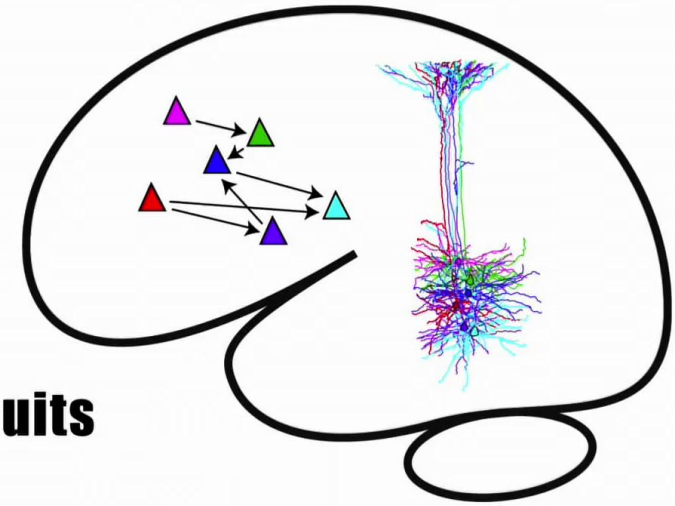
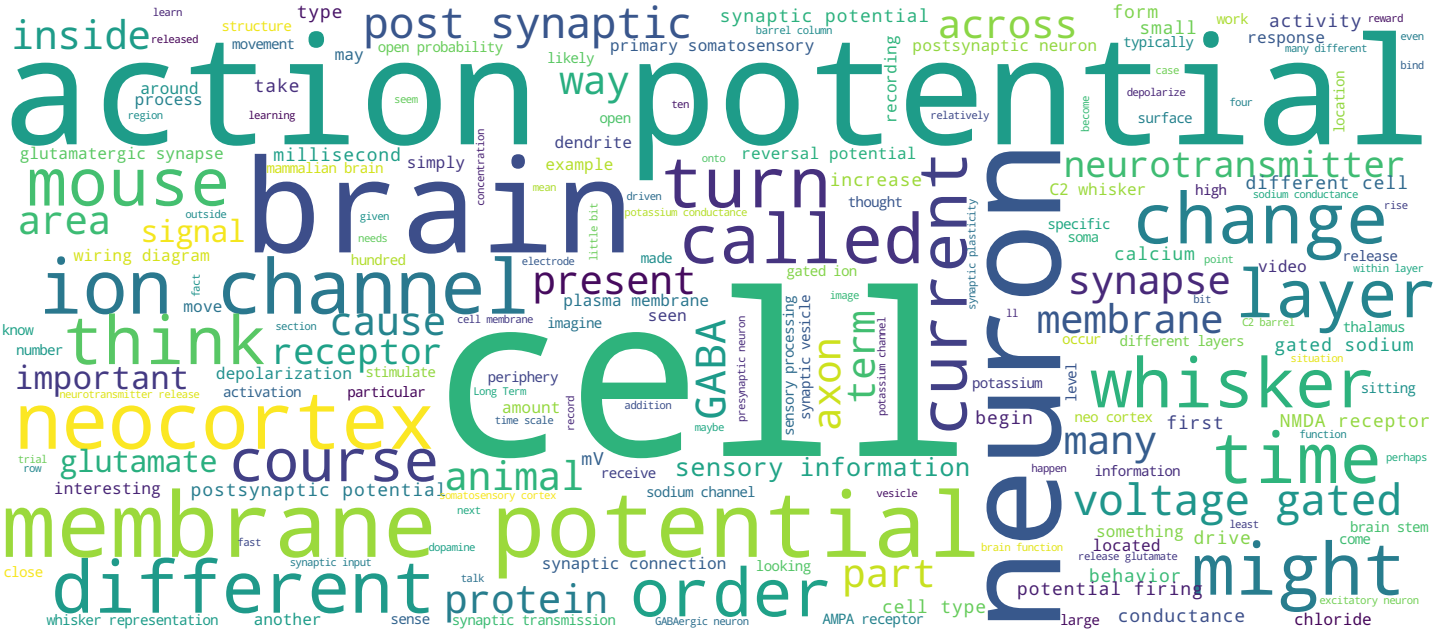


4.3 Glutamatergic circuits

Cellular Mechanisms of Brain Function

Prof. Carl Petersen



Search MOOC

Video

EPFL

Glutamatergic excitatory neuronal circuits

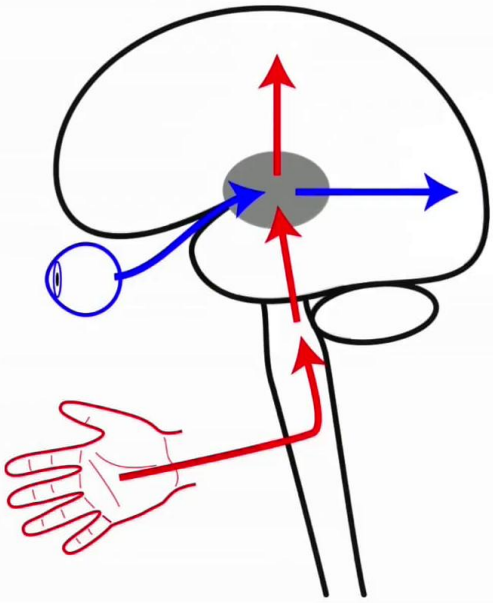
In the last lesson we saw how postsynaptic conductances evoked by glutamate release from presynaptic terminals were converted into postsynaptic potentials and integrated in complex ways across the dendritic arborization. We found that most unitary excitatory postsynaptic potentials (EPSPs) that originate from a single action potential in a single presynaptic neuron that most unitary EPSPs are small in amplitude. They have an amplitude typically of 1 mV and the last 10-20 msec in duration. An excitatory neuron is typically at rest at around -70 mV, so something like 30 mV away from action potential threshold. In order for a postsynaptic neuron to contribute to further computation in the brain, it must receive those excitatory synaptic signals depolarized beyond threshold and fire action potentials of its own at the appropriate time. This means that many synaptic inputs must be integrated in that individual neuron. In order to understand how that neuron integrates sensory information, we need to know where its presynaptic neurons are. We need to map the wiring diagram and the connectivity of how synaptic input arrives onto postsynaptic neurons.

Notes

Summary

0m 05s

Circuits for processing sensory information



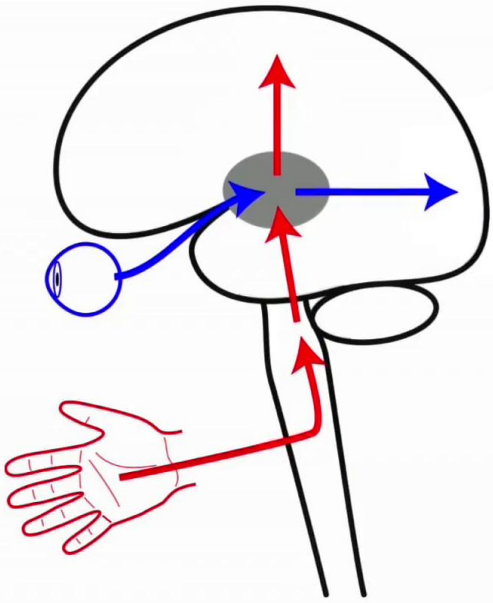
In this lesson we'll begin to think a bit about how synaptic wiring diagrams can be constructed at least in a probabilistic way and how that might relate to sensory processing in the mammalian brain. We'll begin by considering sensory processing in the mammalian brain and its dependence upon glutametergic synaptic signals. We'll think about vision and touch: two important senses for us humans. If we think about the sense of touch, there are mechano-gated ion channels sitting in our skin inside axons of sensory neurons. Touch on the skin causes action potential firing in the sensory neuron and these sensory neurons release glutamate from their synaptic terminals somewhere here in the brain stem. The brain stem neurons in turn receive those excitatory glutametergic signals and send them on to the thalamus, the somatosensory part of the thalamus deals with the sense of touch. Neurons there receive the glutametergic signals from the brain stem neurons and in turn they generate PSPs, action potentials and send the sensory signals up the neocortex where it's thought that our conscious precept of touch is driven by the activity of the neurons in the neocortex.

