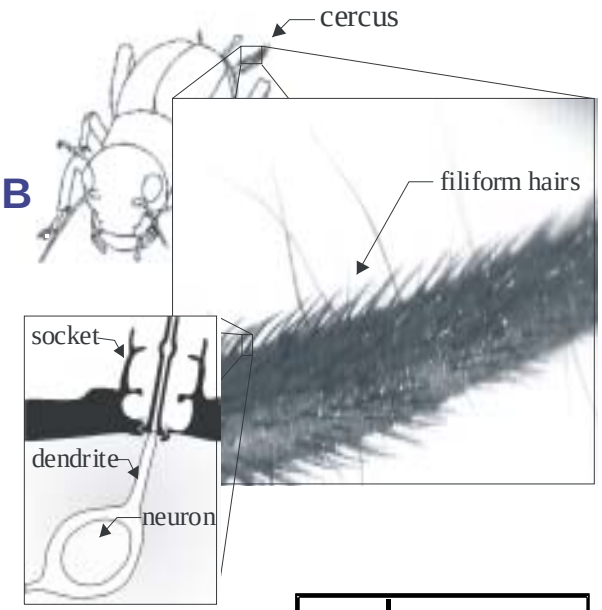
A. Vogt et al, J. Neurosci. Meth. 134 (2004) 191-19

CICADA:

