Визуализация физиологической активности головного мозга: оптические методы.

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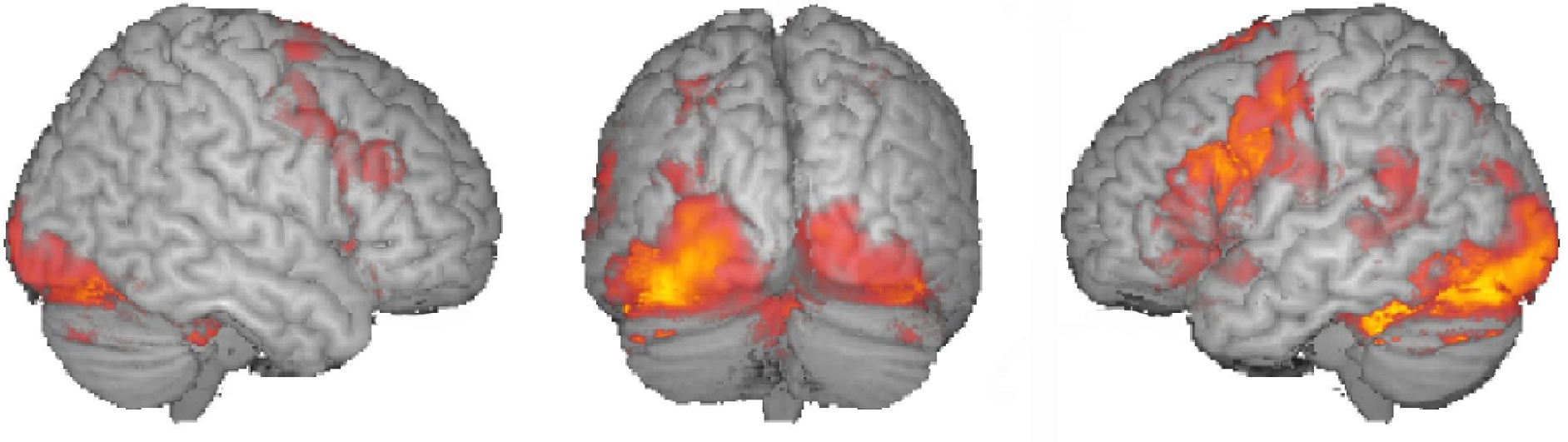
The base:

- Localization of functions is one of the central question in fundamental and clinical neuroscience

- Functional imaging gives us detailed information about the location of brain activity.

- The light absorption, scattering and fluorescence is highly responsive to functional changes in neural tissue.

Functional Magnetic Resonanse Imaging (fMRI)

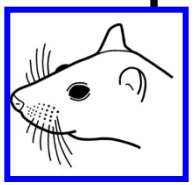


fMRI:
Golden Standard of the Functional Brain Mapping

Brain Optical Imaging: Tasks and Abilities

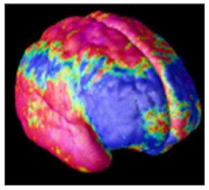
- **Intrinsic Optical Imaging:** Indirect Representation of the Neural Activity by the example of Auditory Cortex

- **Extrinsic Optical Imaging (including Voltage-Sensitive Dye) :** Direct Representation of the Neural Activity by the example of Auditory Cortex and Barrel Field



- **Photoacoustic Imaging:** Physical Base and Biological Application

- **Other methods:** Animal Models and Vizualization



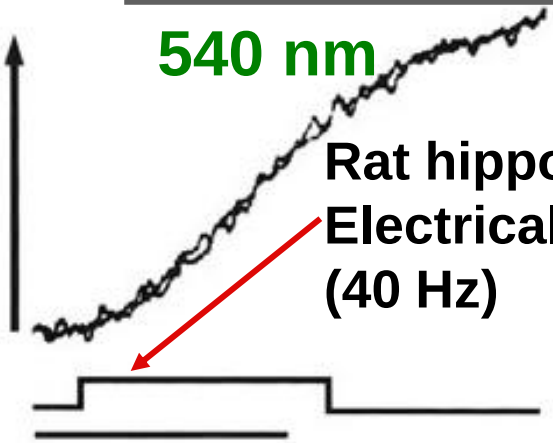
Optical Imaging Intrinsic Signal (IOS)

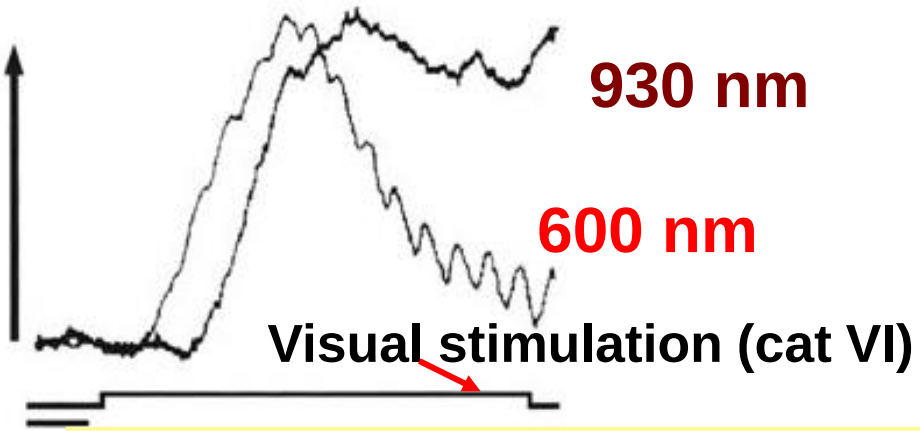
1. **Based on the monitoring intrinsic activity-related changes in tissue reflectance.**

2. **Offers a distinct advantage over extrinsic signal imaging, which may cause phototoxicity**

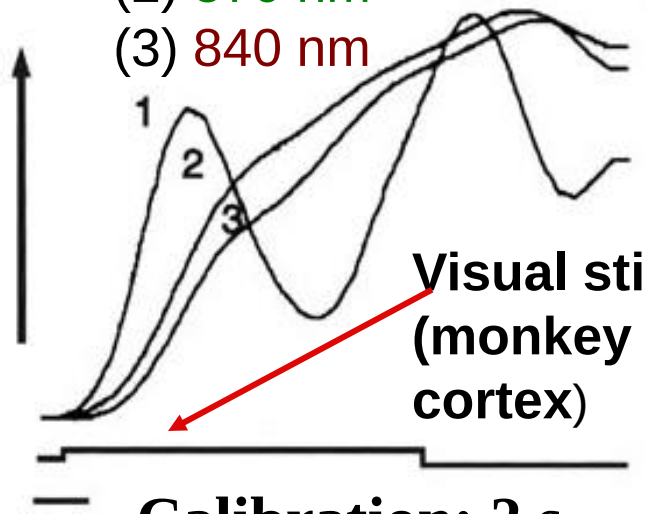
3. **Not requiring any contact with the tissue of interest.**

Intrinsic Optical Signal: Depending Time Course of Wavelengths





(1) 600 nm
(2) 570 nm
(3) 840 nm



Main sources of Intrinsic optical signal:

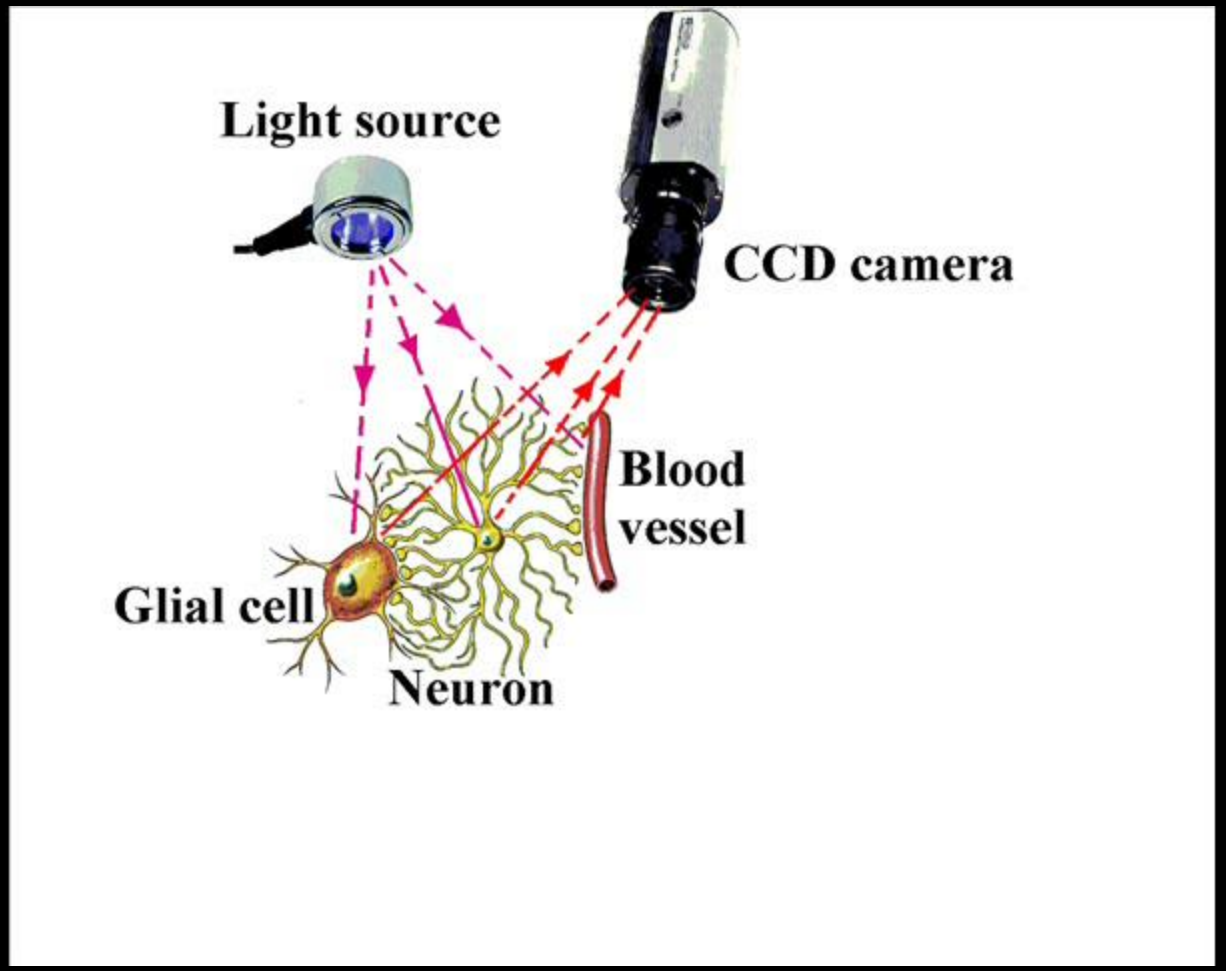
1. An increase in the deoxyhemoglobin concentration, resulting from elevated oxygen consumption of the neurons due to their metabolic activity

