Cognition and its Computation
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Lecture - 09
Single Neuron Level Measurements

Welcome. So, you have been introduced to the ideas of measurement physiological measurements that have to that can be linked with cognitive aspects or some aspects of behavior from humans. So, measuring the activity of the brain and different parts of the brain and specifically if we go deeper and deeper from single neurons, is the key process by which we understand how brain processes information and how I mean we understand about the steps or the computation done by the brain.

So, you have been introduced to the ideas of EEG, FMRI and MEG. And now we will talk about more basic level information which is usually gathered from experimental animals and sometimes some of these can be applied in humans in very specific cases.

So, these methodologies which are the basis for all the knowledge that we gather about computation in the brain, is crucial in understanding our lectures beyond this point, where we will be using different techniques or different methodologies of measurement in order to understand perception, in order to understand attention, in order to understand emotions and so on.

What I mean not totally understand, I mean whatever we know is based on such measurements. So, as you know or as you have been informed that it is the electrical activity of neurons in the brain that convey the information that we need and as you have learned the EEG signals are summed sort of summed and very highly filtered version of the signal produced by many many neurons together.

And here we will try to see how single how we can measure from single neurons and what are the drawbacks and when can we perform such experiments, when we cannot perform such experiments, what are the advantages of such methodologies and so on. So, most of the knowledge to date about single neuron activity in organisms in animals is from the method that is called single unit electro physiology.

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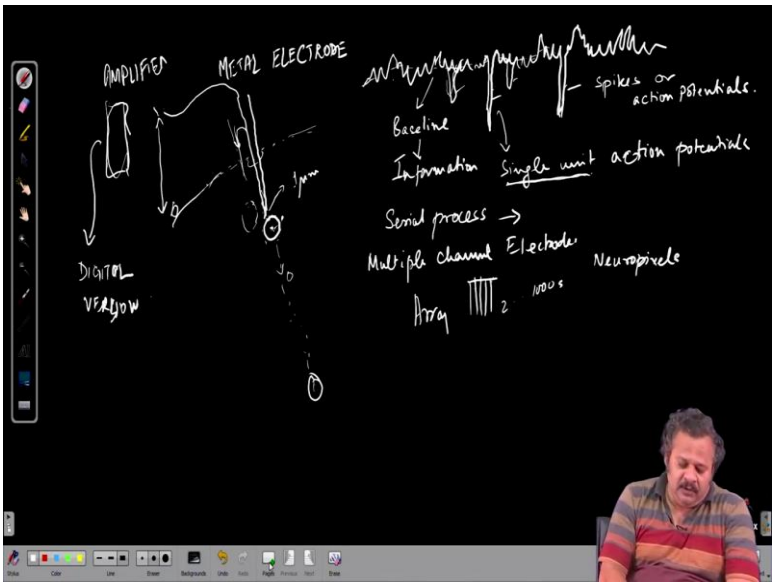

So, when a metal electrode a metal electrode is inserted into a brain tissue into some brain tissue. So, this tip may be up to even a one micron in diameter. And let us say we are able to insert the electrode beyond the skull by making a tiny craniotomy into the required region of the brain. And let us say we are close to very close to some neuron as we have mentioned it is the membrane potential or the potential or voltage across the inside and outside of the neuron that carries all the information.

It is we will try to get that information based on such electrodes, but only an extracellular signature of that signal. These signals are going to be small in nature, a very small in the sense about in micro volt ranges whereas, we will later discuss that the actual signal can vary between 100 milli volts to 0 milli volts. I mean in that range an order of 100 milli volts.

So, if we are sufficiently close and let us say this electrode is now connected with some reference ground on the skull and we are measuring the potential across this. And then it is amplified impedance matched and amplified through an amplifier and then read into a computer after making a discrete version or digital version of the signal. We can get a signal that would look something like this, some noisy waveform and sometimes there may be events in them, that look some like excursions that look like this.

So, this other signal is what we will call the other signal as in the signal that is apart from such voltage excursions is basically the baseline. However, it has a lot of information in

it in terms of what might be going on in the overall surrounding at the tip of the electrode. And these events are what we will later say or define and understand in more detail are the spikes or action potentials.