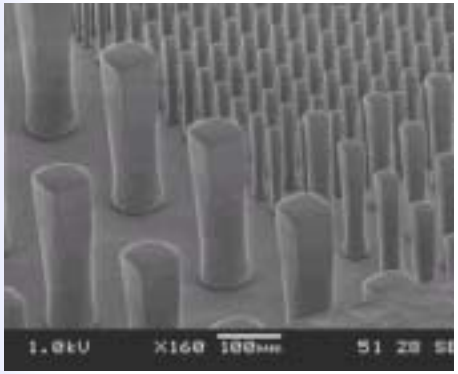
Wind-evoked escape reaction of crickets

University of Tours, F

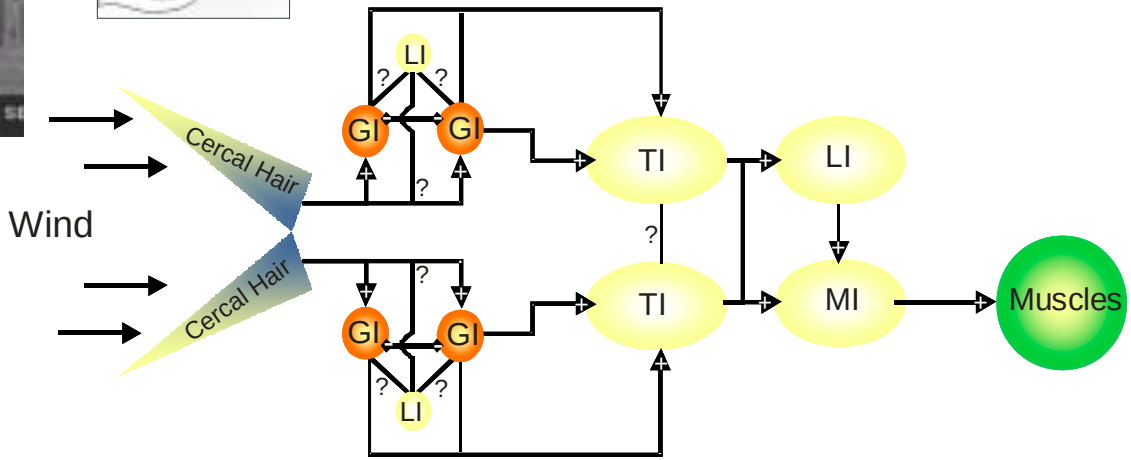
University of Reading, GB



Gryllus bimaculatus



University of Twente, NL

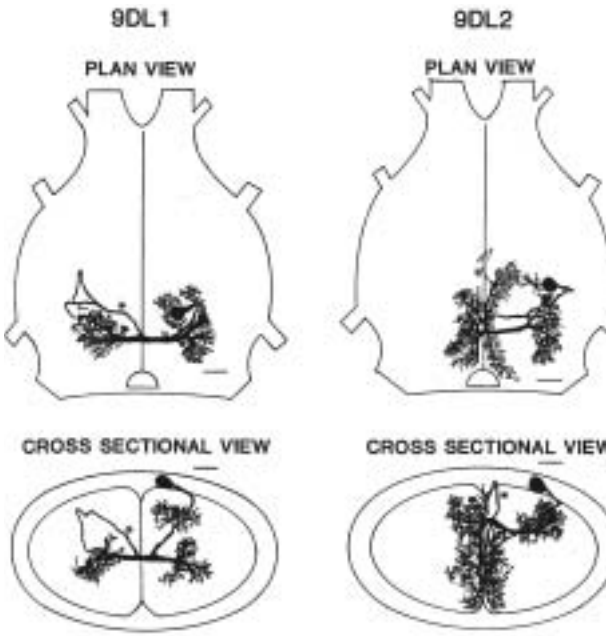


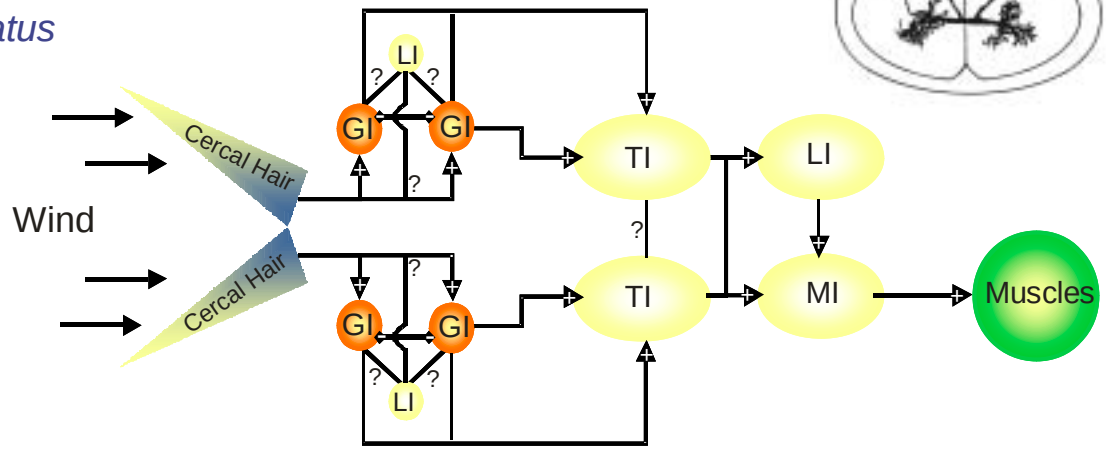
Forschungszentrum Jülich, D

Neuronal information processing

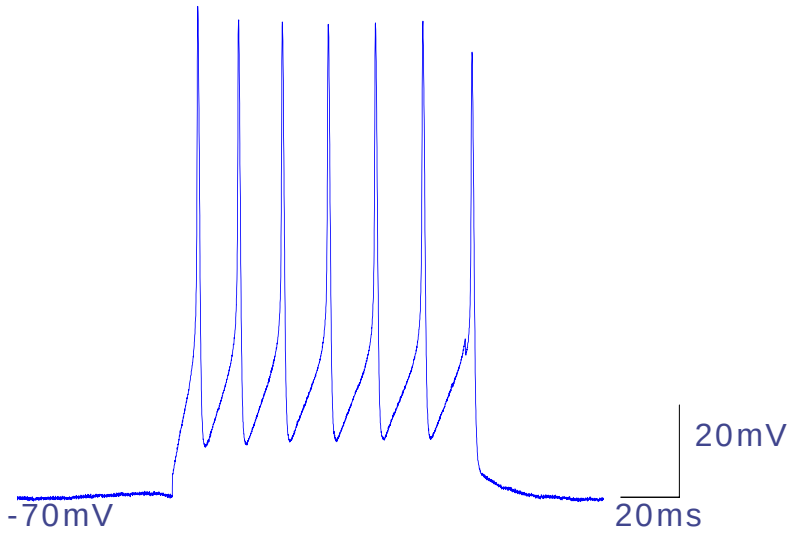
Neuronal circuits of insects

Gryllus bimaculatus





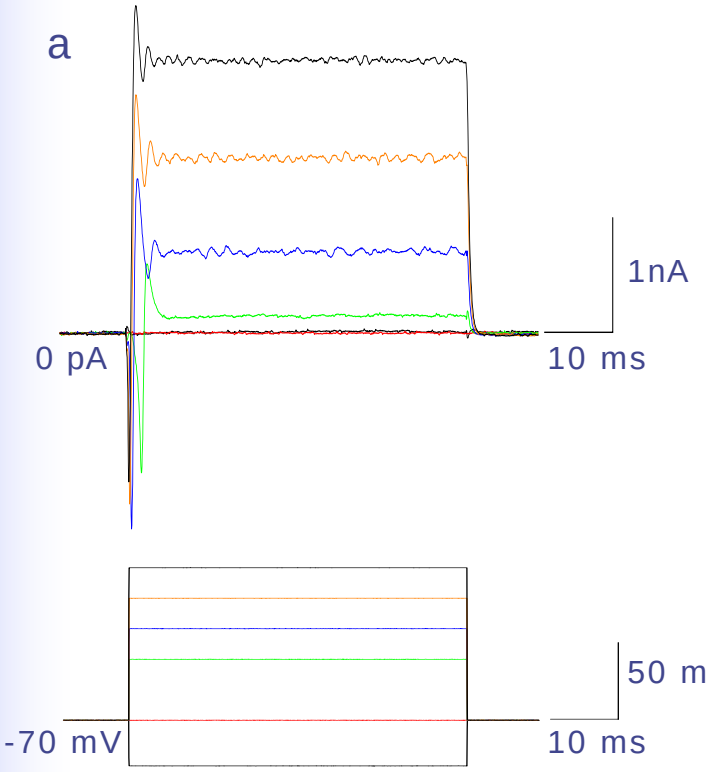
Electrophysiology – single insect cell

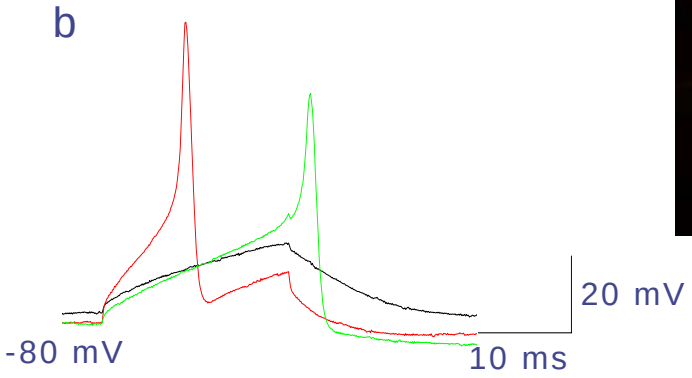


Source: P. Schulte, S. Weigel

Patch clamp recordings of a cricket neuron after 3 days in culture.
Current clamp mode. (v_{mem}: -57mV)