2. Second component is a delayed one: an activity related increase in blood flow, causing a decrease in the deoxyhemoglobin concentration

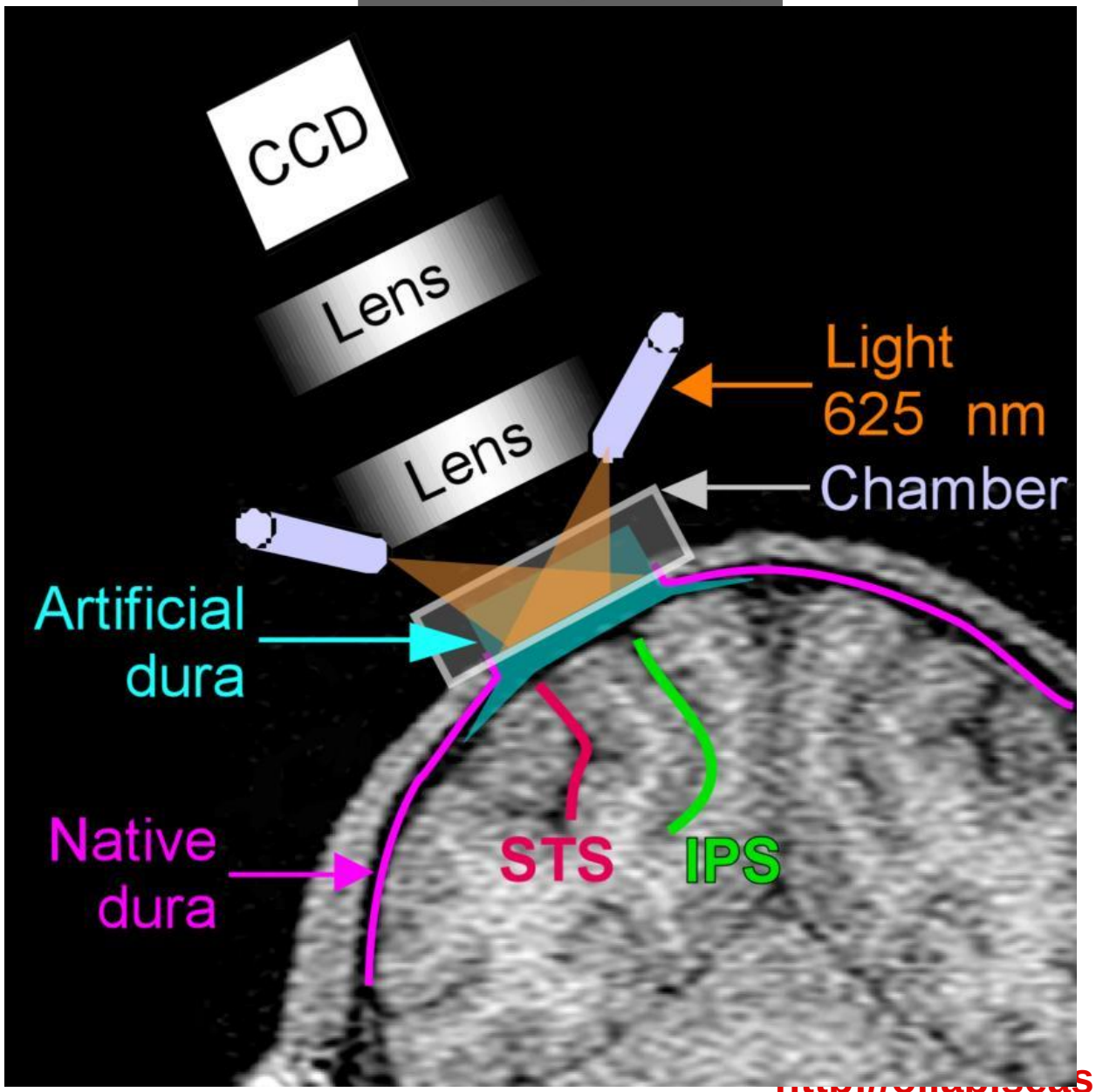
3. Light scattering

(Grinvald *et al.* 2001. In-vivo Optical imaging of cortical architecture and dynamics)

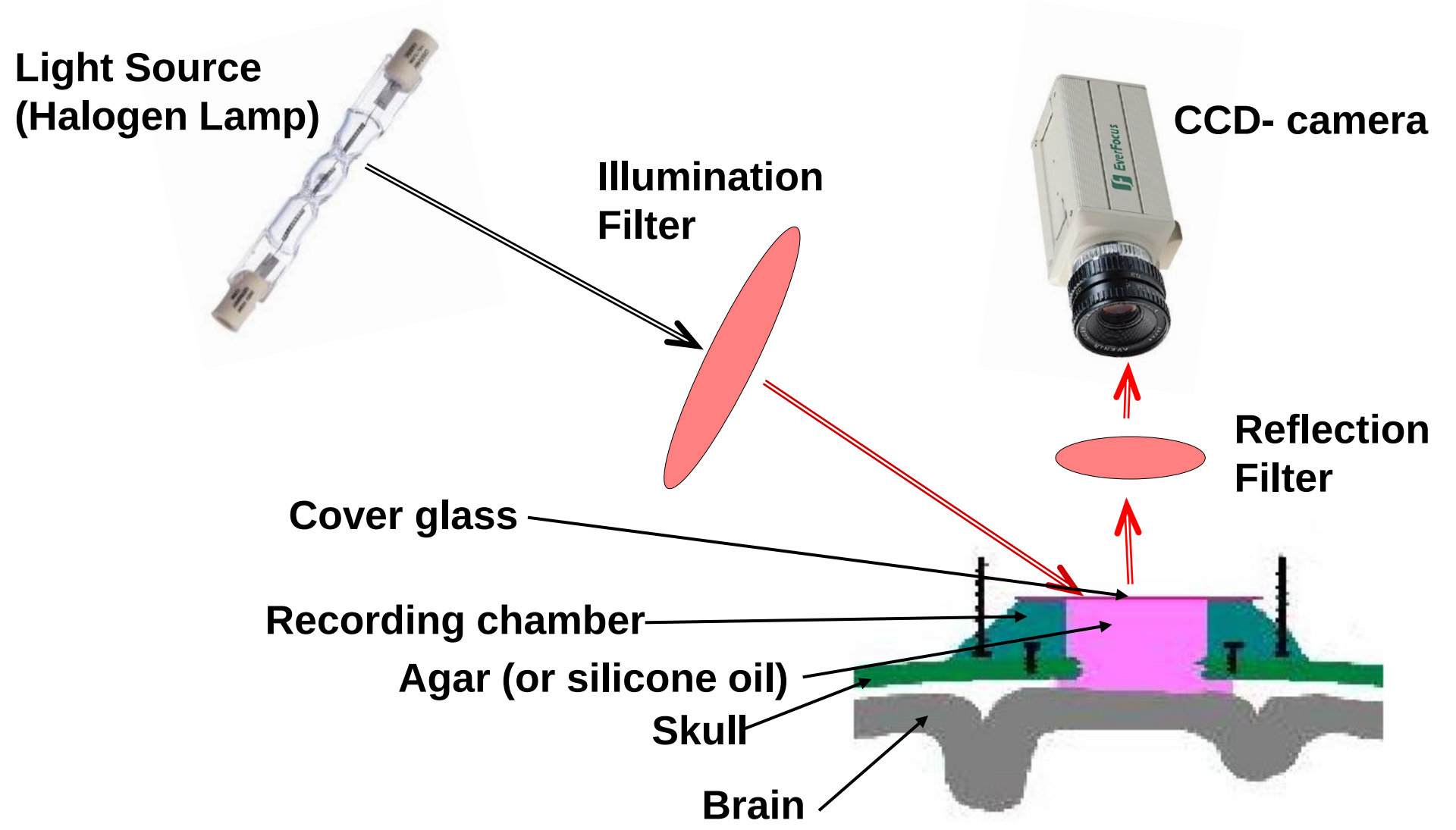
Intrinsic Optical Imaging



Imaging Setup

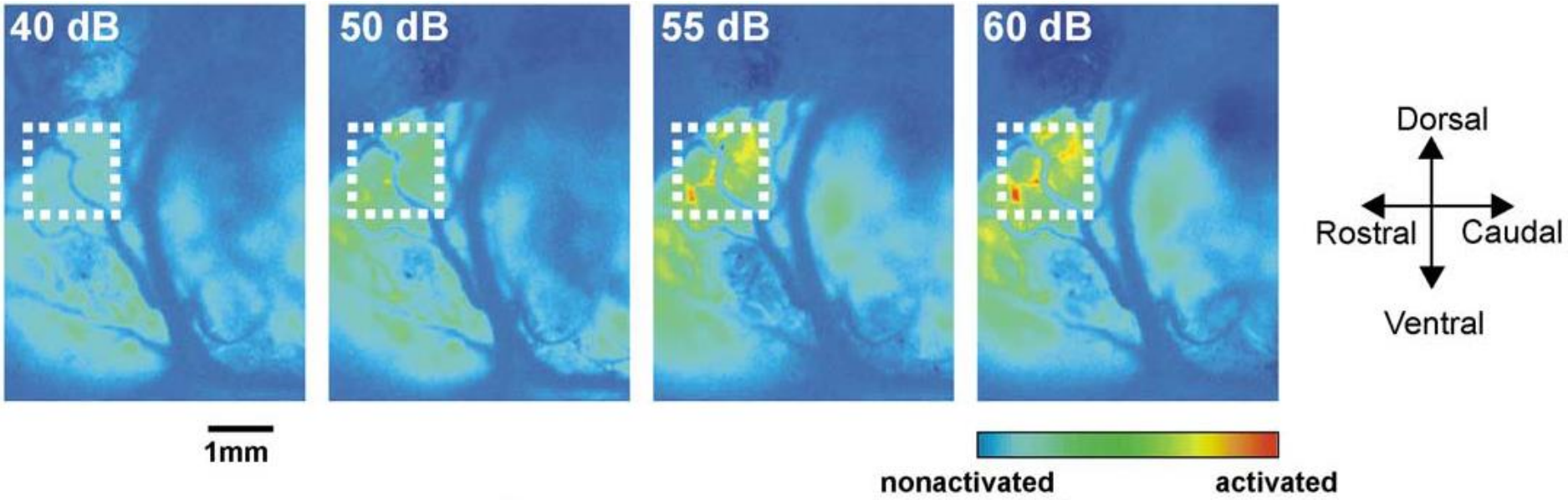


Principle of Intrinsic Optical Imaging (IOS)



(Grinvald *et al.* 2001. In-vivo Optical imaging of cortical architecture and dynamics)