Notes

Summary

1m 35s

Circuits for processing sensory information



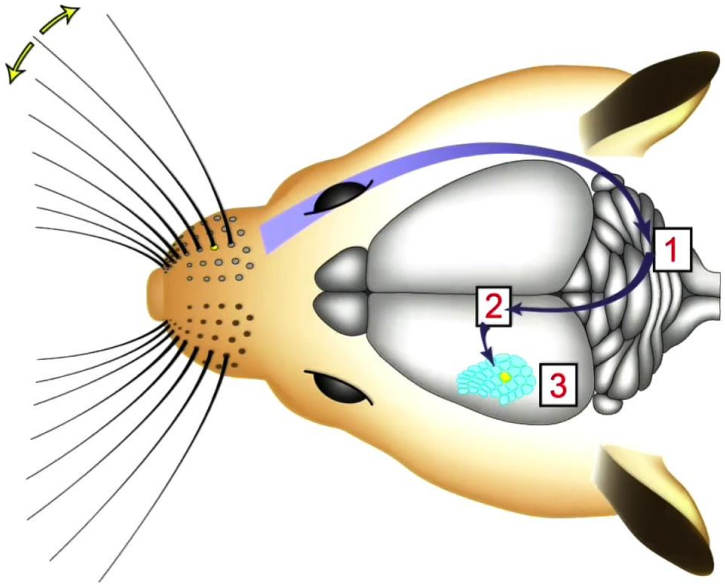
Again, most of that activity is driven by glutamatergic synapses. And so we have here a pathway of brain stem synapse and thalamic synapse and neocortical synapses, all of which are primarily glutamatergic in nature and essential in order to convey a sensory signal from the periphery to the neo cortex, where it might generate sensory percepts. Similarly, if we think about our sense of vision, this also depends heavily upon glutamatergic synapses. Within the eye, the photoreceptors that detect photons arriving from the visual field, they release glutamate on to second (inaudible) so called bipolar cells in the retina. These bipolar cells sense the glutamate and in turn release glutamate at their own synaptic terminals, on to retinal ganglion cells. It's the retinal ganglion cells that then receives its glutamatergic inputs as far as action potentials and transfers the visual information first to the thalamus where again a glutamatergic synapse conveys that information further to the visual cortex. Again, glutamate is the second molecule there. And so glutamatergic synaptic transmission is a fundamental importance of getting sensory information from the periphery to the neocortex where it drives sensory perception. And that's true for all of our senses.

Notes

Summary

3m 03s

Mouse whisker sensation



1. Brainstem
2. Thalamus
3. Neocortex

Petersen, 2007

Cellular Mechanisms of Brain Function

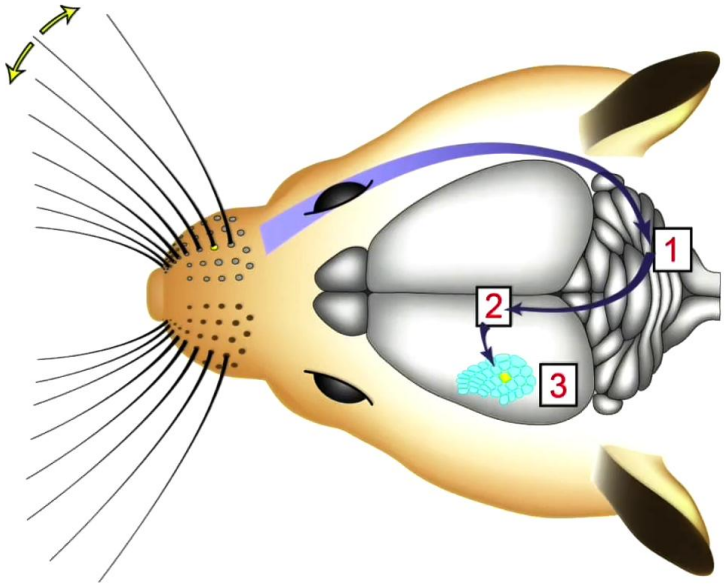
In this video I'd like to focus particularly on one sensory pathway that has some specific features that are useful for detailed analysis in terms of microcircuits and synaptic connectivity. We'll think about the *Mouse Whisker System* and the mouse is an attractive animal in terms of neurophysiology because of its extensive genetics and its extensive use in biological research. Mice are nocturnal animals, they live underground in tunnels and so vision is not one of their most developed senses. Instead, one of the ways in which they gather a lot of sensory information about their immediate surroundings comes from their array of whiskers that are present on the snout of the animal. These are arranged in a highly organized manner and using their whisker system, the animals are able to navigate in tunnels, they can see whether a hole is a sufficiently large aperture for them to enter in, they can detect the shape of objects and their sense of texture discrimination with their whiskers rivals that of the human finger tip. So this whisker system is an immensely sensitive organ through which the mouse can learn a lot about its immediate spacial environment.

Notes

Summary

4m 33s

Mouse whisker sensation



1. Brainstem
2. Thalamus
3. Neocortex

Petersen, 2007

Cellular Mechanisms of Brain Function

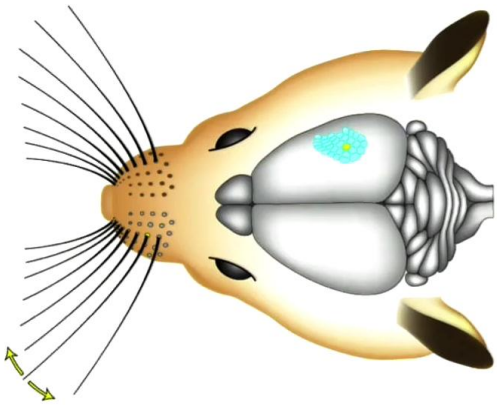
Deflection of one of these whiskers activates mechano gated ion channels sitting here in the whisker follicle and that drives actual potential firing here down the trigeminal nerve. There's a first glutamatergic synapse in the brain stem and neurons in the brainstem receive that glutamatergic signal postsynaptic potentials are generated, some of the neurons in the brainstem fire action potentials and those signals in turn, are relayed to the thalamus. Again, thalamic neurons here receive the glutamatergic input post synaptic potentials are generated and integrated in the thalamic neurons, action potentials are fired and subsets of those thalamic neurons and information is then further relayed to the primary somatosensory cortex where presumably, whisker sensory perception takes place for the mouse. So there are three glutamatergic synapses that relay sensory whisker information from the periphery through to the neo cortex of the mouse, and in particular it's the primary somatosensory neo cortex of the mouse that deals with whisker sensation.