Backfill staining and electrophysiology



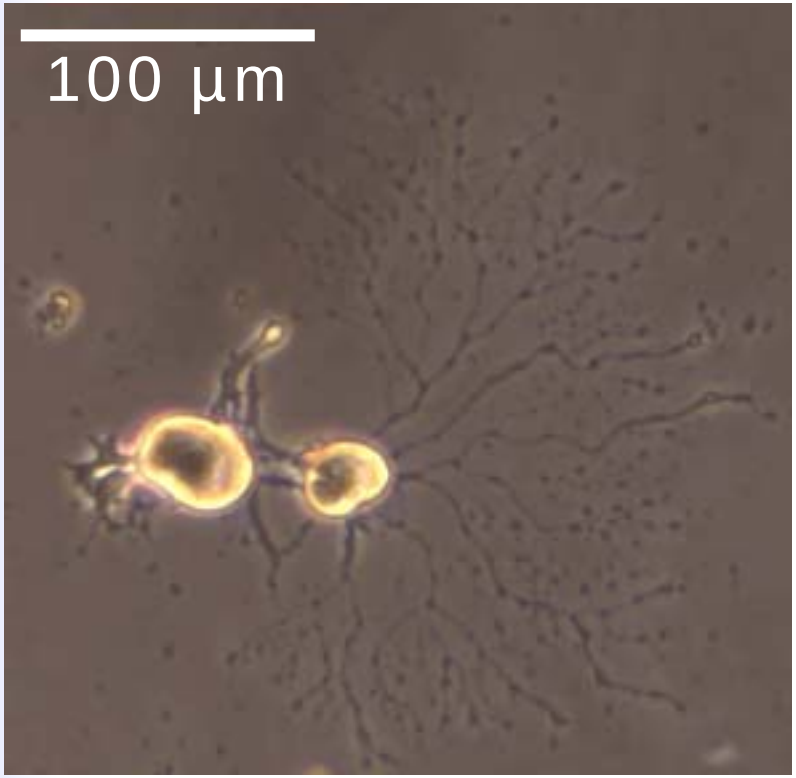


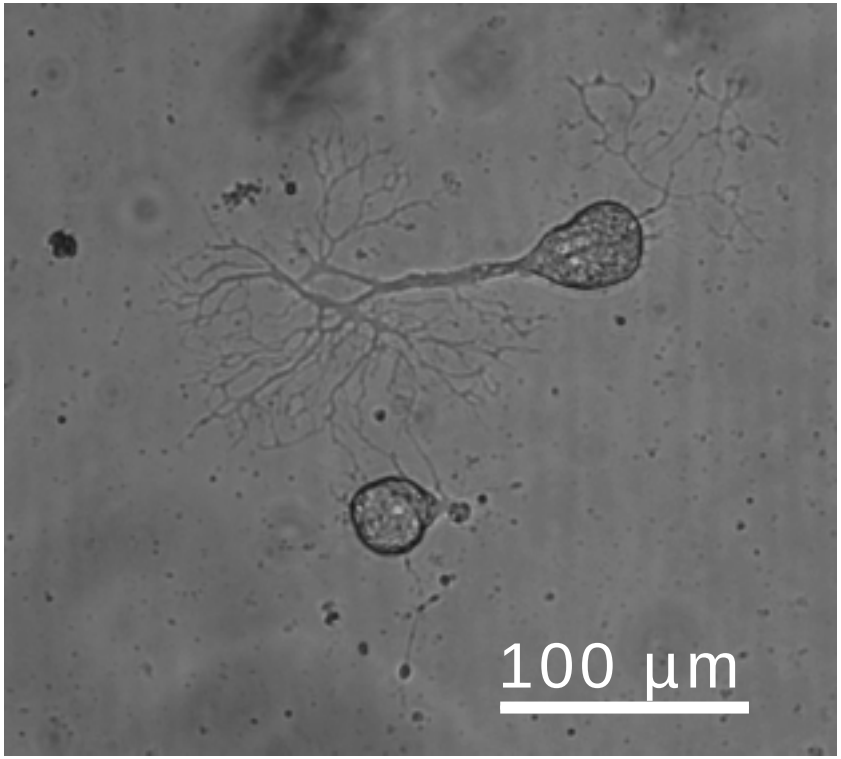
signal

Source: P. Schulte, S. Weigel

Patch clamp recording of a stained cricket neuron after 3 days in culture.
a: voltage clamp mode; b: current clamp mode. (v_{mem}: -58mV)

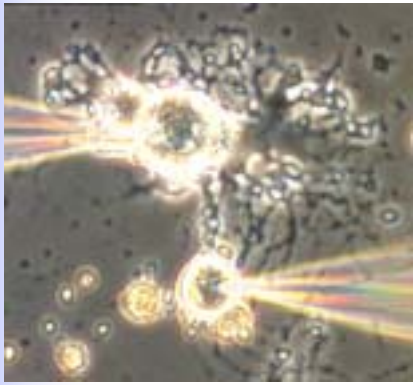
Outlook: neuronal circuits in insects





Source: P. Schulte, S. Weigel

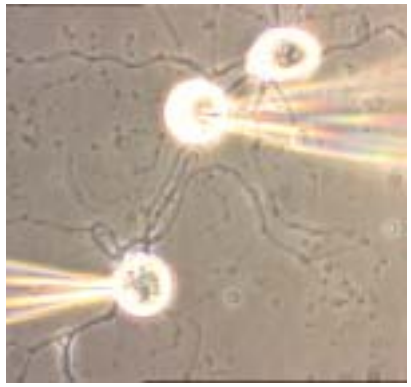
Electrophysiology of neuronal networks



Patch clamp recording (current clamp of simple network of 2 locust neurons after 8 days in culture. (cell1:v_{mem} -42mV; cell1:v_{mem} -35mV)

Source: P. Schulte, S. Weig

Electrophysiology of neuronal networks

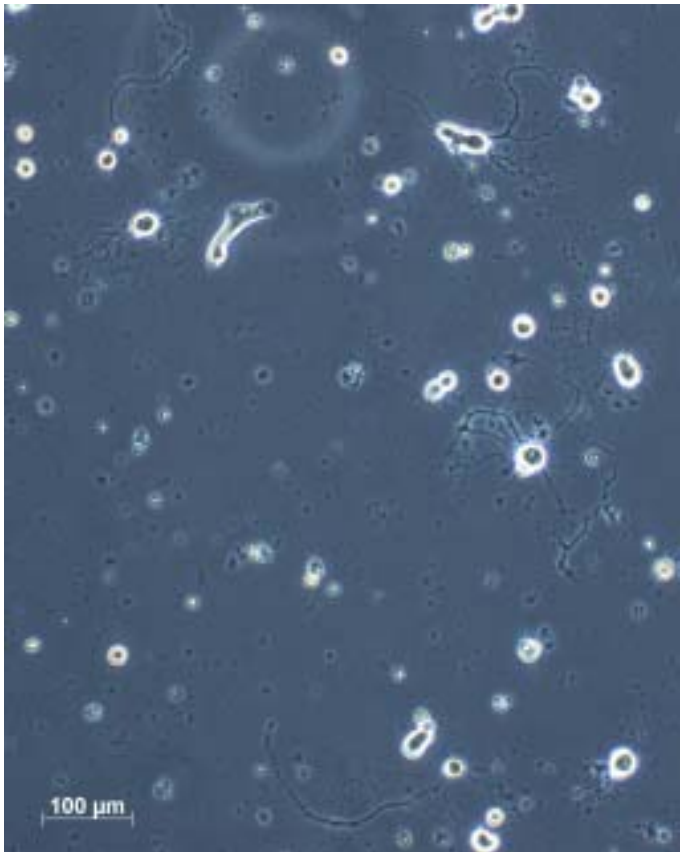


Patch clamp recording (current clamp) of simple network of cricket neurons after 8 days in culture. (cell1:v_{mem} -42mV; cell1:v_{mem} -35mV)

Source: P. Schulte, S. Weigel

Searching cell repulsive substrate

Cricket neurons plated on PEG-coated surface after 6 days in cell culture. None of the plated neurons adhered. PEG provided by the group of Prof. Spatz, Heidelberg.