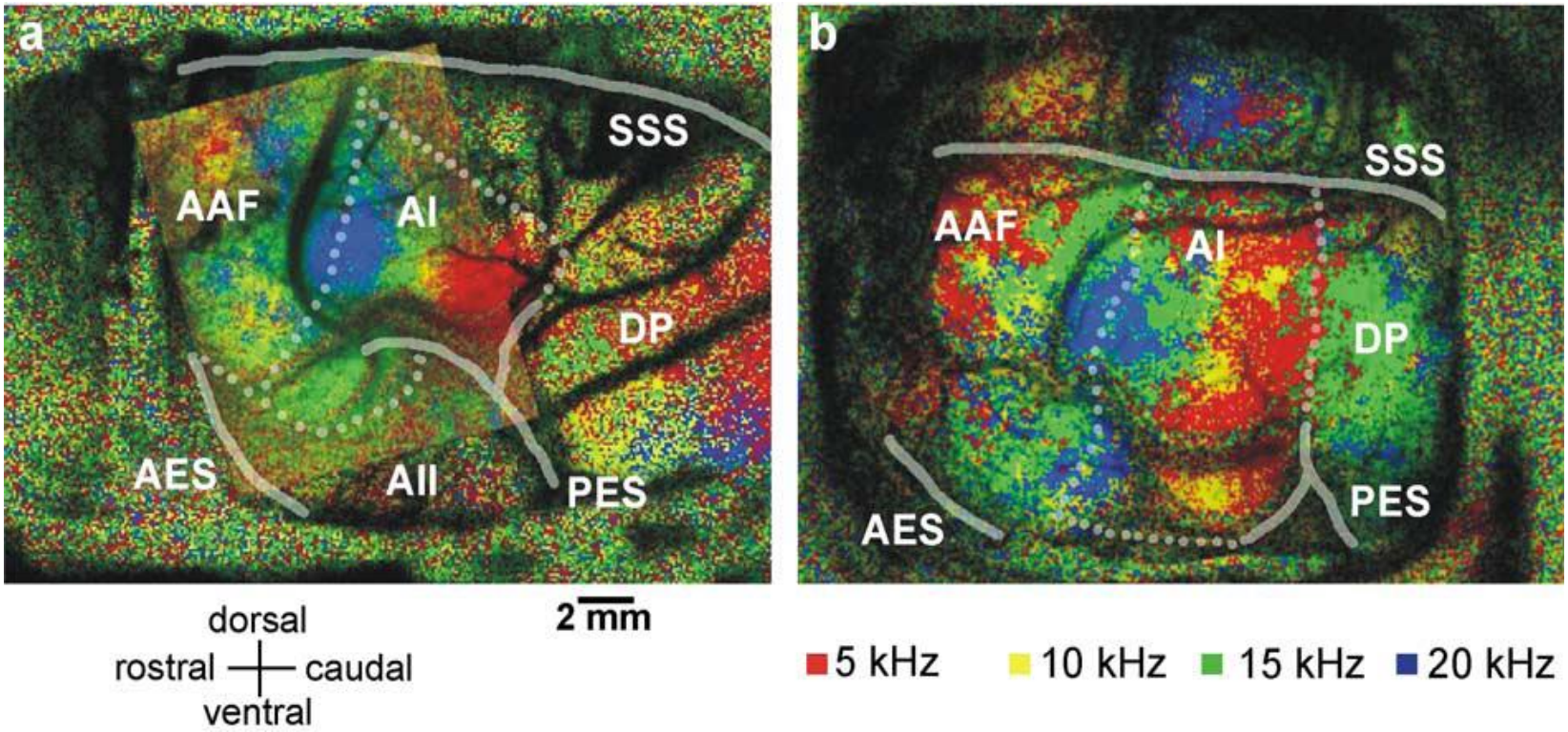
Intrinsic Optical Imaging of Auditory Cortex



Dependence of Signal Strength of Pure Tone Binaural Stimulation (2 kHz) from Stimulus Intensity

(Tsytsarev et al, 2004. Sound frequency representation in cat auditory cortex)

Tonotopicity of Auditory Cortex: IOS data

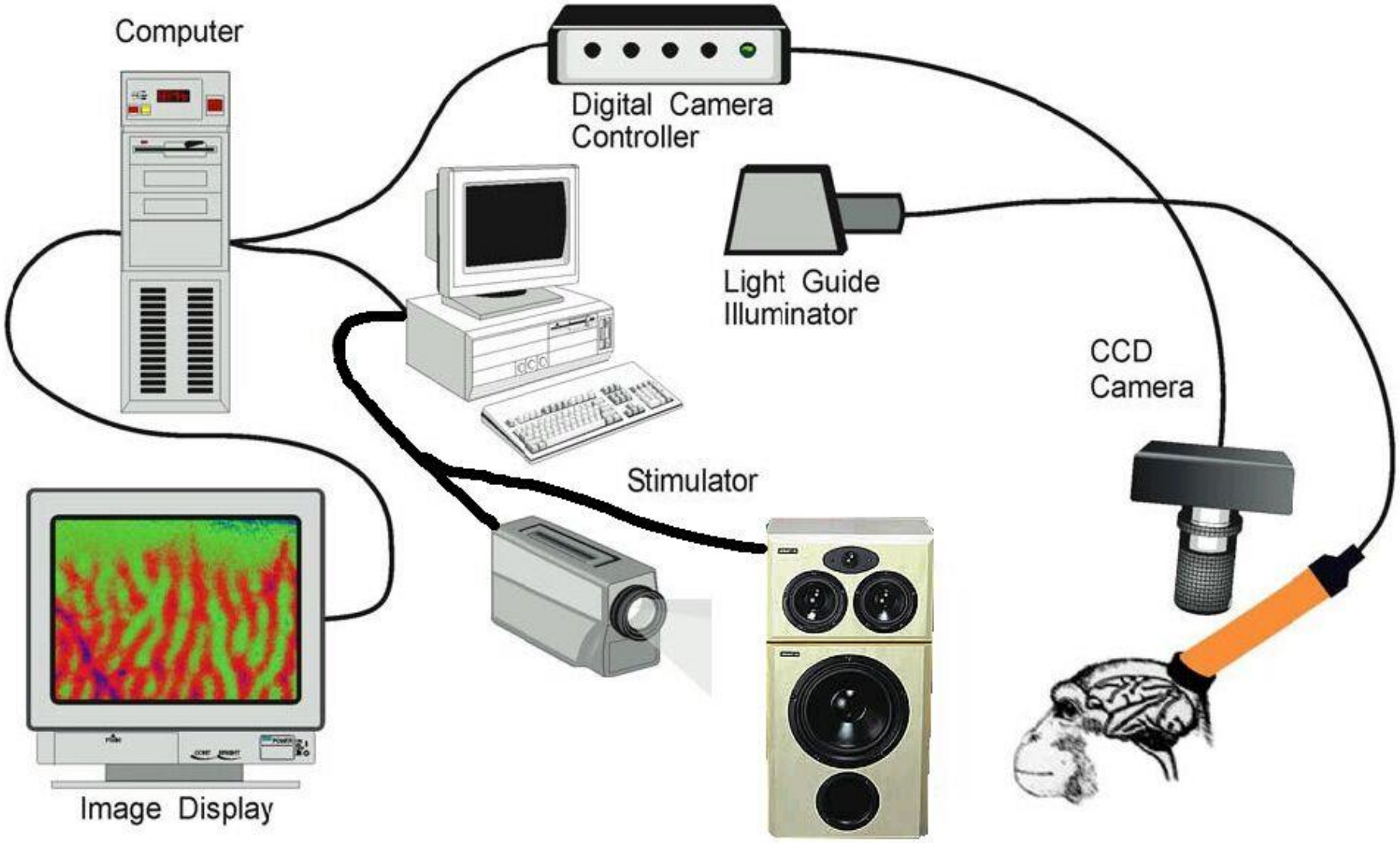


AAF: anterior auditory field
AI: primary auditory field
AII: secondary auditory field

SSS: suprasylvian sulcus
DP: dorsoposterior auditory field
AES: anterior ectosylvian sulcus
PES: posterior ectosylvian sulcus

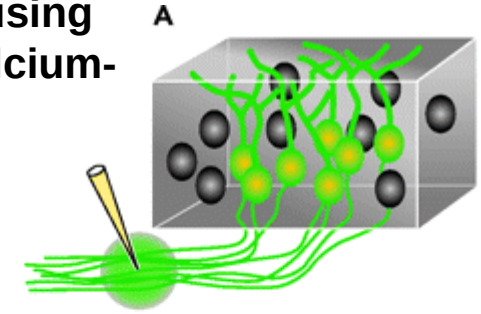
(**Tsytsarev et al, 2004.** Sound frequency representation in cat auditory cortex)

IOS: Functional Brain Mapping Application

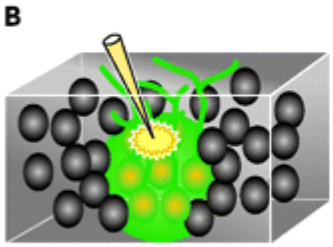


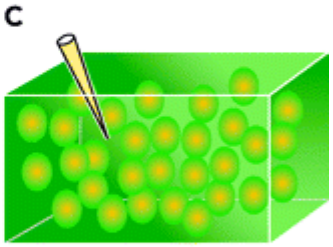
Ca-sensitive Fluorescence Dye Application

A:retrograde labeling using dextran-conjugated calcium-sensitive dye.



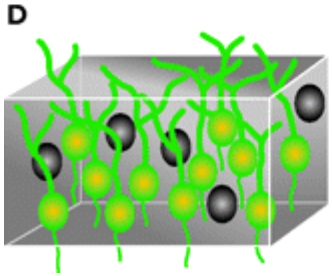
B:labeling of a subset of cells using local electroporation.

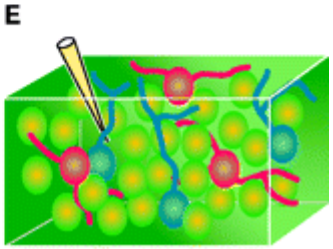




C:multi-cell bolus loading (MCBL) by injection of calcium indicator; results in unspecific cell loading.