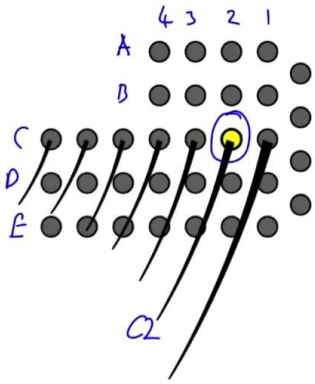
Notes

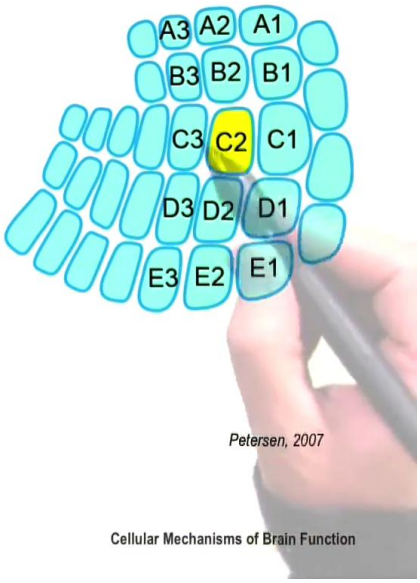
Summary

5m 57s

Whisker map in somatosensory neocortex







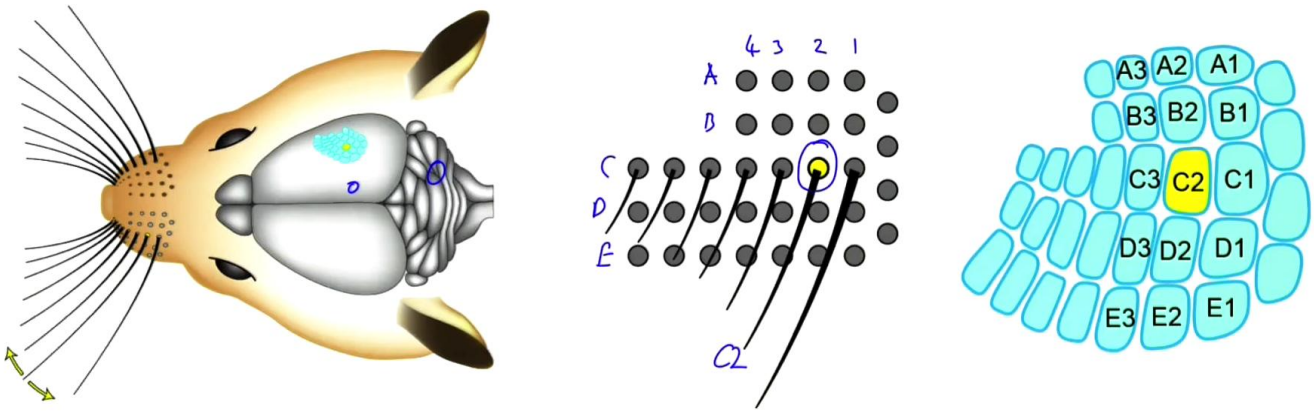
The organization of the primary somatosensory neo cortex of the mouse is remarkable in the sense that it has obvious anatomical units that appear to be in a direct one to one correspondence with the layout of the whiskers on the periphery. So the whiskers here, the hairs that eminate from individual whisker follicles are laid out in a highly stereotypical patent where every mouse has a same organization of the whisker lay out and indeed mice and rats have also the identical layout. So because it's highly stereotypical we can then label these whiskers in specific ways and so this is called the *A* row, the *B* row, the *C* row, *D* row, *E* row and equally they're considered to be archs *1*,*2*,*3* and *4* in terms of the whiskers and so this one here that's highlighted in yellow is a so called *C2* whisker. If we now look at the new cortex across the horizontal extent of the neo cortex when looking down on the face of the neo cortex across a so called somatotopic map, we'll see that there are corresponding units here drawn in blue that correspond to the whiskers. Again, highlighted here in yellow is one particular anatomical unit.

Notes

Summary

7m 13s

Whisker map in somatosensory neocortex



Petersen, 2007

Cellular Mechanisms of Brain Function

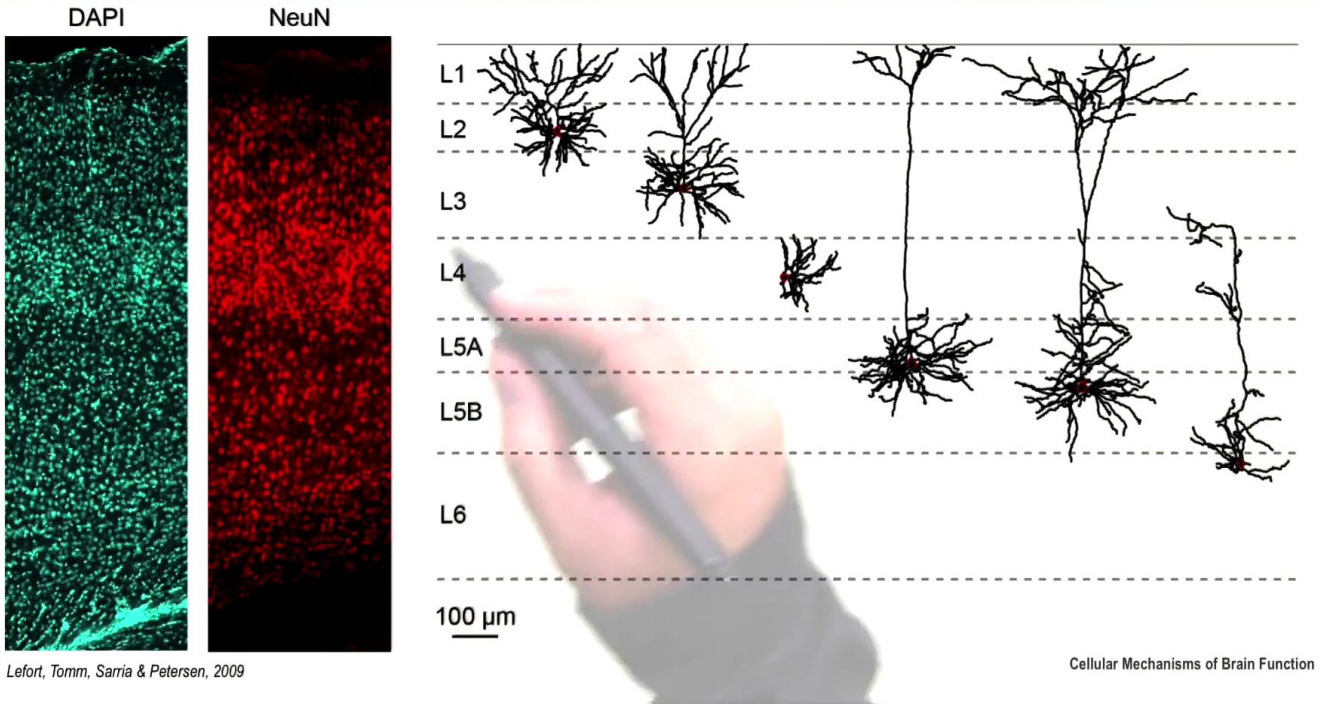
The so called *C2* whisker representation and these round things here are called barrels. So this is called the *C2 barrel*. Present in primary somatosensory cortex and because of its obvious analog in terms of the whiskers on the periphery one might think that sensory information relating to the *C2* whisker is primarily processed in the *C2* barrel column in the primary somatosensory cortex, and indeed, at the various stations in the brain stem and the thalamus there's a nice representation also of the individual whiskers in terms anatomically identifiable units.

Notes

Summary

8m 38s

Excitatory neurons of the C2 barrel column



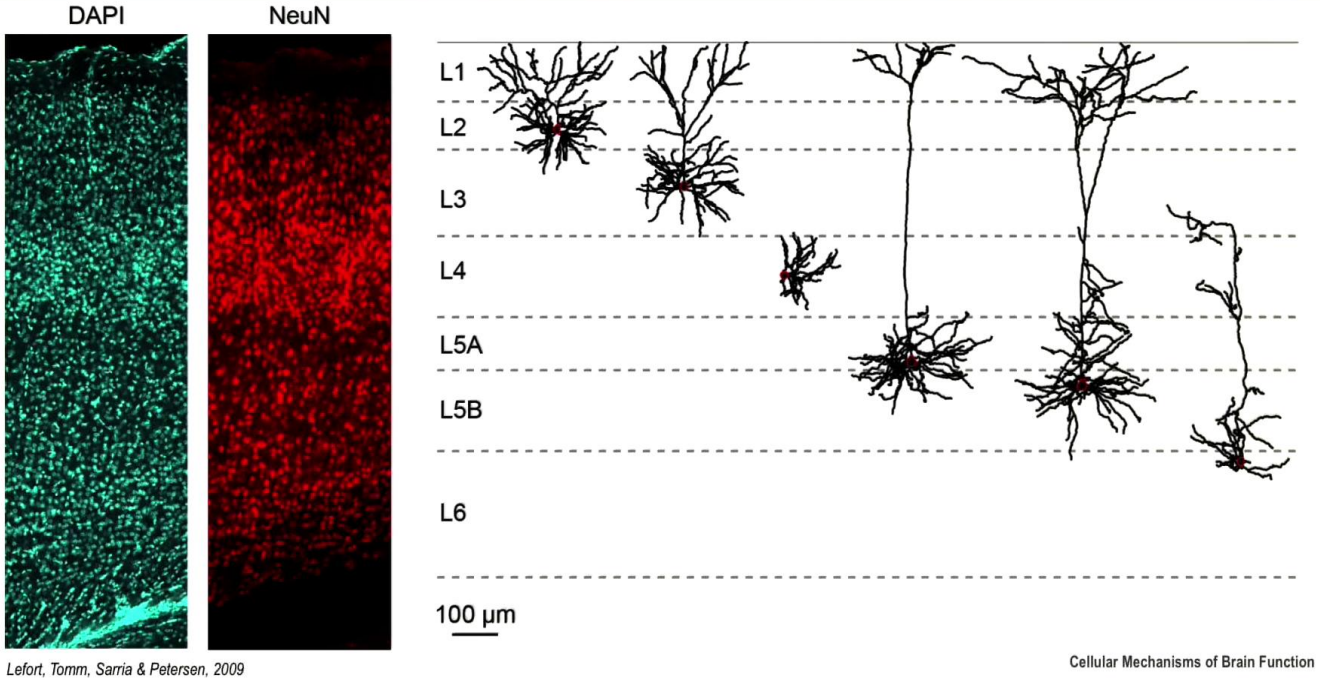
Now if we look at a section of the brain where we cut through the thickness of the brain and so before we were looking at the top of the brain and we saw that there was a nice C2 representation of the whisker. Now we look inside the brain and we see that it has some depth. It's about 1mm thick, the neocortex in this particular location in the mouse. In here maybe you can make out the walls of the C2 whisker representation that's here, high cell density represented here by this fluorescent stain of DAPI that stains the cell nuclei and you can also see it here in this stain of neurons present in this air, you'll see that there are these walls here that represent these limits here of the C2 whisker representation and now this would be the surface of the brain up here, and here we go deeper into the brain and this is the thickness of the neocortex. Its about 1.2mm thick. Now there are some obvious differences at different depths in the neocortex and people have divided the neocortex into different layers, and so there's a layer one, the most superficial, the outside area of the brain that has very few cells in it. It's mainly composed of synapses, axons, dendrites and synapses.