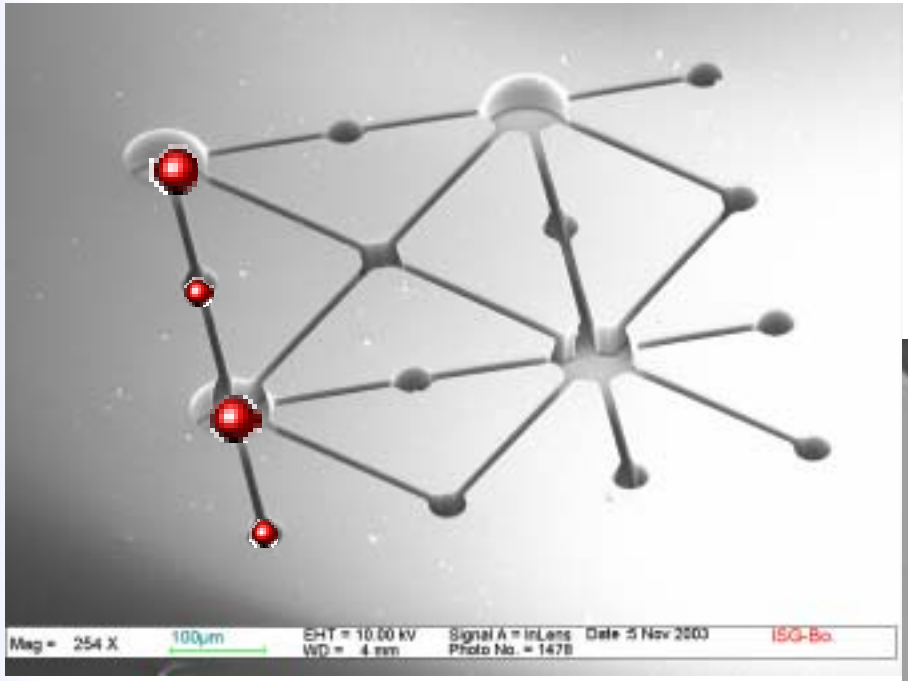


Cricket neurons on glass after 6 days in cell culture (control).

Source: P. Schulte

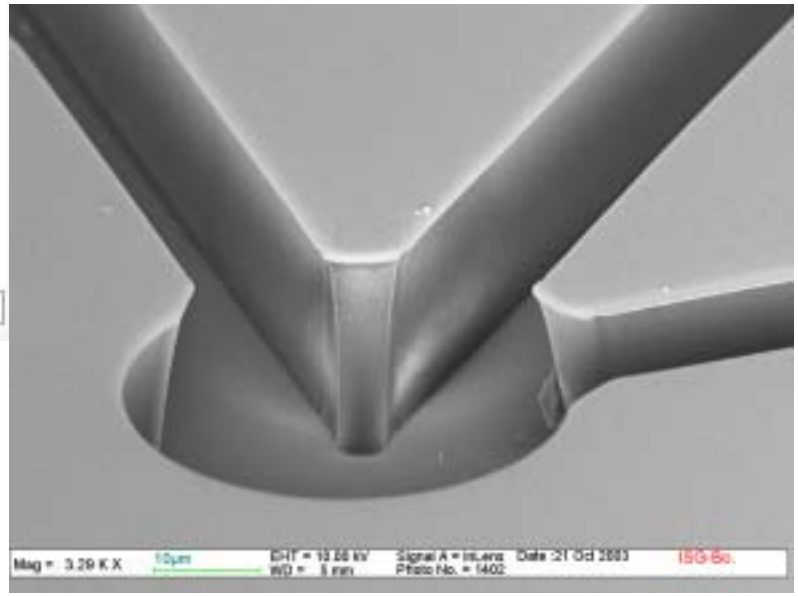
Defined neuronal circuits in insects

Microchannels for the guidance of cell outgrowth
Positioning of single (identified) neurons into the holes