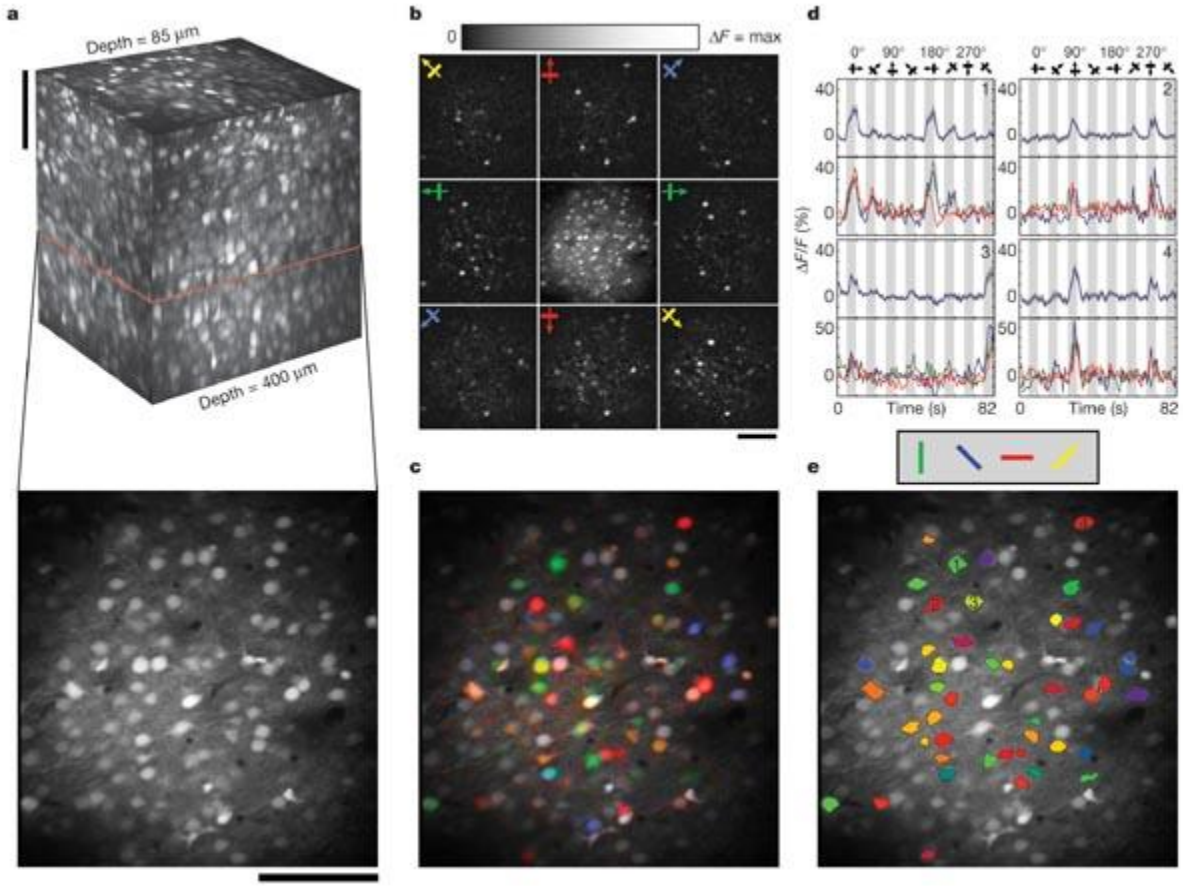
D: expression of a genetically encoded calcium indicator in a particular cell type.





E:multi-wavelengths labeling by combining MCBL with specific counterstains. Various fluorescent dyes can help to distinguish different cells

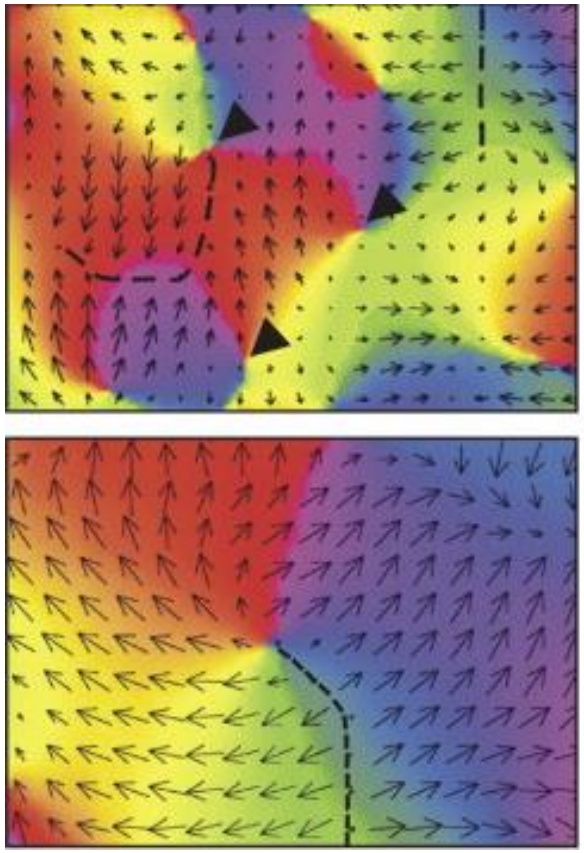
Ca- sensitive imaging: Cellular Resolution



Voltage-Sensitive Dye Imaging (VSDi)

1. **Voltage-sensitive dyes are organic molecules which reside in a cell membrane and change their optical properties in response to a change in membrane potential.**

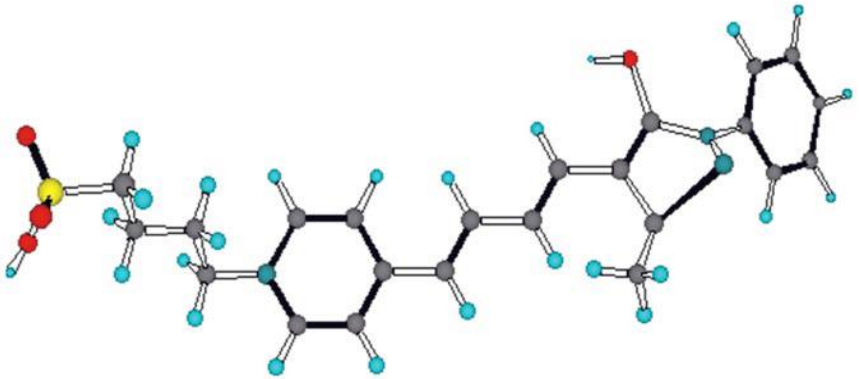
IOS: Orientation Columns in the Visual Cortex



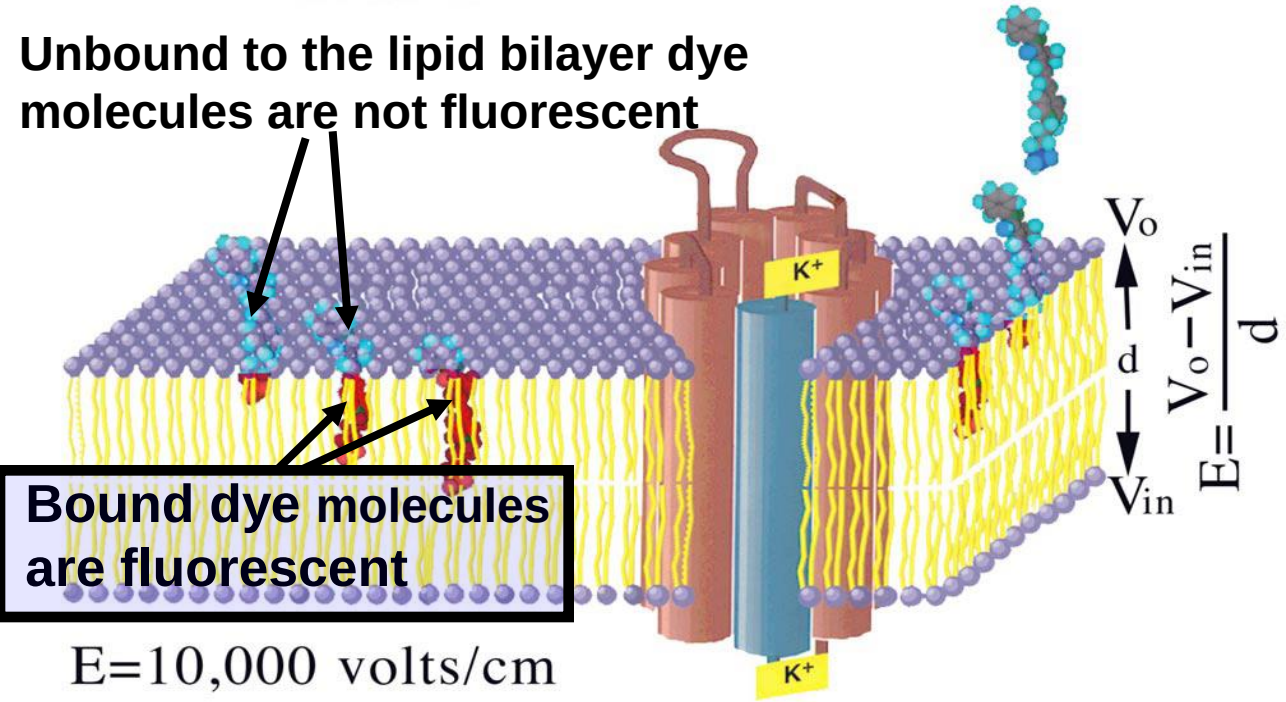
An orientation preference map, with color coding for the preferred orientation of a visual stimulus, according to the legend at the right.

Orientation and direction maps from ferret visual cortex obtained by optical imaging of intrinsic signals.