Notes

Summary

9m 24s

Excitatory neurons of the C2 barrel column



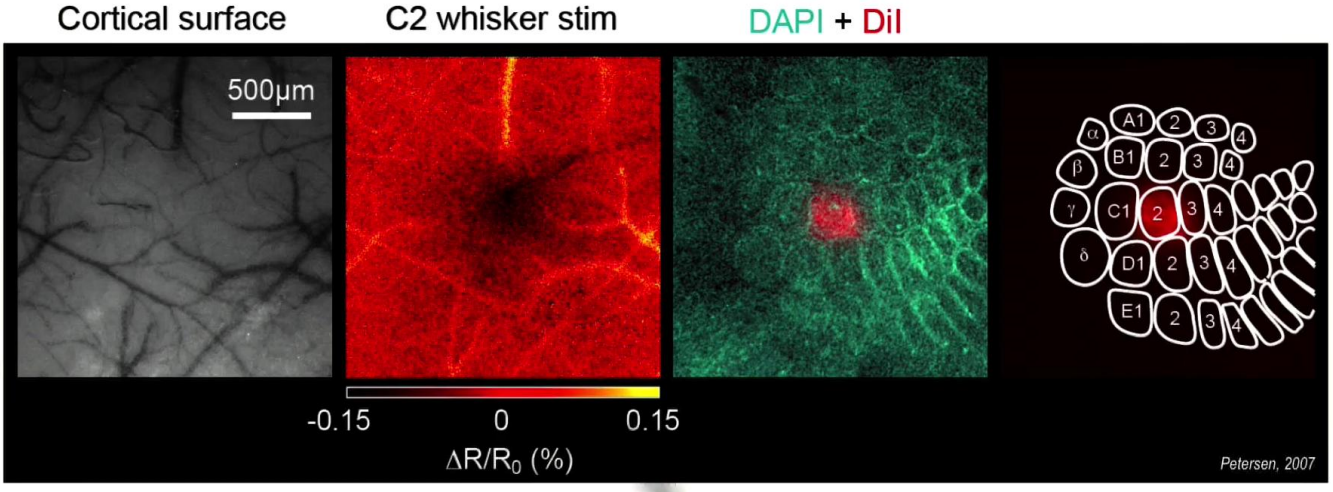
The cells themselves begin here in layers 2 down to layer 6, and you'll see that there are different densities of cells in different layers. And here we've labeled some individual neurons that are present of these different layers and you can see that there are some different morphologies of the cells. And so there are very small cells that are present here in this so called *Layer 4* area that's actually where the main sensory input arrives in the neocortex and in the superficial layer, there are so called *small pyramidal* cells small excitatory neurons, and in the deeper layers, there are much larger cells with much more extensive dendrites. And it turns out that the microbiology, the electrophysiology and the synaptic properties of the cells in different layers are different, and so we can think of these as different cell types. All of these neurons here are glutamatergic neurons that have their axonal outputs release glutamate. So they are of the one cell type in terms of their neuro transmitter but we can subdivide them into many different subtypes of excitatory neurons and one of their ways of subdividing them is their laminate position within the neocortex that tells a lot about the synaptic connectivity. Which is what we're going to look at further.

Notes

Summary

10m 49s

Functional mapping of barrel cortex



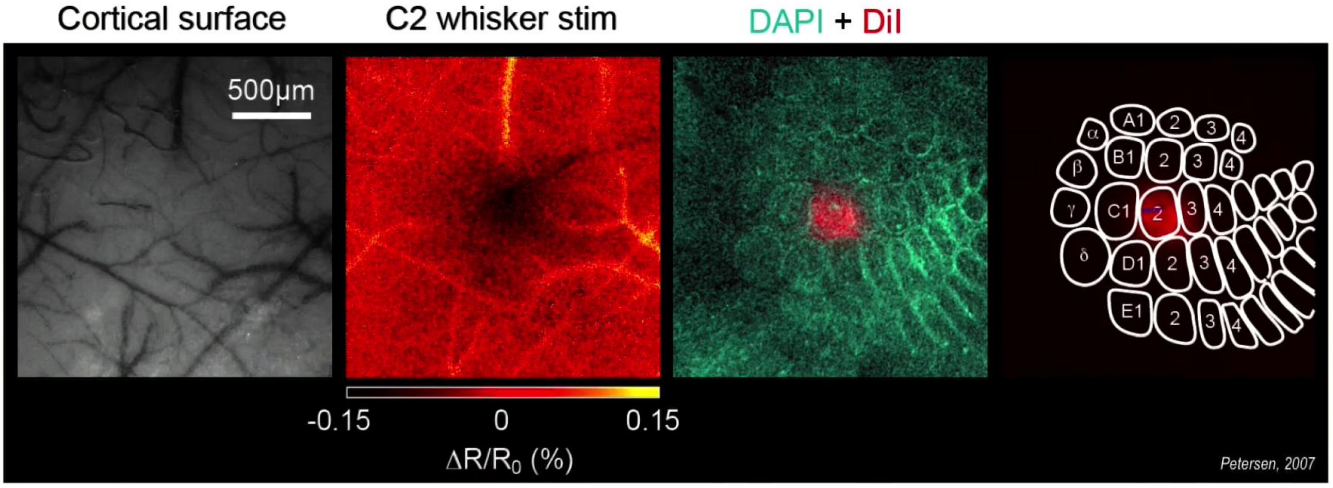
Now in order to functionally map the wiring diagram of the C2 barrel column, we first need to identify it in the living brain. It turns out that the anatomical barrel map that we've looked at so far is nice in anatomical sections but when you look at the living brain, you simply see the surface blood vessels without an obvious indication of where different whisker representations are. However we can perform a simple optical imaging technique where we can shine a red light on the surface of the brain, we can then stimulate the C2 whisker that then causes activity in a localized region of the brain, the C2 whisker representation, and that changes in turn, the optical properties of this area of the brain. There are changes in blood flow, damaged from neuron activity, and there are also swelling of different processes that affects how light enters and leaves this area of the brain. So, simply by shining a red light on the surface of the brain, stimulating the whisker, we can see changes in deflection of this red light and it turns out that when an area of the brain is activated, less light is reflected, and at the level of hundreds of microns, we can now map the location of the C2 whisker, it causes a response in the small area here of the brain.