SU-8 structure

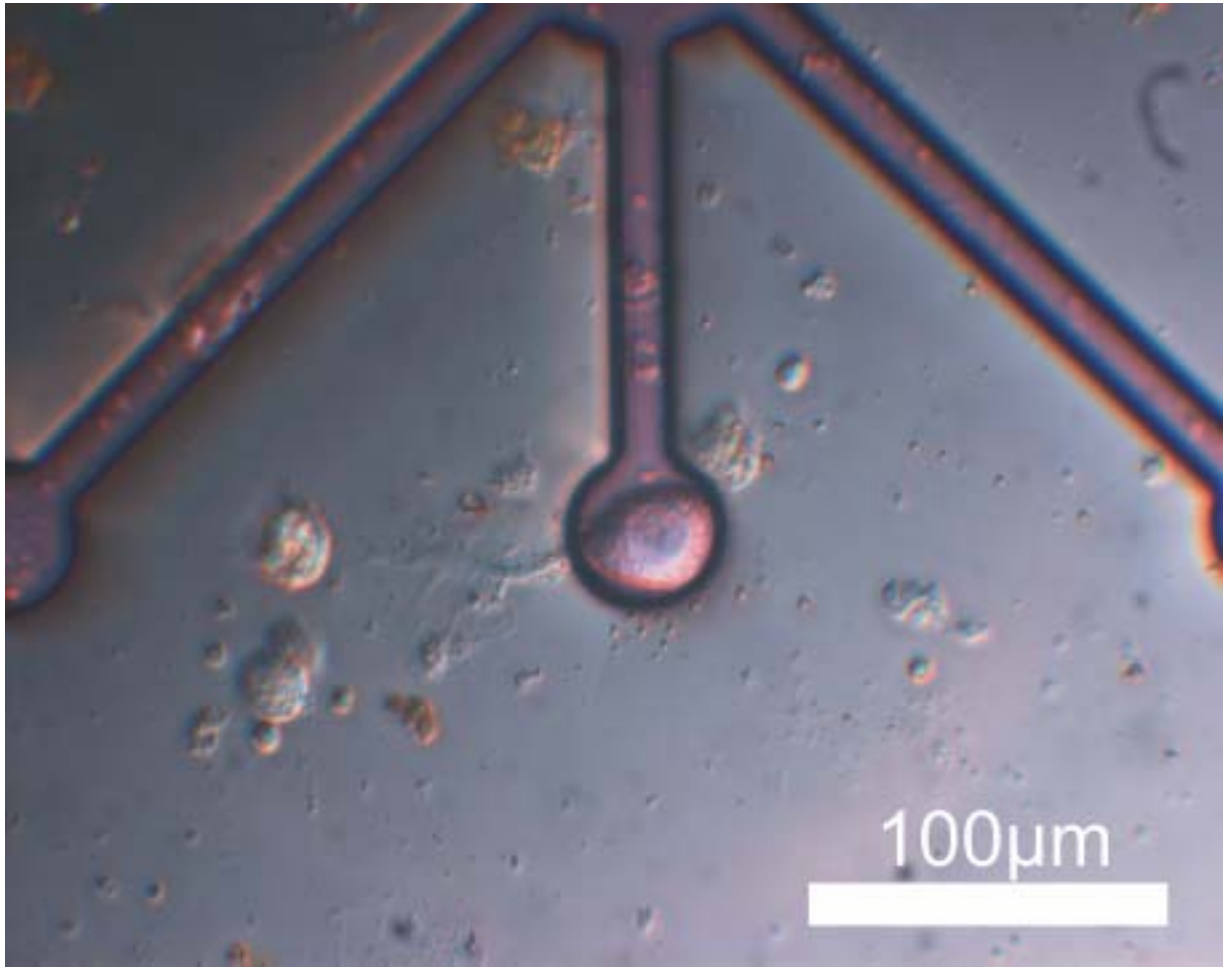
Concept derived from the work of the Fromherz and the Fujita laboratory e.g. Griscom, L.; Degenaar, P.; LePioufle, B.; Tamiya, E.; Fujita, H. *Jpn. J. Appl. Phys.* **2001**, *40*, 5485-5490; Merz, M.; Fromherz, P. *Adv. Mater.* **2002**, *14*, 141.