Principles of Voltage-Sensitive Dye Optical Imaging



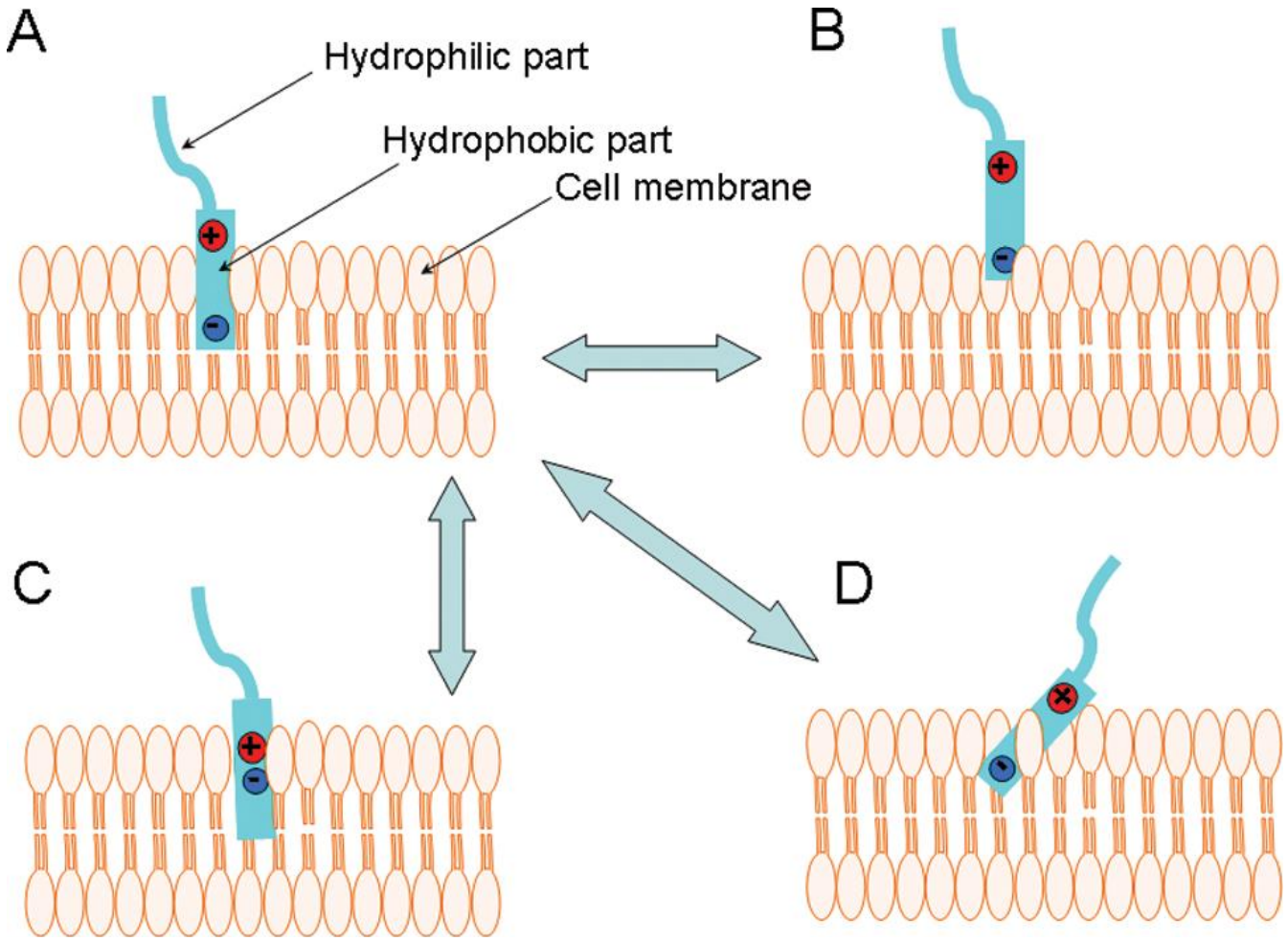
RH-795 chemical structure



Dye interactions with the lipid bi-layer

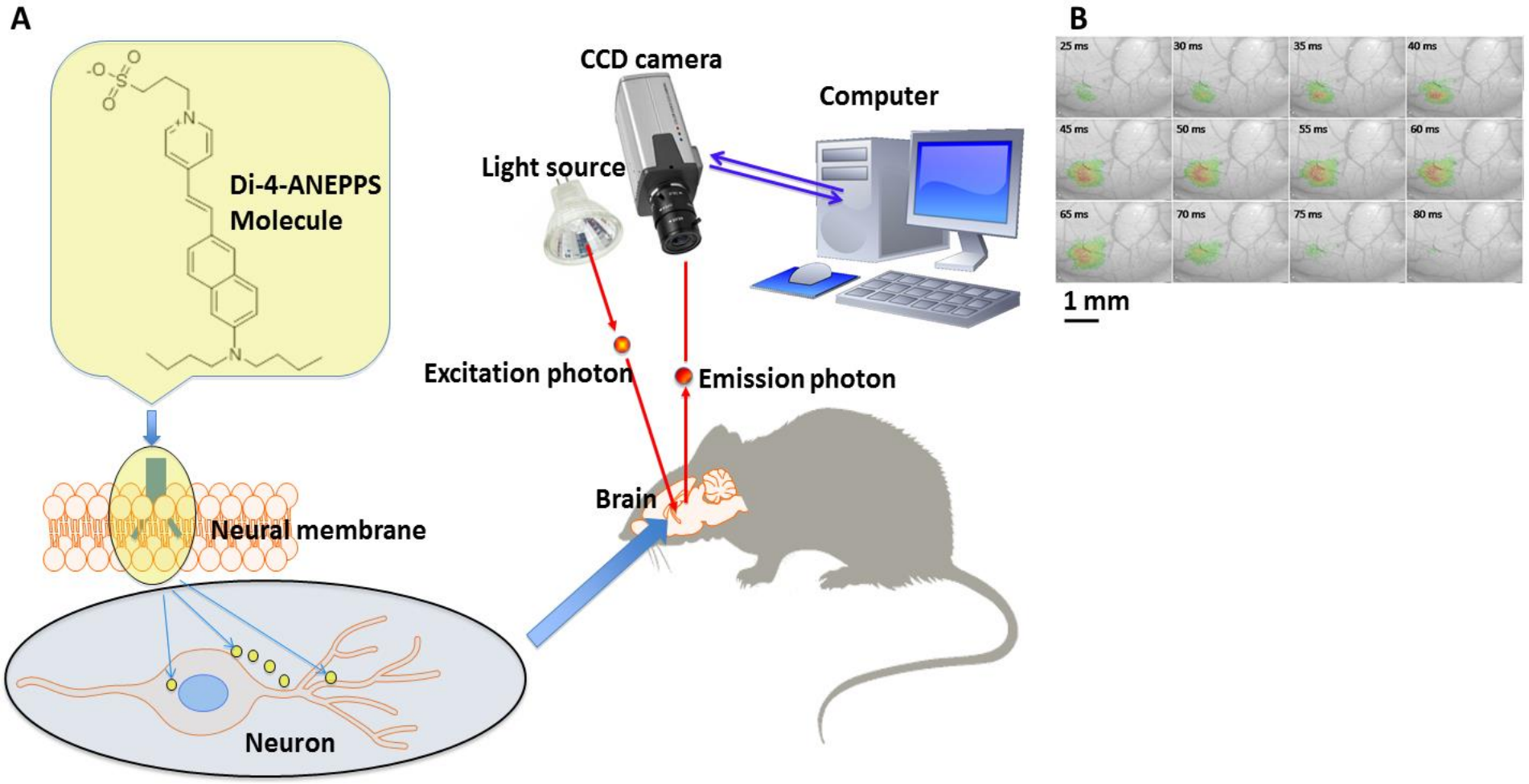
(Grinvald *et al.* 2001. In-vivo Optical imaging of cortical architecture and dynamics)

Mechanism of the VSD optical features changes

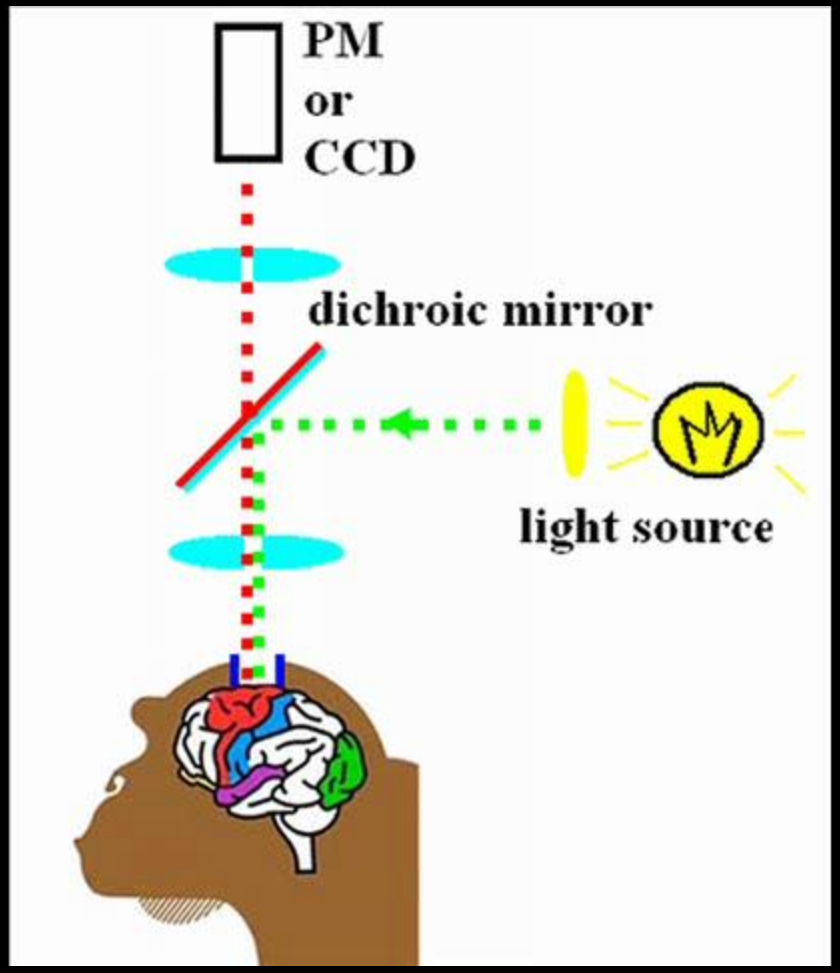


Redistribution (A), reorientation (B) and the direct electrical modulation of the electronic structure of the dye molecule (C).