We will show that these are all are non events and it forms the basis of computation in the brain. So, with metal electrodes and extracellular signals that is the electrode is picking up the signal from outside the neuron near the cell body. So, for reasons that can be shown based on ah theoretical analysis that we can indeed get a signature of the v in or the voltage inside minus outside at the tip of the electrode, along with a filtered version of all the electrical activity in a sphere around that electrode tip.

And if we are close enough to one neuron we will pick up these events which are called single unit action potentials. And it may not be that it is always a single unit that is observed sometimes, we can get another neuron. Sometimes we can get another neuron that is also close by and the events from that neuron are also embedded in the same signal may be with a different shape or different height and so on depending on the geometry and the electrical properties of the tissue around the metal electrode tip.

And we may be able to differentiate the two different events based on the shape which is what we call as spike sorting and in that case we can say that we are recording from two different single units. So, we are gaining access into the brain from single neurons in that particular region of the brain or single units, because we are blind to what actually is the structure that we are recording from.

We may be recording from thick dendrite where also action potentials can happen we may be recording from the from an axon, we may be recording from an axon that is thick enough and the electrode manages to pick it up. Usually not on metal electrodes, but what single unit means that this is a single entity not necessarily a single neuron, but in most cases it can be that we are recording from the single neuron. But it is not right to call it a single neuron, because we do not know exactly where we are recording.

As you can imagine we it is like a fishing experiment in the sense that we are sticking an electrode into the brain tissue, let us say a mouse ah that an experimental mouse into its cortex. And we hope that we can get such events on our electrode tip and then we can manipulate the environment or around the mouse or the experimental animal by using different stimulus sets of different modalities or during engaging the mouse in some sort

of particular behavior after it has been trained or so on, depending on what question we want to ask.

We can then record these activities or action potentials or spikes of single units. Then we can with precisely move the electrode further down along the tissue and may be record from another set of neurons and so on. In the process we may be able to get responses or the activity of a few different neurons in the particular region. And then we may move take out the electrode and move it to a location beside it and go through the process.

So, this is a very serial process not a, I mean it is a serial process. And we can ah I mean we can we have limited number of neurons or single units that we can actually record from in such a process. But most of our knowledge in the neurosciences in more in terms of single neuron activity has come from such methods, in terms of the system level activity of neurons.

So, this being a serial process we are unable to understand what a network of neurons in a population are actually doing in order to understand, how activity of a population of neurons encodes or actually represents the external world. Or how it is how it reflects the behavior or the status of an animal or what have you depending on the questions we are asking.

So, that is a drawback and; obviously, people have come up with better techniques which essentially are multiple channel electrodes, multiple channel electrodes where essentially you have an array of electrodes that are implanted or actually gradually moved into the brain tissue. And there can be as few as two to as many as thousands of electrodes in a single array depending on the design and type of electrodes.

And more recently people have worked out even better techniques ah that is called the neuropixels probe which gives access to many thousands of neurons simultaneously. And that has its own problems also being that this being an invasive method, there are there are damage that is done to the tissue and it requires recovery and certain things that we cannot always control for.

And; however, we do have access to parallel channels that is we can understand or gain information about neurons simultaneously, under the same conditions from many

different neurons at the same time. So, that is the big advantage of using a multiple channel electrodes and we do experiments mainly in animals in experimental animals.

However, in special cases with such electrodes we may be able to also record from humans and many people have done that. And that is essentially in cases where humans or subjects have to go through a surgery where an electrode implantation is necessary. For example, epilepsy where we can piggyback experiments on top of the surgery, when the subject is willing and all the other ethical considerations are taken care of.

We can again access to such data from humans itself, but it is now becoming more common. However it represents a very small fraction of in terms of how much knowledge we have gained from such experiments and the total knowledge that we have right now based on animal experiments. So, these cases where we can record from humans are obviously, very valuable because we can never ask many such questions in animals.

And those become therefore, key features of our experimental knowledge about cognition and about the science or you know about neurons in general in the human brain. So, these are all ways in which we gain access to electrical information in terms of single unit activity or at the same time multiple units from multiple different electrodes. So, to go into a little more detail about the information that is available from the signal in the electrode.