Source: A. Reska, Y. Mourzina, R. Stockmann

Defined neuronal circuits in insects

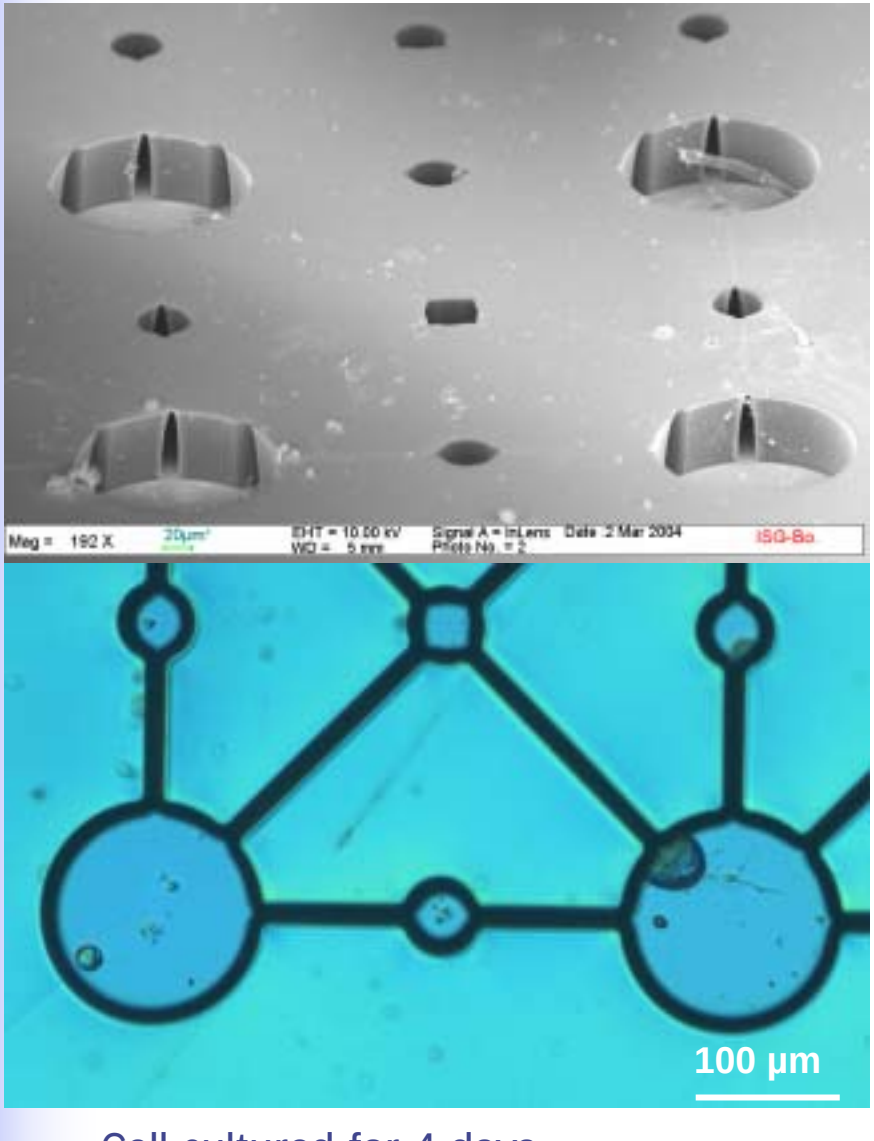
Network formation



Source: A. Reska

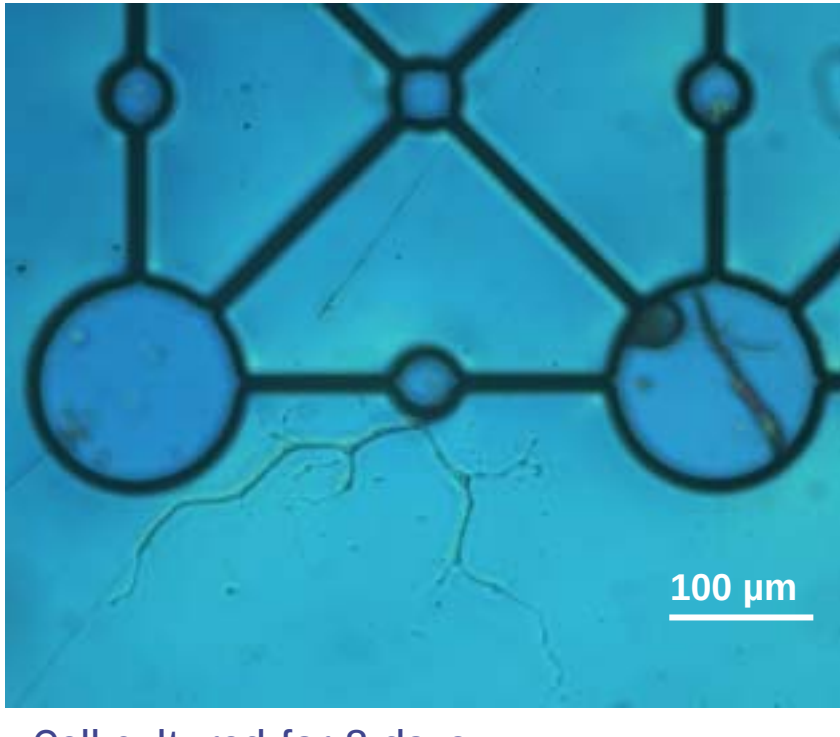
Neuron on a concanavaline A coated SU-8 structure after 5 days in culture