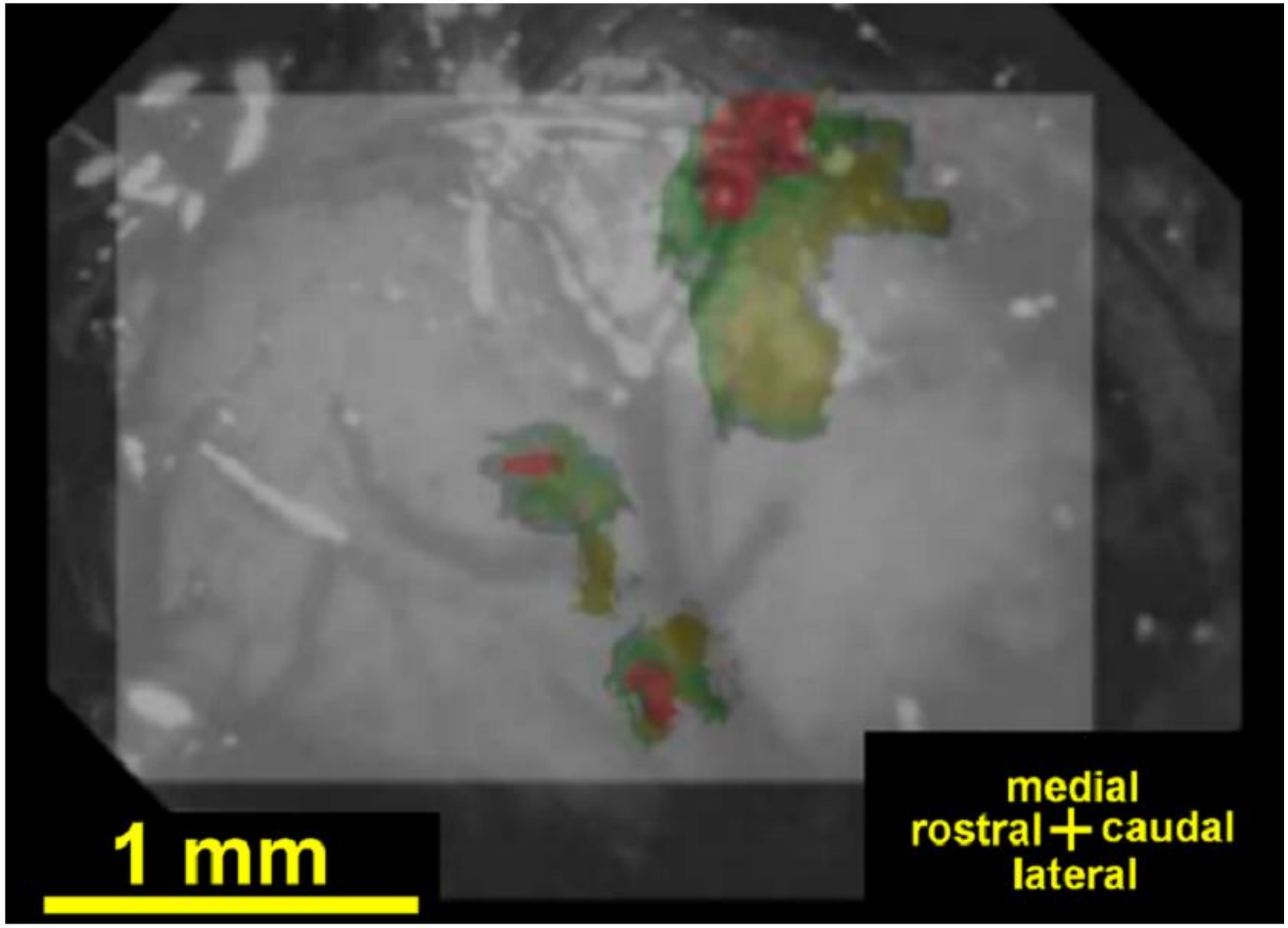
Main Parts of the VSD Imaging Setup



Excitattion and Emission Light Separation



VSD Imaging of Tonotopicity



1 kHz 5 kHz 7 kHz

Color-coded tonotopic map of sound frequencies showing the organization of subfields of the auditory cortex

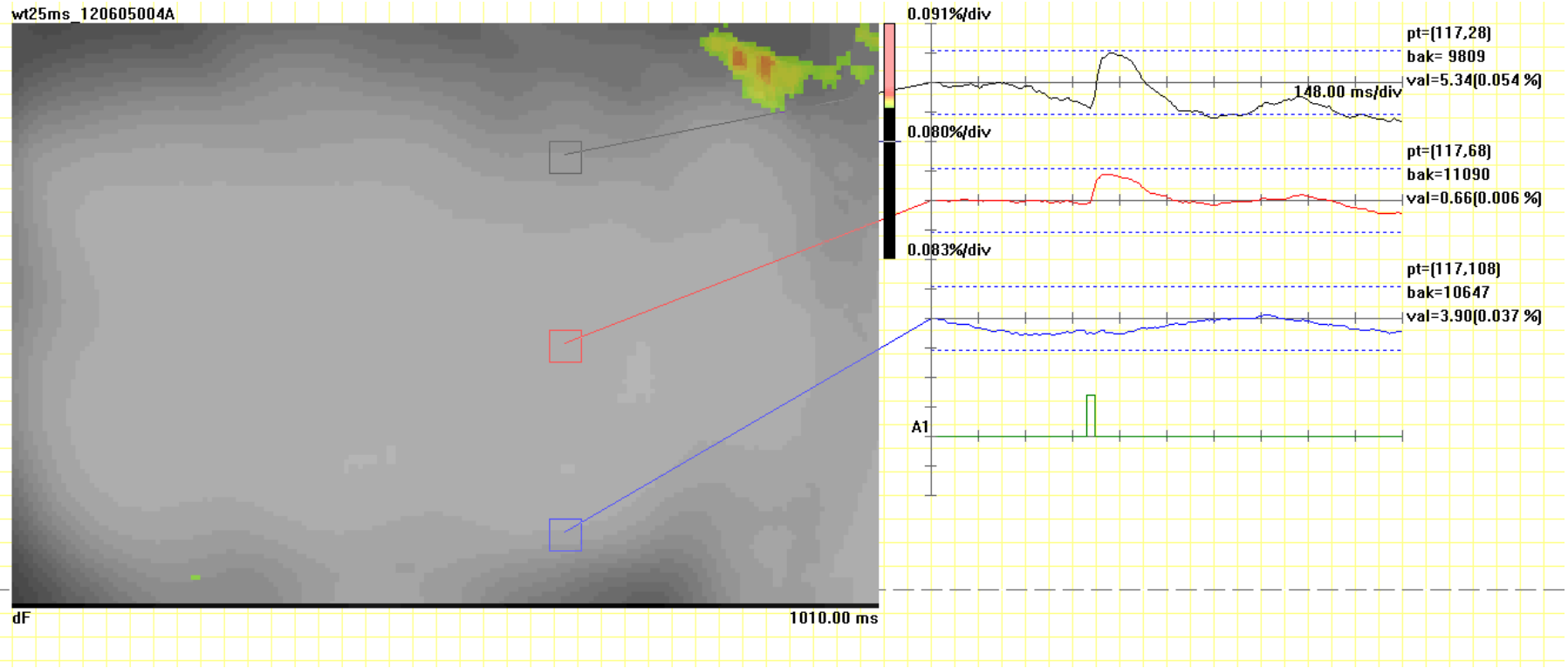
(Tsytsarev *et al*, 2009. Optical imaging of interaural time difference representation in rat auditory cortex)

Vibrissae System: Optical Imaging Research

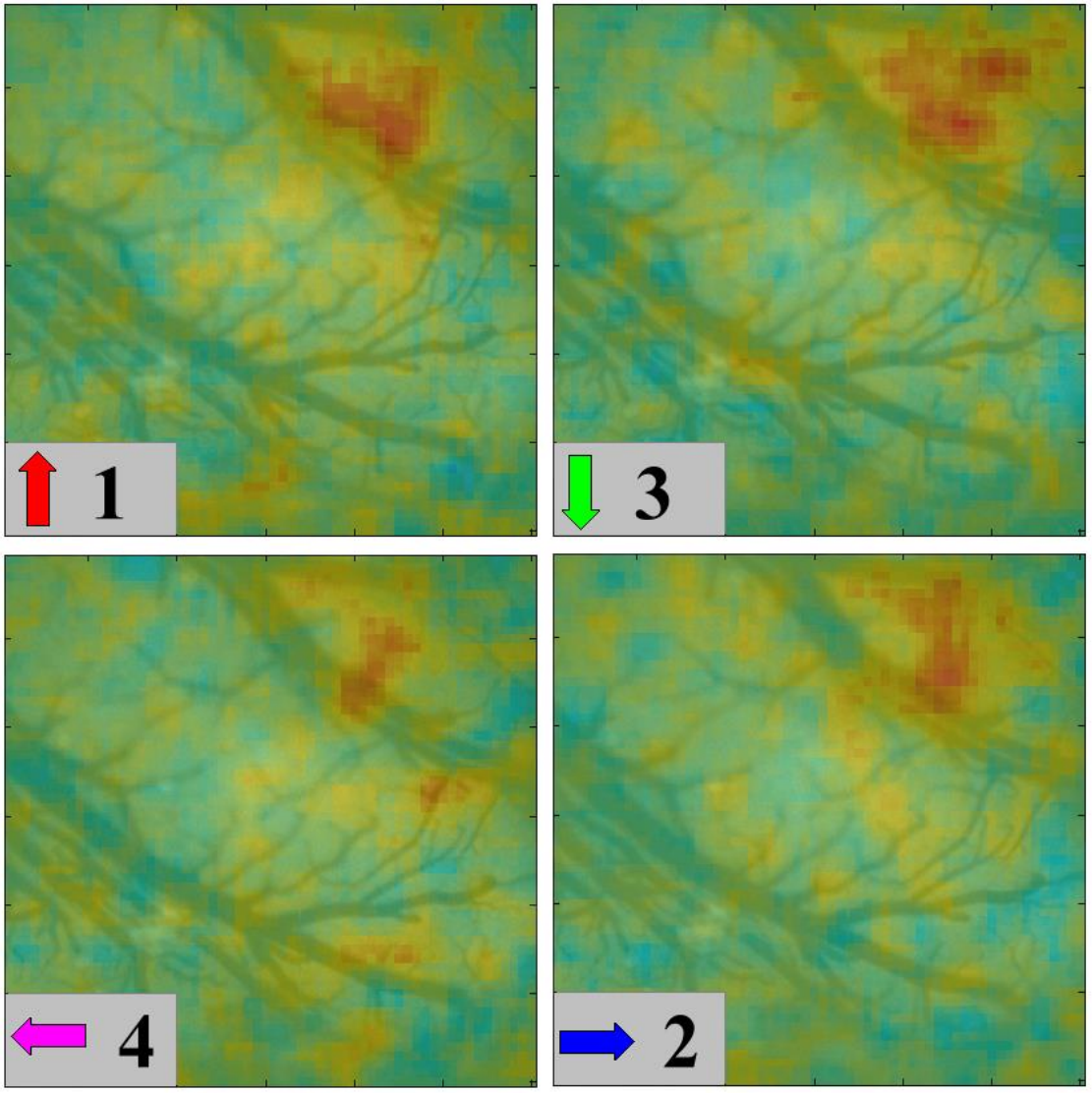
The main goal:

to create a functional map of the directional sensitivity of the barrel field using Voltage-Sensitive Dye Optical Imaging

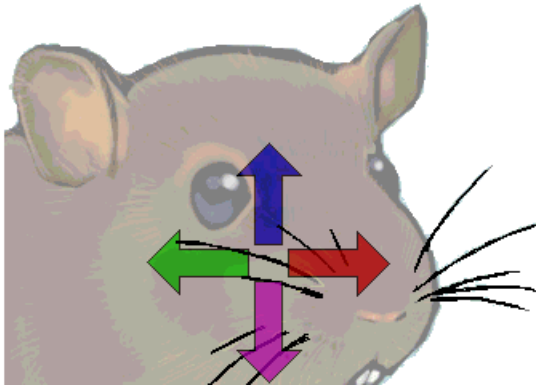
VSD signal in the Barrel Field



Vibrissae System: Angular Selectivity in the Barrel Field?