Notes

Summary

12m 11s

Functional mapping of barrel cortex



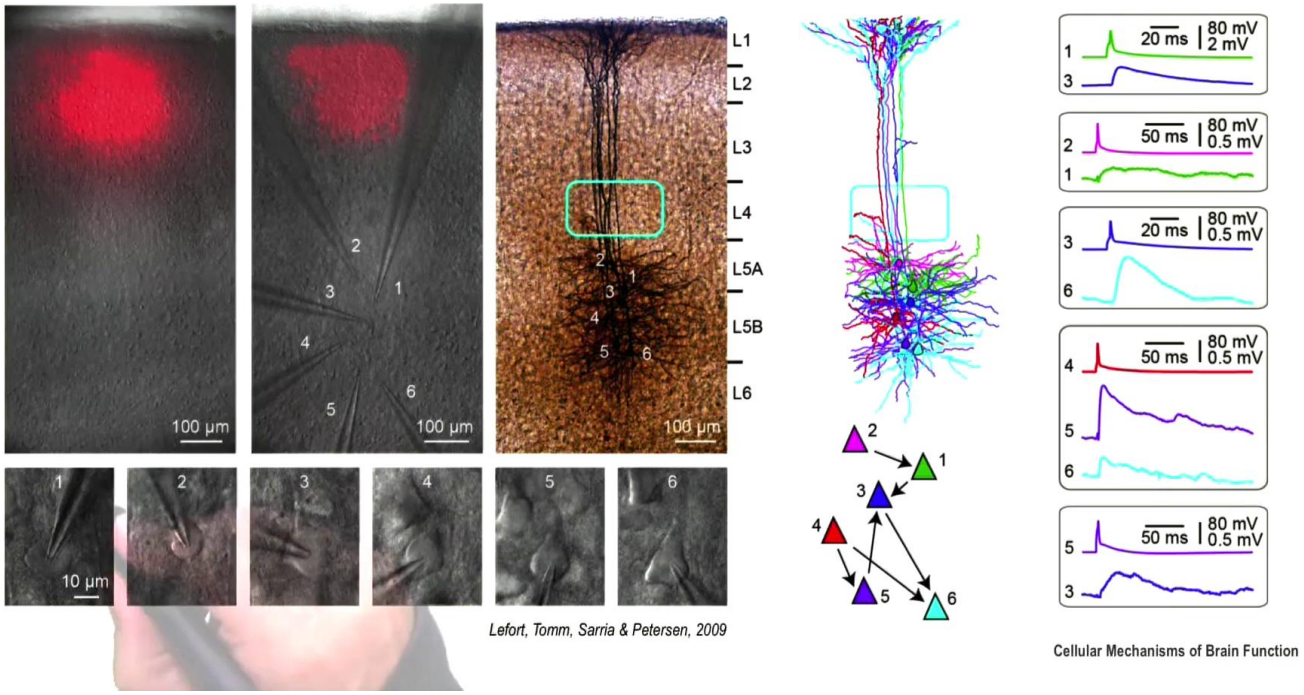
We can then furthermore put a bit of red dye at this location inside the brain and at later times we can cut horizontal sections of the brain and see that the red dye that we placed inside the brain is located inside the map of the whiskers and in particular, it corresponds to the C2 barrel column inside the neocortex. As one would expect, there's a nice mapping of the functional location of where C2 whisker evokes responses and the anatomical map that matches them nicely. You can also see that the spacial extent of this whisker representation is really at this level of hundreds of microns. The C2 whisker representation is maybe some 200 by 300 micrometers in extent, and as mentioned, the neocortex is about 1.2 mm thick at this location. So we can then do this type of experiment, inject some red dye, cut brain slices through the thickness of the cortex, and we can then begin to analyze how are neurons that are located in different layers communicate with each other.

Notes

Summary

13m 42s

Synaptic microcircuits in the C2 barrel column



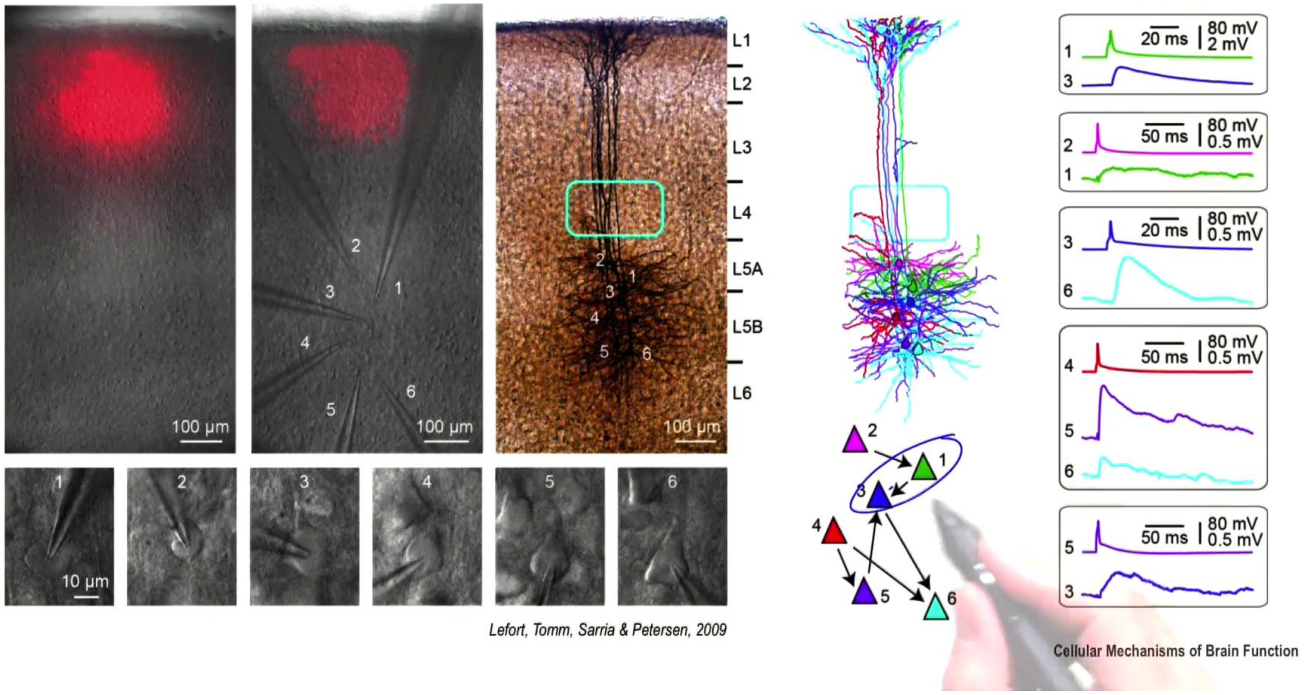
So here's a picture of a living brain slice, we injected red dye to label the C2 barrel column, you might be able to make out the so called barrel walls here, present inside the layer 4 of the neocortex, and in order to analyze synaptic connectivity, we can then simultaneously record from a number of different nerve cells. In this particular experiment, we're recording from six neurons that are located here in layer 5 of the neocortex, so some depth from the surface of the brain would go to the infragranular layers and here we've got six wholesale recordings that are located on six different neurons, so in each case there's a wholesale glass electrode that's making contact with this neuron as a membrane potential recording and so there's a continuity between the patch electrode and the cell membrane here, so we can record the membrane potential. In order to study synaptic connectivity, we can then initiate action potential in one of our cells. We can simply inject current into *Cell 1* cause it to depolarize, fire an action potential, and then see what happens in these other neurons, whether any of them respond with post synaptic potentials.