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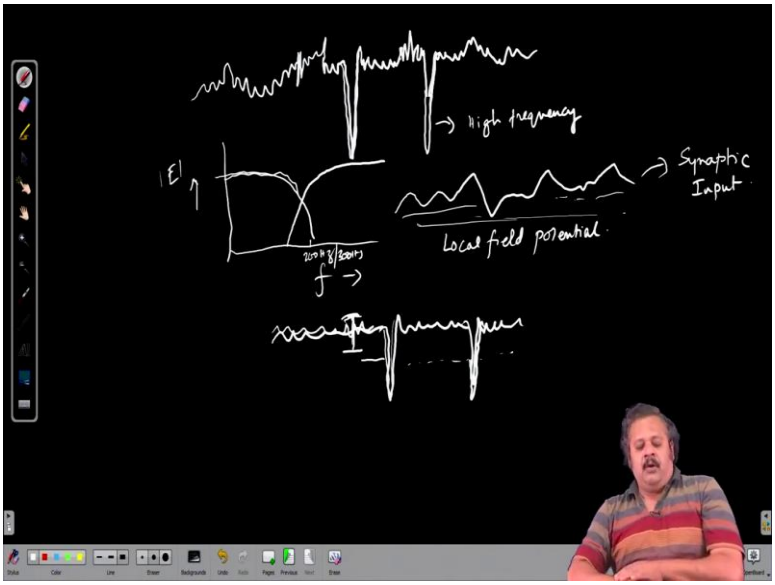

As I was saying let us say the signal looks somewhat like this it is a noisy background and then there are events that are what we will call the spikes. And there are other rhythms or changes in how the baseline is fluctuating over time. And as you can see these being very sharp changes in time are actually of content of very high frequency content, in terms of the overall signal frequency content these are the high frequency regions this represents the high frequency part of the signal.

And if we were to thus filter out the low frequency region this being the frequency axis and this being let us say a measure of energy or power in the signal at over a period of time and let us say we filtered out the signal energy up to 200 hertz or 300 hertz or so. The signal that we will result in is going to be something like this, I mean it is just a depiction. And the frequencies contained in this kind of a signal is up to let us say 200 hertz or 300 hertz.

And this is what is called the local field potential and these correlate well with some studies have shown correlate well with the FMRI signal, but over much ah longer time scales. And the overall energy can be much vary may be varying much at a much faster time scale. So, this local field potential as we discussed in EEG is like also the EEG and again contains the same sort of frequency bands with the same sort of implications in terms of the volume of tissue that we are recording from.

So, in EEG if you remember we are recording from a much larger region under the scalp of the electrode, if we are able to ah reduce the volume conduction from one electrode to other. We can actually localize it to a smaller region, but still it is nonetheless much much larger region over which the signal is captured; as compared to the local field potential that is captured with the metal electrode inserted into the brain.

So, the and the other part usually what is done is a high frequency part is filtered out that is above 200 to 300 hertz up to 5 to 10 kilo hertz. That signal is devoid of all these baseline fluctuations that we see and become more of a high frequency noise flattish baseline, with the events that we were interested in or that we are interested in. And if the recordings are such that we have large enough spikes and only single sort of shapes that is picked up on the electrode.

Then we can simply threshold it based on the baseline noise fluctuation range and find out what times these events have occurred. And as we will show later that since these are

the all or none-events we can actually identify these. And now we can convert this signal into almost a 0 1 signal. So, with these spikes we are only the spikes we are losing a lot of information as well, the local field potential actually reflects most of the synaptic input activity into the volume of tissue from which we are picking up the signal.

However, there is a lot of processing that can be understood and many other aspects of the processing that can be understood, if we can gain access to the overall v in minus v out of a single neuron. That is not extracellular, but being able to directly record the v in minus v out. In that case we will be able to know all the other types of fluctuations or information that is contained or that is based on what the neuron is firing action potentials we can gain access to that.

And these measurements are extremely ah difficult in awake, behaving animals or even if you speak about human subjects, I mean these are almost impossible. However, people have done such experiments where we gain access to the directly the v in minus v out in awake and behaving animals that is we can understand many aspects of cognition and processing at the single neuron level. Now, how do we do that?