VSD optical patterns evoked by different stimuli

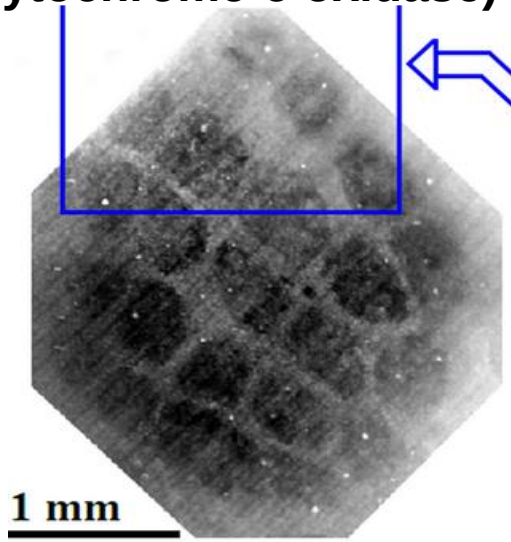


Stimuli Arrangement (color coded)

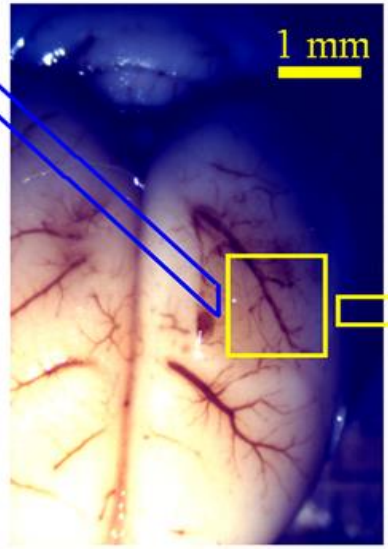
Whisker's Stimulator

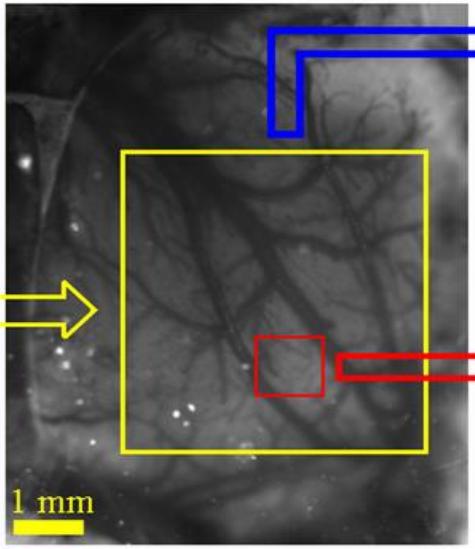
Barrel Field Voltage-Sensitive Dye Optical Imaging

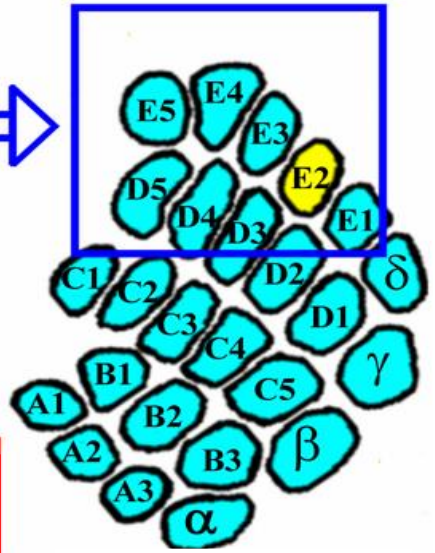
Histology
(Cytochrome-c oxidase)



Extracted brain

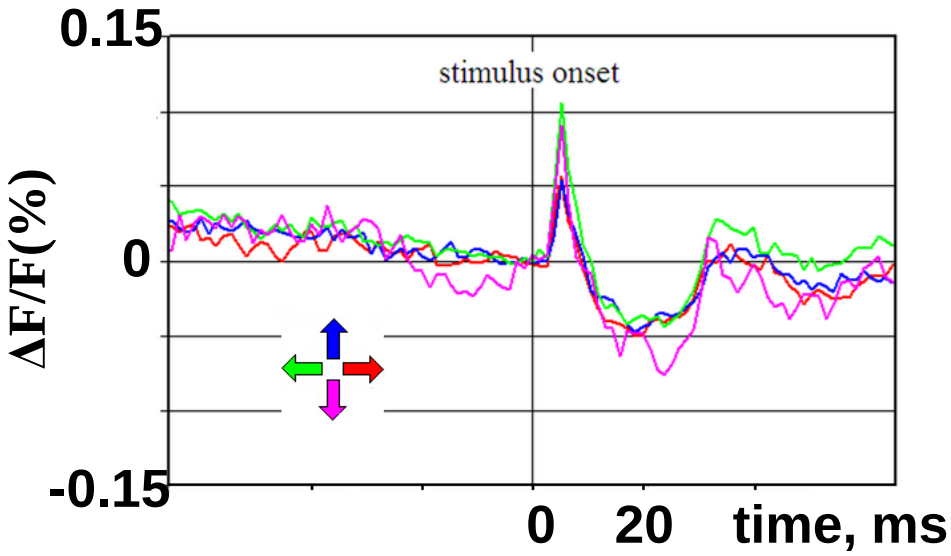






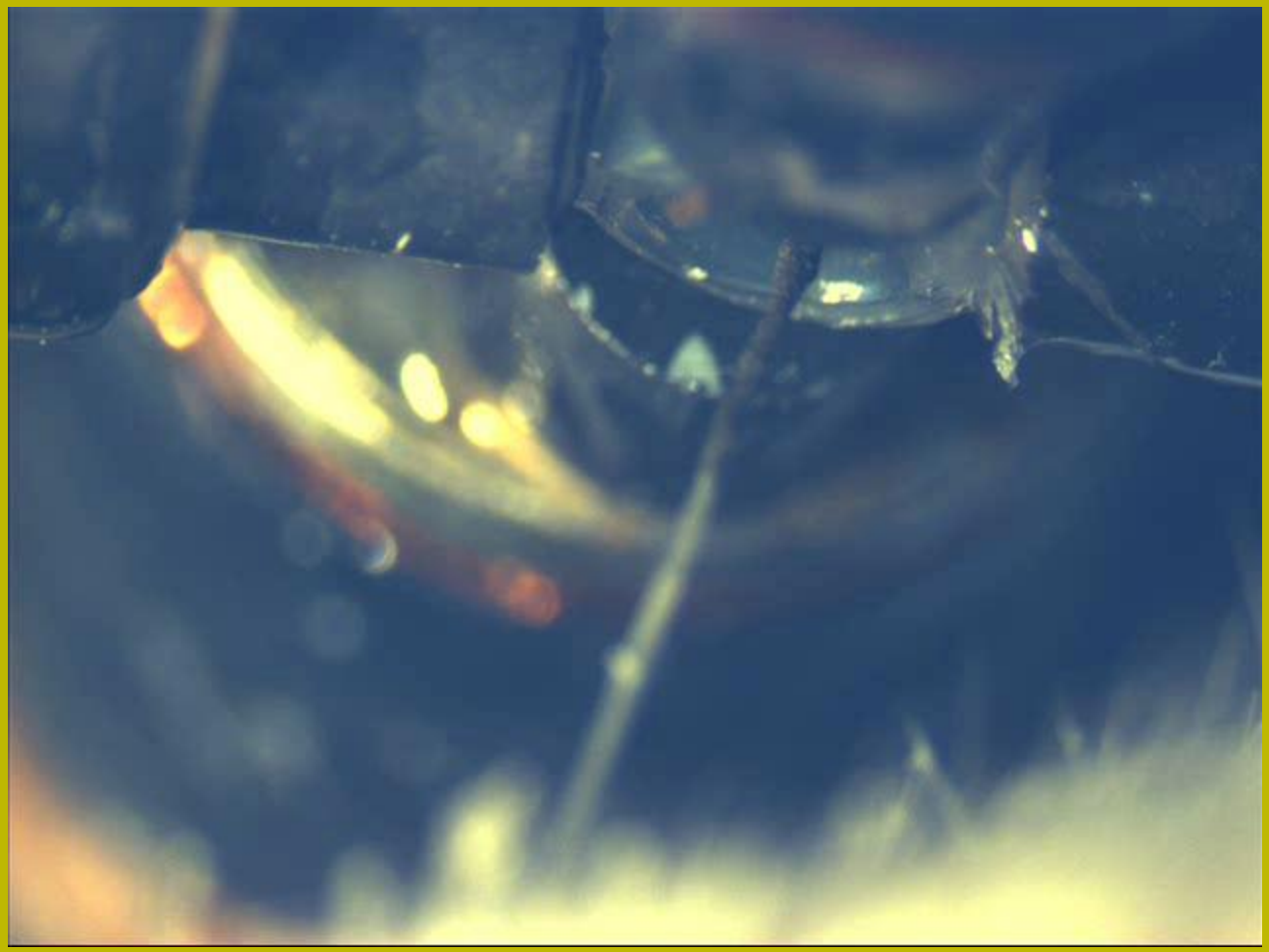
Barrel field

Brain opening and area of recording



Integrated signal in response to different stimuli

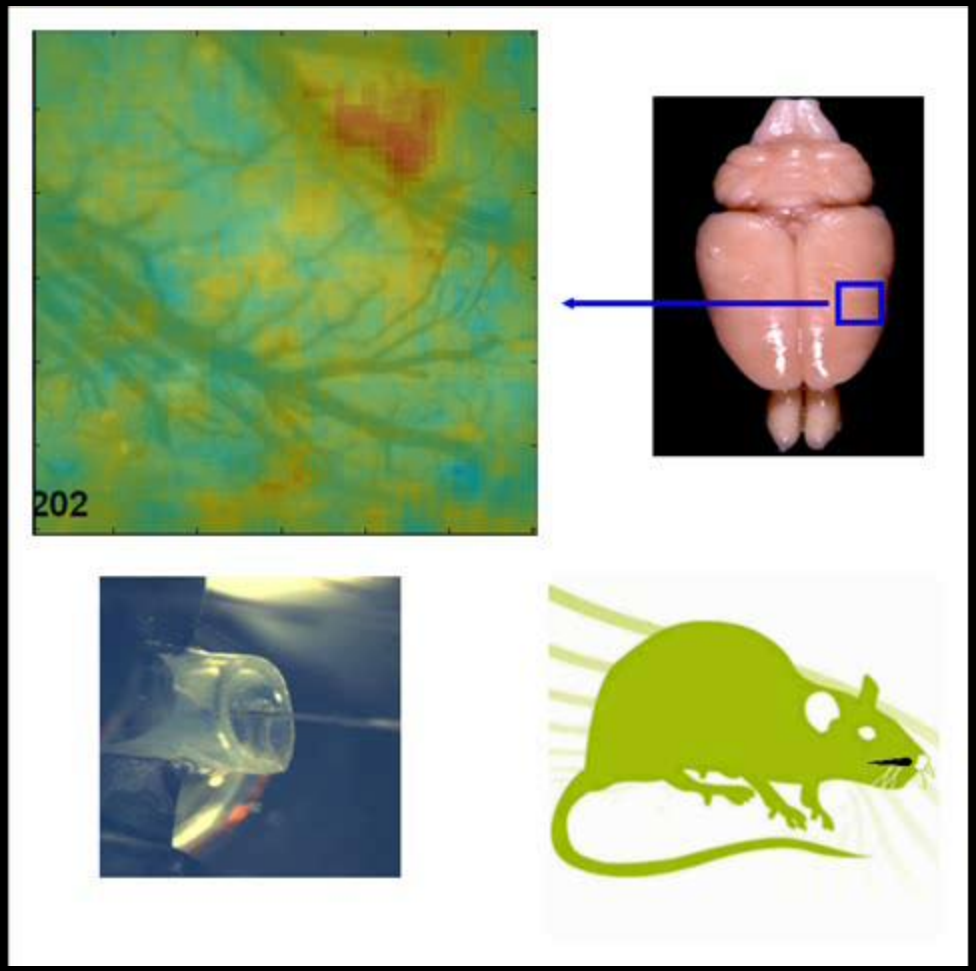
Directional Whisker Stimulation



Studying of the Cortical Representation of Whisker Directional Deflection Using Voltage-Sensitive Dye Optical Imaging