Notes

Summary

14m 56s

Synaptic microcircuits in the C2 barrel column



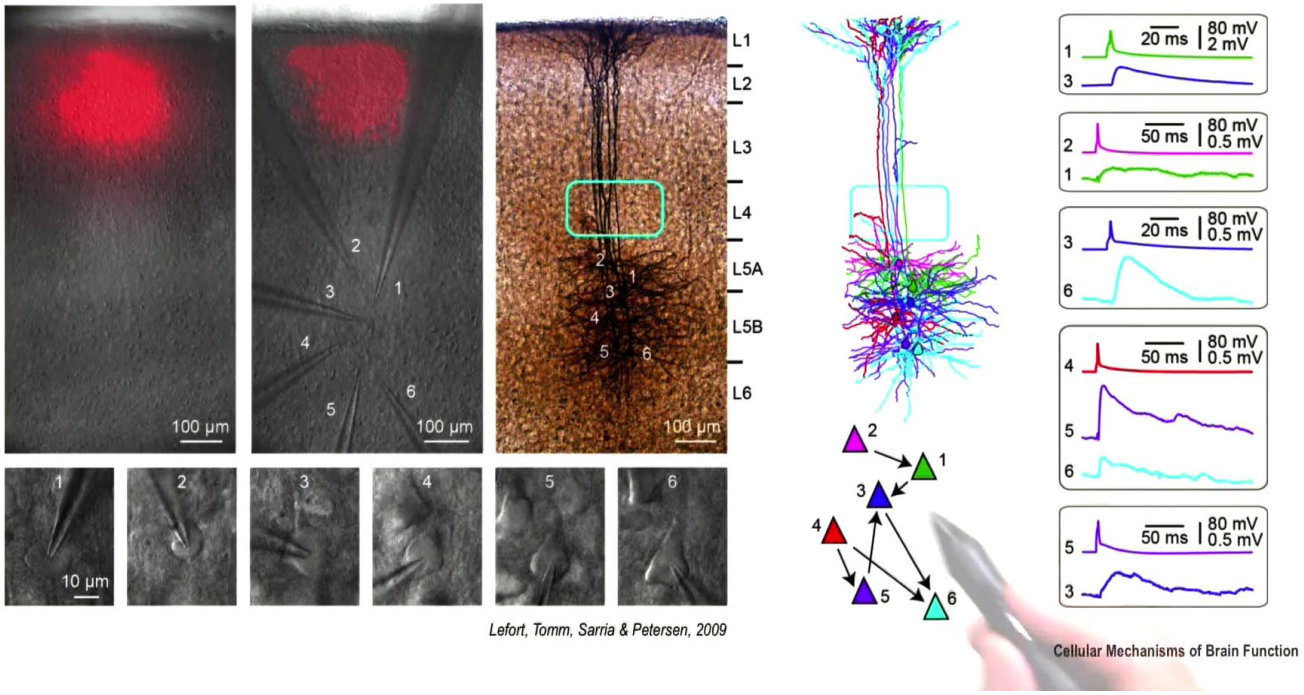
In this particular experiment, *Cell 1* released glutamate and caused post synaptic potentials in *Cell 3*. That's what you see here, an action potential in *Cell 1*, and a post synaptic potential in *Cell 3*. This 80mv refers to the action potential trace and the 2mv refers to the post synaptic potential so the post synaptic potential, as expected is somewhere in this 1mv range lasting some tens of milliseconds, whereas the action potential is brief, it is one millisecond depolarization of about 50 to 80mv. So, *Cell 1* is synaptically connected to *Cell 3*, there's a synaptic connection. But what's not shown here is that, that same action potential didn't cause any effect of the membrane potential in cells 2, 4, 5 or 6 so there's a single connection here from *Cell 1* to *Cell 3* and four of the others have no connection and so *Cell 1* has a 20% probability of having post synaptic partners, only one out of five of these possible cells were synaptically connected to *Cell 1*. So we can think of that as a 20% probability of finding a synaptic connection.

Notes

Summary

16m 17s

Synaptic microcircuits in the C2 barrel column



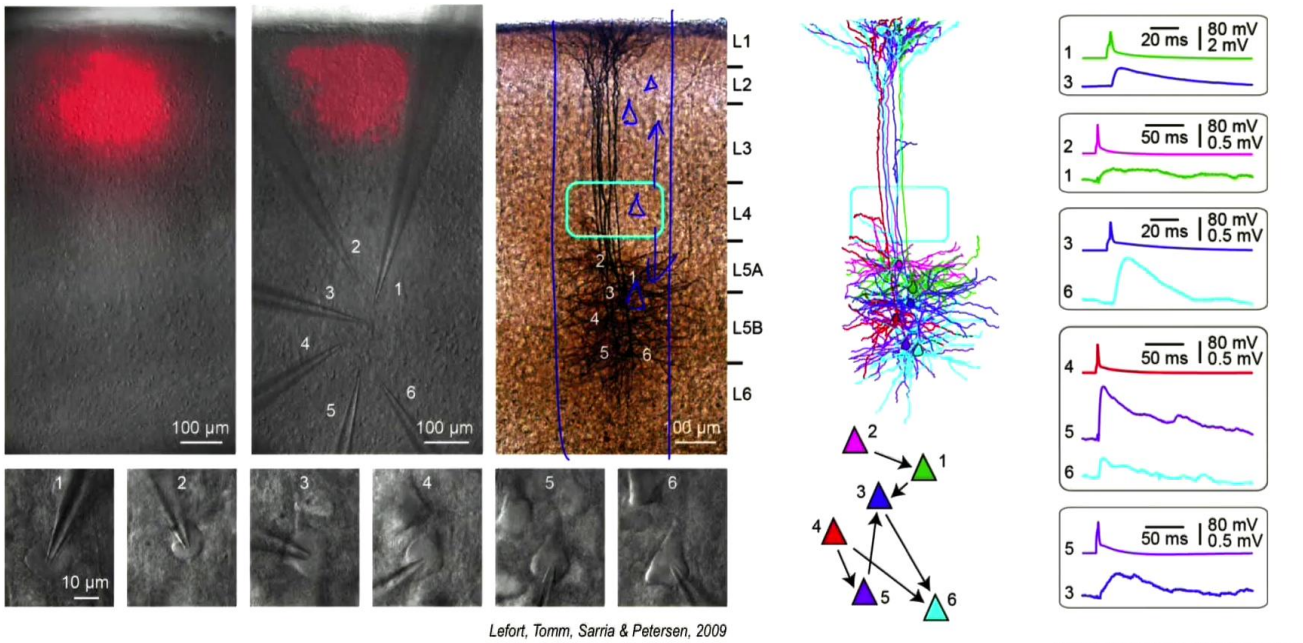
We can then of course, go through the different cells and stimulate *Cell 2* and see whether any of these other cells responded and in fact it turns out that *Cell 2* drives a small postsynaptic potential in *Cell 1*. Note though, the scale is rather different in the time course of the PSP is also different from *Cell 3*. There's some diversity in the properties of the postsynaptic potentials as we've already seen. We then go through all the different cells stimulating cell 1, 2, 3, 4 and 5 in turn, and seeing who responds postsynaptically to that action potential. That then allows us to draw a small wiring diagram for these particular six neurons that we recorded from, here you see the dendritic structure of those cells, here we've separated them, adding different colors so you can see the structure of the different neurons, they're all excitatory, pyramidal neurons and they're connected in this particular wiring diagram pattern where *Cell 2* connects to 1, one to three three connects to six, but it receives an input from *Cell 5*, four connects to both five and six and so there's a divergent output here from *Cell 1*, causing postsynaptic potentials in both of these two partner cells that are postsynaptic territory.

Notes

Summary

17m 33s

Synaptic microcircuits in the C2 barrel column



Lefort, Tomm, Sarria & Petersen, 2009

Cellular Mechanisms of Brain Function

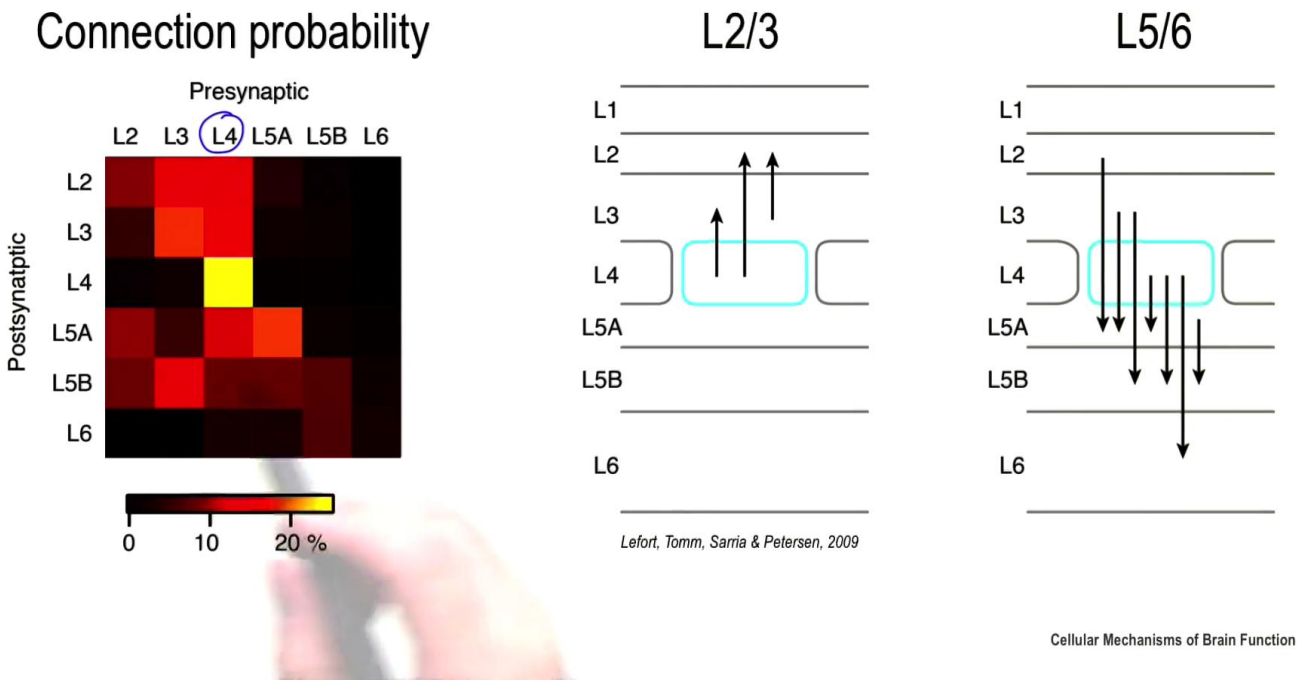
So we can then repeat this experiment over and over again in different layers recording cells in *Cell 2*, in *Cell 5*, we have four and simply see what the probability of finding synaptic connections is across a population of neurons. We can't record from all the cells at the same time so it becomes a statistical argument in the end where through analyzing many different brains, focusing on the one particular area of the brain and nicely delimited column of the neocortex, we can get the statistical wiring diagram of how neurons in different layers, different cell types if you like, communicate with each other at a statistical level and that then, helps us understand how sensory information is processed in this part of the brain.