Defined neuronal circuits in insects



Cell cultured for 4 days

Towards network formation

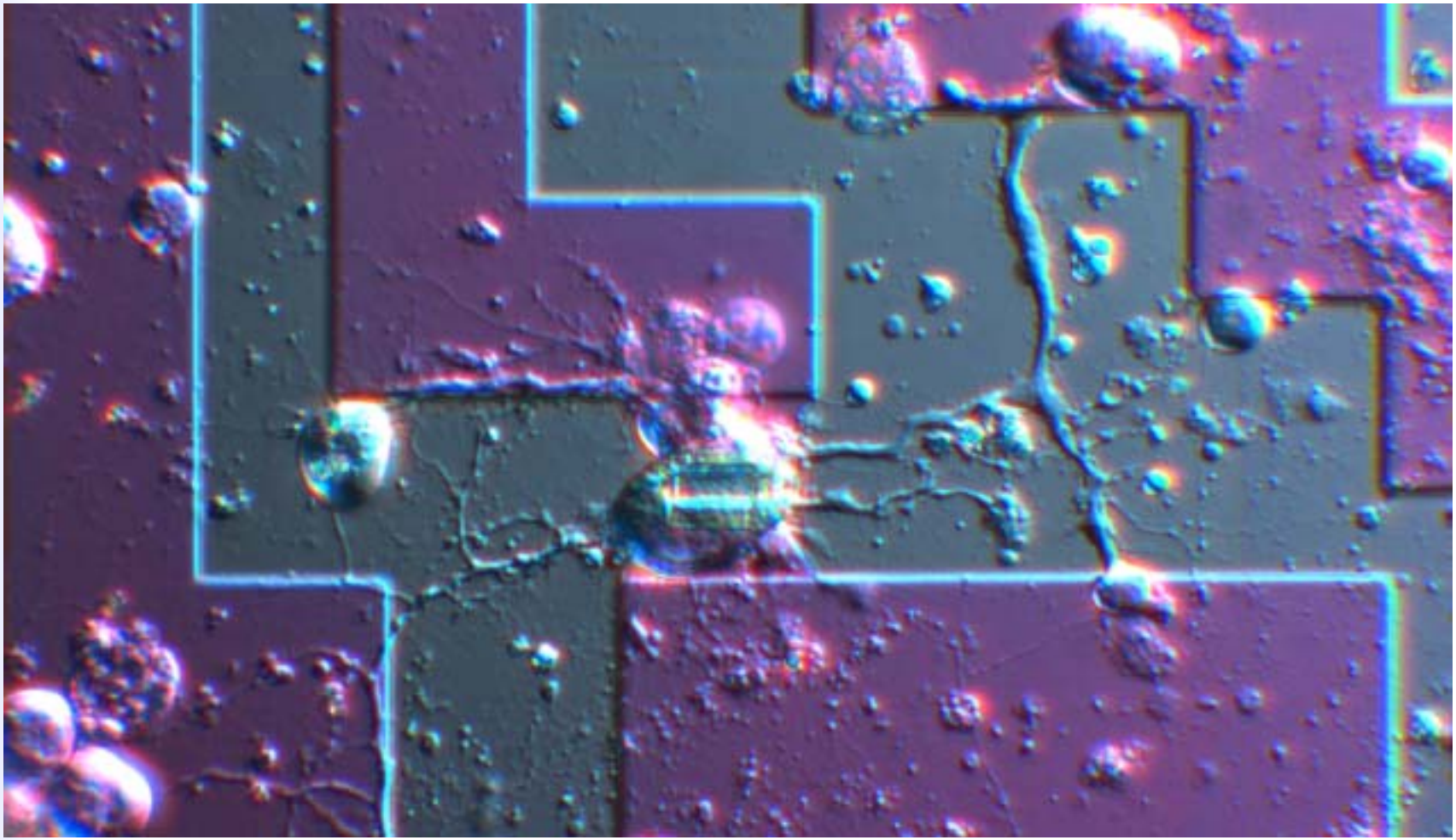


Cell cultured for 8 days

Source: A. Reska

Extracellular signal recordings

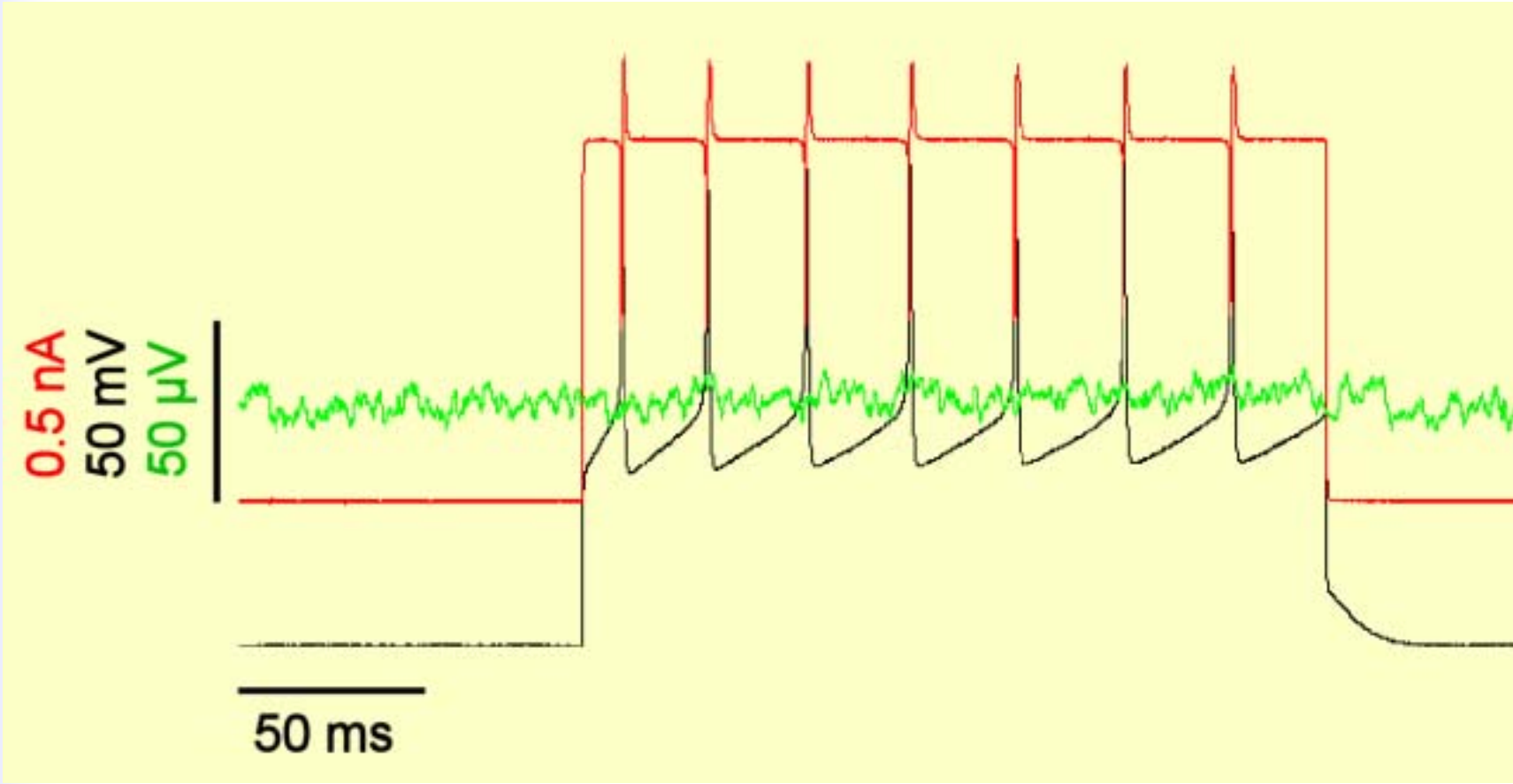
Cricket neuron on FET



Source: G. Wrobel, S. Weig

Extracellular signal recordings

Extracellular recordings of insect neurons with FETs

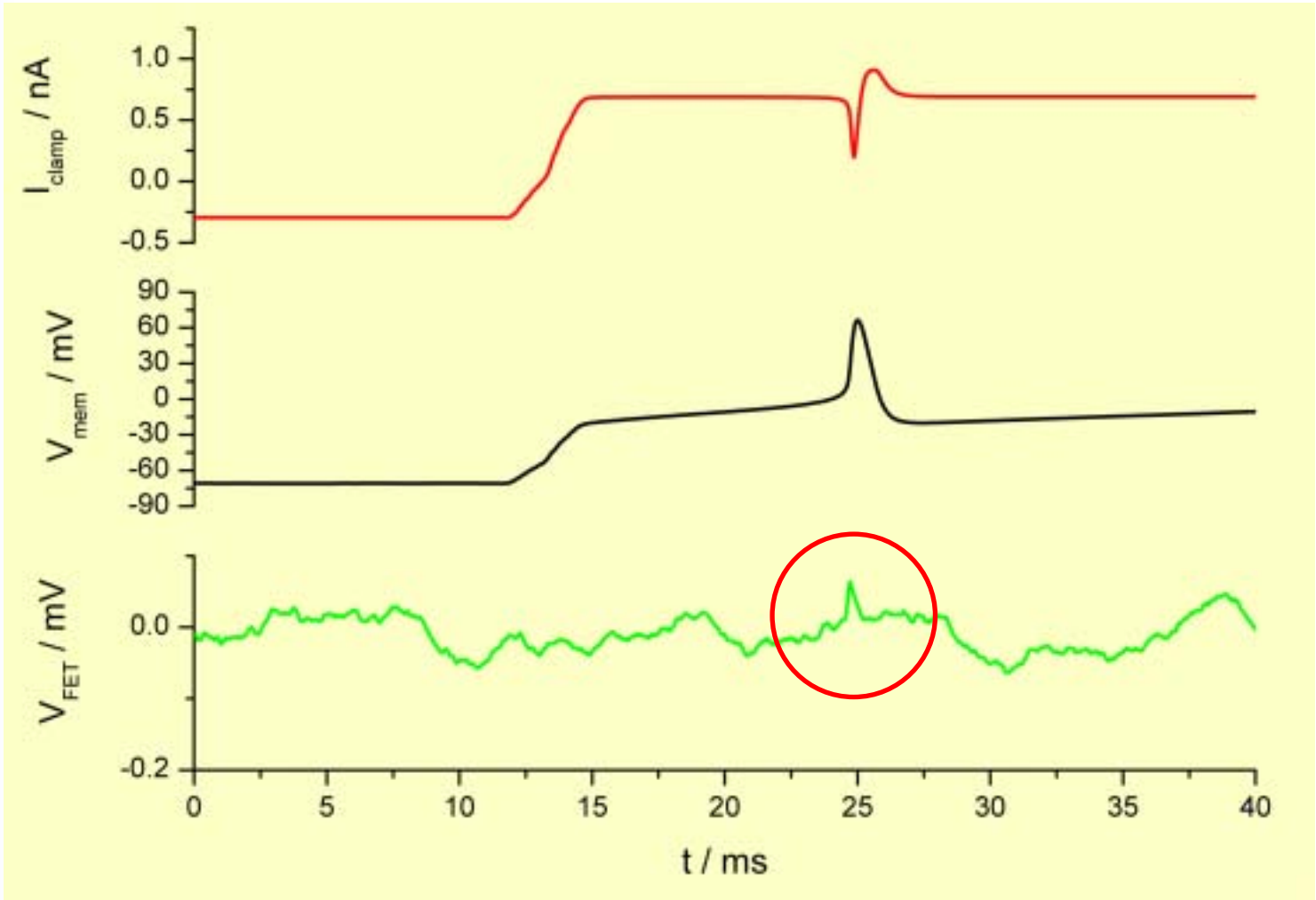


action potentials of Locust cell (cc-mode)

Source: G. Wrobel, S. Weig

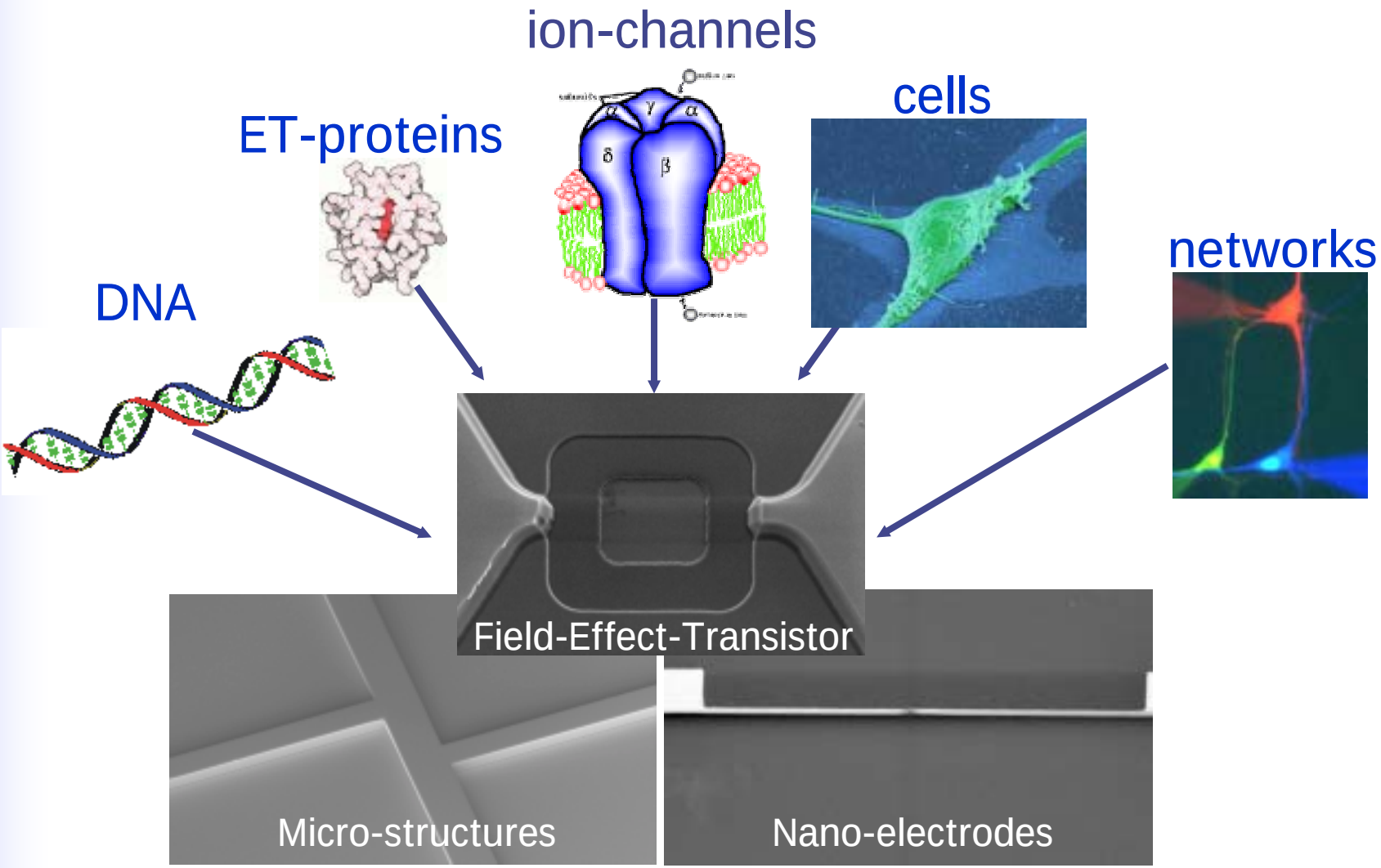
Extracellular signal recordings

Extracellular recordings of insect neurons with FETs



Source: G. Wrobel, S. Weig

Bioelectronic hybrids



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and co-workers
Inst. Biol. Information Processes (IBI-2)
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Prof. H. Wagner

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http://www.fz-juelich.de/isg/ISG2

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Inst. Thin Film & Interfaces (ISG-2):

Biomolecular Electronics:
Dr. Dirk Mayer, Oliver Salomon, Stefan Schaal, Daniel Schwab

Electronic DNA-Chip:
Dr. Sven Ingebrandt, Oleksandr Belinski, Yinhua Han

Cell-Device-Coupling:
Dr. Sven Ingebrandt, Dirk Borstlap, Sven Meyburg, Regina Stockmann, Dr. Günter Wrobel, Prof. Yi Zhang

Cellular-Networks:
Dr. Simone Böcker-Meffert, Tanja Decker, Rita Helpenstein, Anna Reska, Dr. Petra Schulte, Stefan Weigel

Microfluidics:
Dr. Yulia Mourzina

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Dr. Jochen Krause, Dieter Lomparski

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