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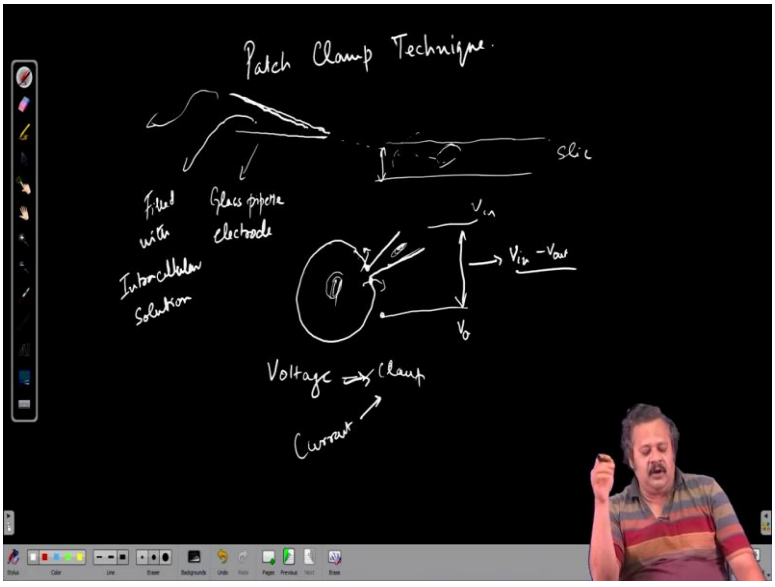

That is basically based on patch clamp methods. So, if we are able to. So, the experiment is done in such a way that under a microscope and sometimes blind, we can go in with a glass pipette electrode. That is basically the glass pipette is pulled into the shape of an

electrode, which has a opening at the tip and there is a metal wire inside that is whether it is going to carry the electrical signal information.

And this pipette is filled up with the intracellular solution of neurons. So, we have a fair good fairly good idea of what sort of ionic concentrations and other molecules are present and this and that solution is what is put into the inside of this metal electrode sorry in the glass pipette electrode. So, intracellular solution of neurons and it may vary from structure to structure based on where we are recording from and special types of neurons that we might be recording from. And for special questions we may be adding different things into the solution.

So, this glass electrode is inserted into the brain tissue either in a live brain slice or in neuronal cultures for other questions or even in an intact animal into the brain and in that case it would be blind. So, if we assume that we are inserting this into a live brain slice which is some 4 500 microns thick that is cut off from the that contains the particular types of neurons that we need and it is alive.

And we are trying to measure the different properties of the neuron. Then we can in we can actually under the microscope be able to see the neurons and approach that particular neuron that we want to record from with the electrode. And finally, what is done is that with the tip of the electrode we can suck the membrane of the neuron up to be tightly sealed to the sides of the glass electrode.

And so, there is no leakage of solution from the two sides or from around the edge where the electrode is touching the neuron. And so now, what happens is that the inside of the neuron and the inside of the electrode become ISO potential. That is they are at the same electrical potential, same solution, chemical potential everything is same. So, inside of the electrode and inside of the neuron is basically the same.

And now if we can measure in reference to the outside this is the v in this is the v out if we can capture this signal and amplify it we can measure exactly what v in minus v out is going to be over time. So, this is an intricate experiment many people do it routinely many neuroscientist do it routinely and this provides a huge amount of information and allows us to ask many different questions about the properties of the neuron and so on. In this particular scenario we will simply introduce two terms and that is two the voltage clamp and current clamp.

And we will be referring to it in only in some special cases. And that is that these measurements can be done under two conditions primarily I mean there are many other ways of doing these recordings.

But we will be talking primarily of two conditions; one is the voltage clamp and the other is the current clamp, which means that the voltage clamp is such that the membrane potential the v in minus v out is set fixed to a particular value over time by us or by the experimenter and we are able to measure the current that is flowing across the membrane of the neuron.

So, we will be discussing in more detail about the current and what we can learn from it. Similarly, we can fix the current that is flowing across the neuron which is called current clamp and measured the voltage v in minus v out over time. So, these two things allow us to control the neuron in particular ways as we will need in our discussions on how we gain access to spiking information or how spikes occur and how plasticity can be induced in between two neurons and so on.

These will be coming up in later lecture. So, we will end here with the ideas of voltage clamp and current clamp the two cases that can be used in order to understand more about the processing within a single neuron. And in the next lecture we will be talking about further other techniques that can be used in terms of understanding network level activity from with other methodologies and manipulating activity with other methodologies.

Thank you.