Notes

Summary

18m 48s

Excitatory microcircuit of the C2 barrel column



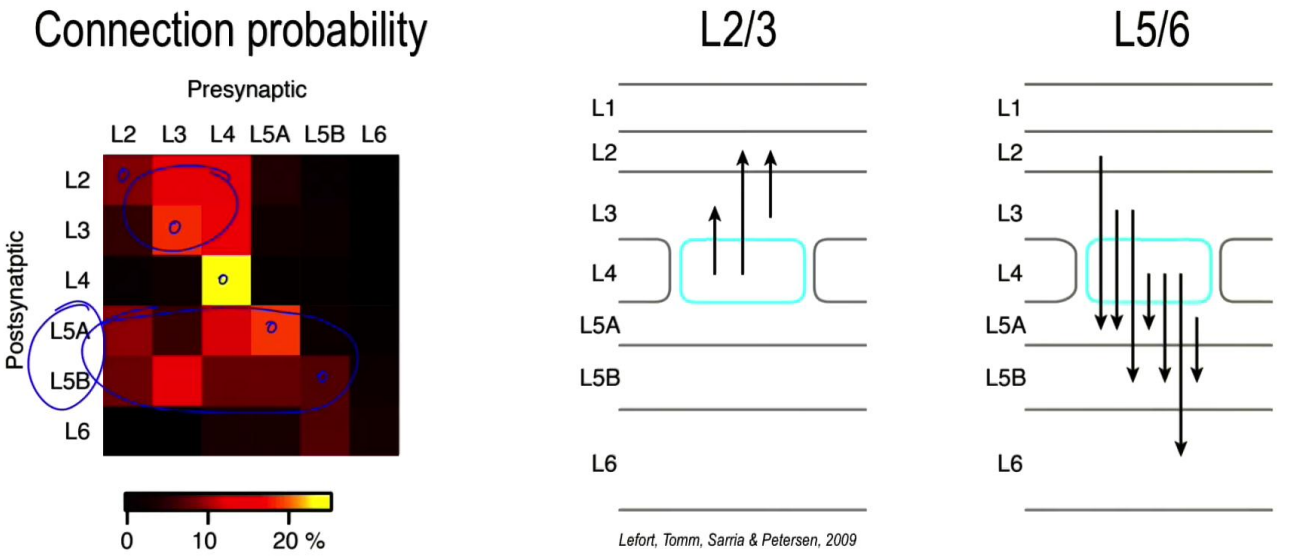
So here's the end result of the analysis after recording from hundreds of pairs of neurons we find that there's a connection probability matrix that we can construct where we take the location of the presynaptic neuron in terms of which layer of the neocortex it's in, and when we look and see how often that connects with postsynaptic neurons again, ordered according to which layer the SOMA is located in. And if we now start with neurons that are located in presynaptic layer 4, we'll see that they make connections onto postsynaptic cells in layer 2,3,4,5A,5B and a little bit in layer 6. So layer 4 neurons send the axons and make synaptic connections with neurons in all other layers of the neocortex. On the other hand, if we think postsynaptically about neurons in layer 4, we'll see that they don't receive any input from layer 2,3,5 or 6 and they almost exclusively receive their input from other layer 4 neurons So layer 4 is a very tight local processing area when neurons within layer 4 like to talk to each other and they also like to send signals to other parts of the neocortical column but they don't want to receive information from these other layers.

Notes

Summary

19m 39s

Excitatory microcircuit of the C2 barrel column



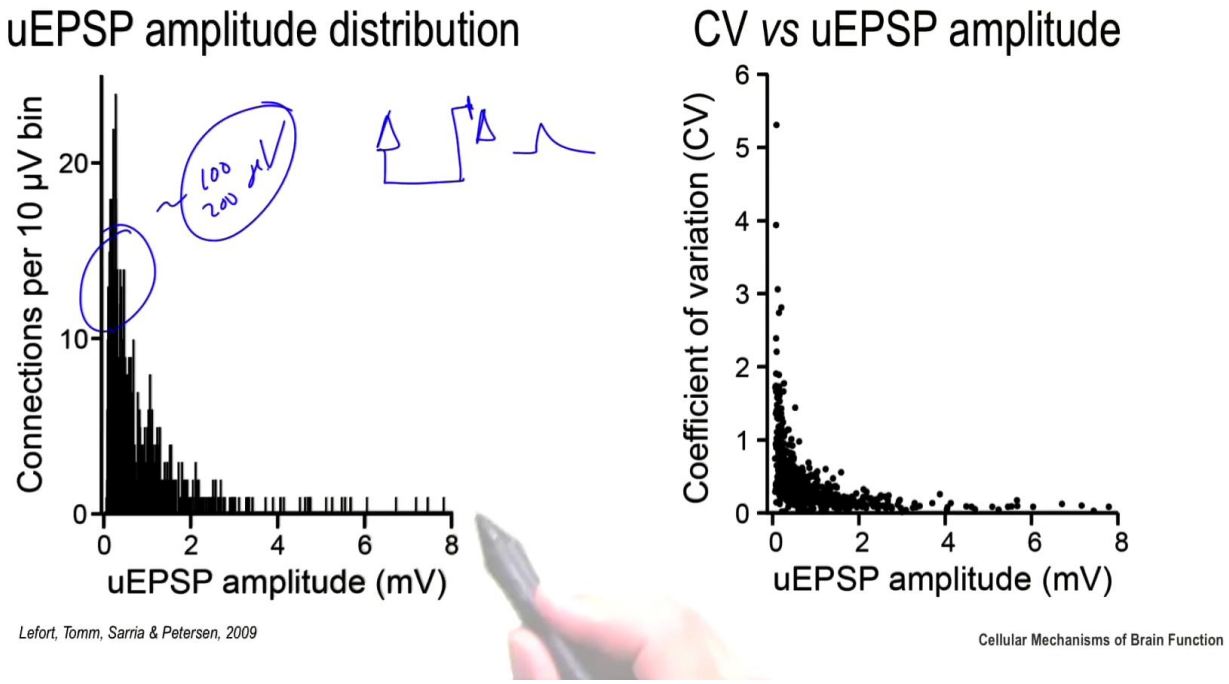
We can also see that there's a strong diagonal component where neurons within layer 2, within layer 3, within layer 4, 5A and within layer 5B like to talk to each other. They like to talk to their neighboring cells that are sitting right next to each other. That's one of the ways in which they make connections, simply by being close to each other. Beyond that, there's further specificity. The superficial postsynaptic neurons, they get information from presynaptic layers 4,3 and 2. And so here, you'll see that the layer 2 and 3 neurons are receiving information from these circuits here. Probabilistically, that's where they're most likely to be getting their synaptic input from. On the other hand, if we look here in the deeper layers, in layer 5, you'll see that they get synaptic inputs from almost all other layers, they get information from layer 2,3,4,5 and perhaps a little bit from layer 6. So these neurons here are integrators, they receive information from all other parts of the cortex and presumably, they use that information to distribute some further processed signals to other parts of the brain. There's a higher degree of specificity in the directions of which sensory information gets distributed to different cells in the neocortex and what remains to be learned from further experiments is what the purpose of that computation is within the neocortex.