VSD Pseudocolor Pattern in the Region of Interest

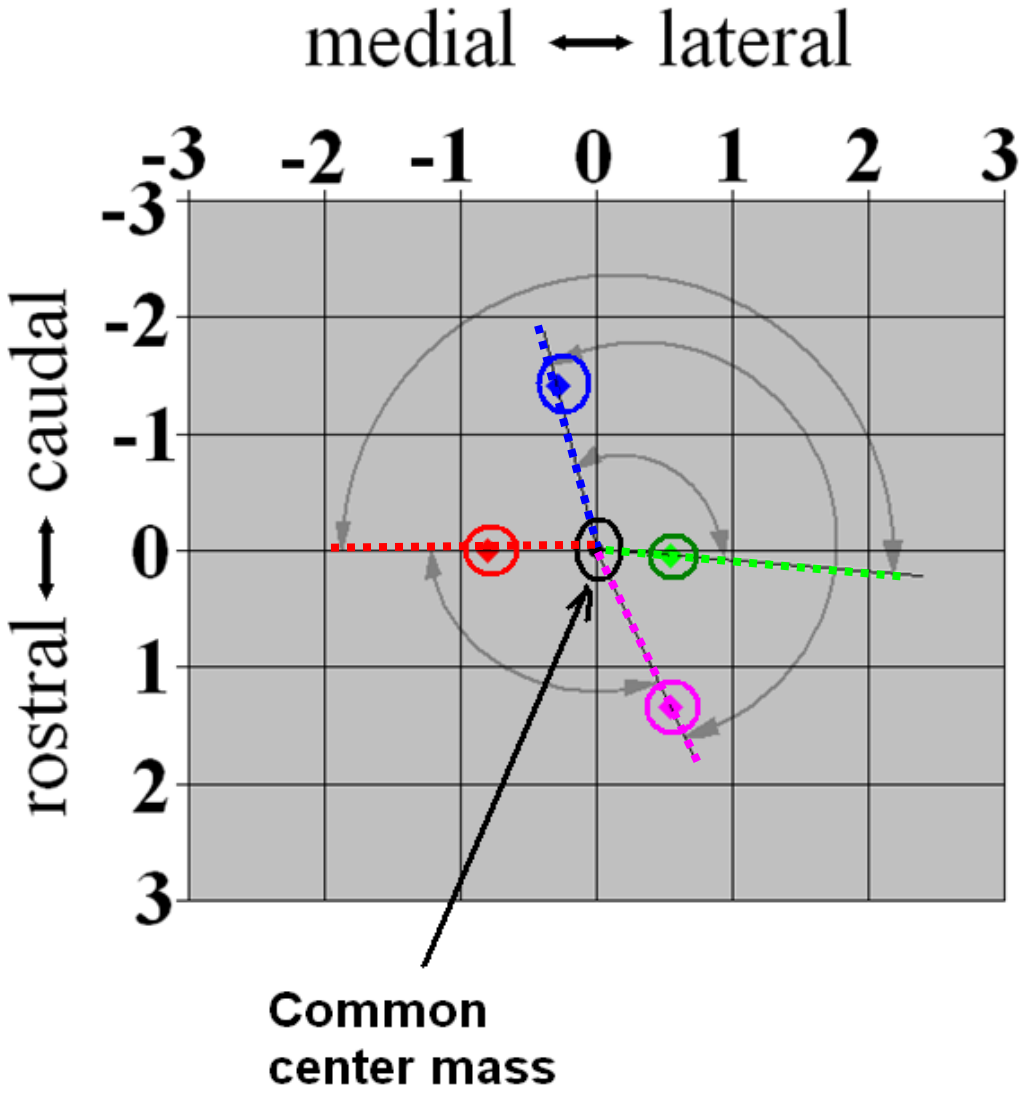
Barrel Field

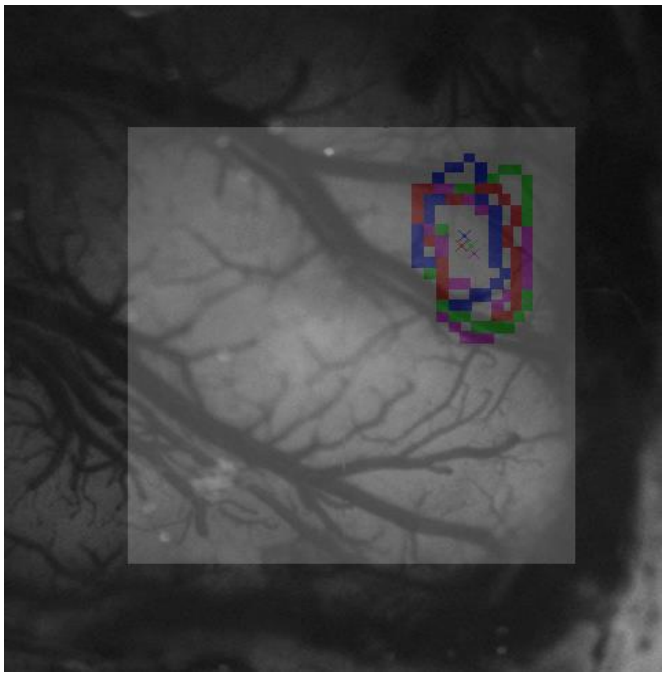


Magnetized Whisker inside the Stimulator

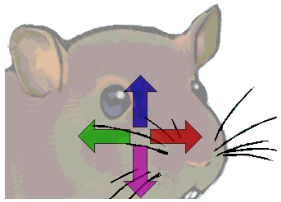
Direction of the Deflection

Activity Centers in the Single Coordinates System





Borders of the Activity Patterns after thresholding



0.5 mm

Sensory Pathway of the Vibrissa System

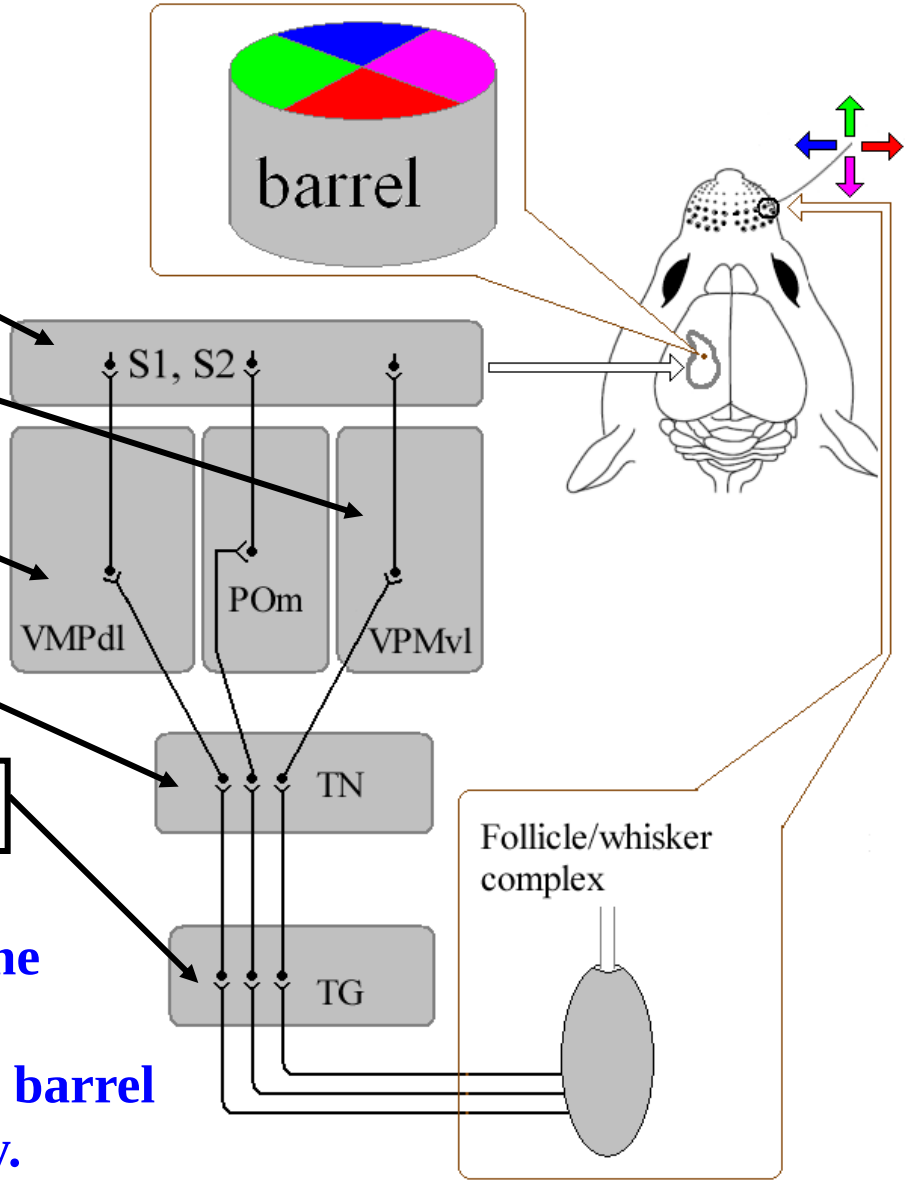
S1, S2 – primary and secondary somatosensory cortex

VPMvl – ventrolateral domain of the ventral posterior medial nucleus of the thalamus

VPMdm - ventral posterior medial nucleus

TN – trigeminal nuclei

TG - trigeminal ganglion



Conclusion:

Barrel cortex neurons are selective for the angle of whisker deflection.

These neurons are localized in the single barrel in accordance to their angular selectivity.

Thank you very much for your attention