Notes

Summary

21m 11s

Synaptic properties



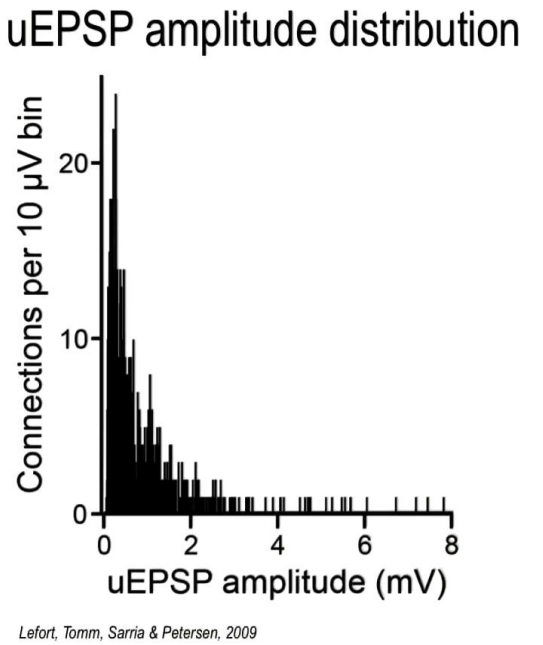
If we now look at the amplitudes of the postsynaptic potentials that are found inside the C2 microcircuit, we find that there's a rather interesting distribution of unitary EPSPs. Most of the synaptic connections are very small in amplitude. They're sitting there somewhere at around 100 or 200 mV. So, an action potential in a presynaptic neuron on the whole gives a rise to a relatively small postsynaptic potential of the postsynaptic neuron. It's just a few hundred mV. Almost nothing compared to the amount of depolarization that's necessary in order to get this postsynaptic cell to fire an action potential, it needs some 30 mV of depolarization, and that 100 mV is going to make a small impact. However there are also some rare synapses within the neocortical microcircuit that are much, much larger and so we can get synapses that are at 5 and even 10 mV in amplitude in response to a single action potential. These synapses then could give rise to a considerable impact, postsynaptically, and contribute strongly to driving postsynaptic action potential firing.

Notes

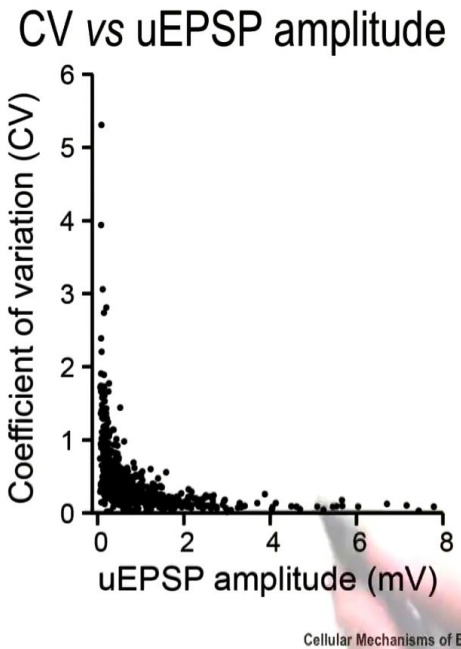
Summary

22m 47s

Synaptic properties



Lefort, Tomm, Sarria & Petersen, 2009



Cellular Mechanisms of Brain Function

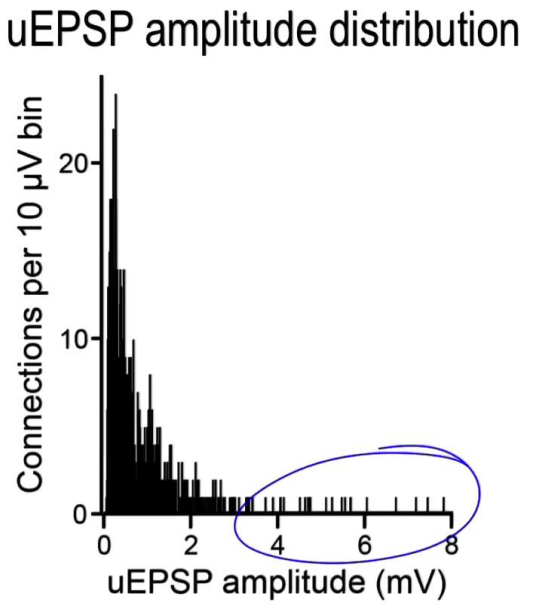
And so we are left in an interesting situation where there's a few very strong synapses and there are very many, very weak synapses and at this time, it's still not completely clear which synapses are most important in terms of driving neocortical microcircuit activity. Computational studies suggest that one can remove all of these small synapses and it makes very little impact upon the neocortical microcircuit activity. It may therefore be that larger amplitude synapses are the main drivers for reliable sensory processing in the neocortex, and it may be that a very sparse set of connections, which are large in amplitude, might dominate over a seed of very small connections that make almost no impact upon the larger neocortical microcircuit. Large unitary EPSPs might be a fundamental importance for reliable signaling in the brain. Further indications that some of the liable processing might occur through these big amplitude connections, comes if you're looking at the variance of the EPSP amplitudes. in here we compute the coefficient of variation which is equal to the standard deviation over the mean.

Notes

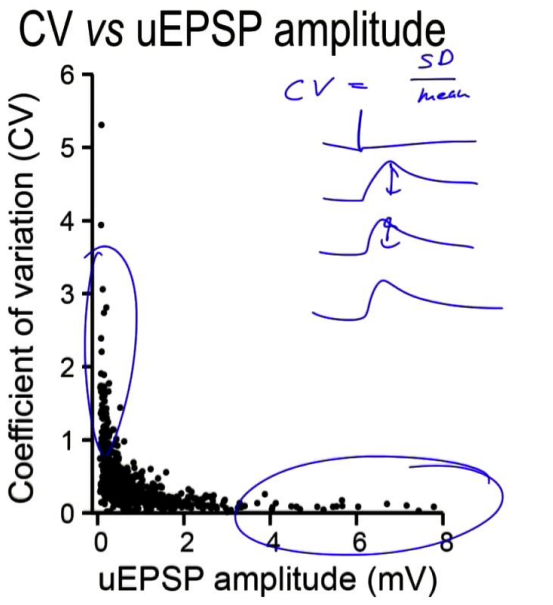
Summary

24m 12s

Synaptic properties



Lefort, Tomm, Sarria & Petersen, 2009



Cellular Mechanisms of Brain Function

In terms of the trial by trial EPSP amplitudes that we measure for the same connection in response to a given action potential, and you'll remember that there's some variance. We've already discussed that large synapses might have low variance whereas small synapses have much more variable responses. That then, is another indication that if you want to have reliable sensory processing, you might want to use these big connections that not only have a big impact on the postsynaptic neuron but they're also reliable with low variance from one trial to the next.

Notes

Summary

25m 37s

Glutamatergic synaptic circuits

- Excitatory glutamatergic neurons send long-range axons linking distant brain areas, *e.g.* glutamate synapses are involved in signaling all sensory information from periphery to cortex.
- Glutamatergic synapses in local neocortical microcircuits connect specific cell-types with diverse synaptic properties.

Cellular Mechanisms of Brain Function

So here in this video we've seen some important aspects about glutamate synapses. First of all we've seen how glutamatergic synapses are essential for relaying all sorts of sensory information from the periphery to the neocortex where it's thought that sensory percepts are generated, so glutamatergic signaling is essential for sensory perception. We've also seen that within the micorcircuits of the neocortex, there are highly specific circuits that are made where some types of neurons like to talk to other types of neurons but might not want to receive information from other types of neurons, and so there's a great deal of cell type specificity in terms of where glutamatergic signals are made and how the sensory processing is likely to occur within the neocortex. In addition we've seen that there are some extremely large amplitude synapses that are one or two orders of magnitude, larger in amplitude compared to the large sea of small synapses that are present in the brain and these larger amplitude synapses might be particularly relevant for reliable sensory processing within cortical microcircuits. These synapses may in part be determined by genetic mechanisms.

Notes

Summary

26m 17s

Glutamatergic synaptic circuits

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Cellular Mechanisms of Brain Function

The different cell types express different genes and so genetically, one can distinguish different cell types in the neocortex and there may well be genetic factors that skew the wiring diagram of the neocortex in one way or another. It's also very likely that experience makes a big difference to the wiring of the neocortex. We learn how to perceive the world around us and it's likely that some of that learning as to how to interpret the sensory information occurs in the wiring diagram of the primary sensory areas. So the distribution of large amplitude and small amplitude synaptic connections in the brain may in part relate to experience depend on learning. Over the next video, we're going to see how synaptic plasticity through different patterns of activity can alter the strength of synaptic connections.

Notes

Summary

27m 39s