BioMEMS and Microfluidics
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Lecture - 31
Review Lecture of 20, 21 and 22

So, we would like to now review lectures twenty through twenty two and this lecture was mostly dedicated to the different strategies of hybridization based demonic detection. We started with the famous work by Alivisatos where there was an evidence of a Nano particle, the gold Nano particle. He mobilized to a single strand of DNA on its surface, would kind of pair up and accumulate together under a transmission electron microscope when there would be complementariness between the different DNA strands as they mobilized on to the surface of this gold particle. So, this was, in a way, one of the first example of bio nanotechnology, that how inorganic moiety at the Nano scale could be assembled or disassembled based on, if there is a complementary bonding between the different base pairs of two single strands of DNA molecule.

We also looked in detail at the work of, sort of immobilizing or hybridizing DNA again to capture probe, which was bound on a surface of a gold film and which had biotin target on one of the ends. And then, this polystyrene bead, which would be otherwise mobilized on the surface with an avidin would get coated on to this or which would get captured on to this biotin target, ok. And this, essentially, would depend on the complementary nature of the two DNA strands, which would be interconnecting the biotin target with the capture probe, which is immobilized on to the surface of the gold.

So, therefore, if there is a bonding, there is a complementary nature of the DNA, which is actually the target DNA with the probe DNA, which is there on the surface, there would be a presence of the polystyrene beads, as very clearly evident by the air from the scans or the air from the records and if there is no complementariness between the two probes, then there would be no beads on a surface.

Similarly, we also talked about the various strategies for doing hybridization based micro area detections, and in light of that we understood, that that there is large library of different probes, which are immobilized on to chip scale and then, each probe would be responsible for binding, for a binding event corresponding to its own target and this target would be labelled

fluorescently. So, therefore, wherever there is a presence of a target, would open up a pixel or a dot, which would be machine readable. And based on the intensity of the dot, one could figure out about the concentration of the target and also, the nature, the molecular nature of the target just because it bound to certain species, which was there in as a part of this particular array.

So, we also learnt, that there are two different strategies for doing this mobilization and making this micro chip one was the electronic placements strategy where would use electric field on small electrodes to get the DNA very close to the surface, so that it could bind on to a small, you know, lysine layer on the top of the surface. And the other strategy, and so, this was followed, typically, by the Nanogen and this would also be better known as the Nanogen approach.

The other that we discussed was the light directed synthesis of the DNA where there would be a base pair. By base pair fabrication, by different lithography steps added on to the whole assay where you could eventually grow a DNA or a probe sequence of the desired type with several combinations, which are available on the way, that you change the chemical patterning technique. So, it is basically a photochemical patterning technique that you are using for growing up DNA on the surface. So, this was popularly called the Affymetrix. Both these companies actually have sold quite a bit of these microchips all around the world for molecular diagnostics.

So, we also detailed, discussed about certain other mortalities of sensing of DNA using electrochemical labels. For example, we referred to the famous Motorola biochip where using molecular wires we could sense whether DNA would immobilize on to the surface with a ferrozine level or a probe, which would be electronically sensible. So, essentially, with electrochemical sensing we could see whether the right binding event was happening on the bed of a molecular Nano wire.

Similarly, we also looked at various other sequencing mechanisms and in context of that we discussed the Sanger's reaction we also talked about how AFM tips are ideated to be popularly being used for surface electrophoresis of these DNA, so that you could have a fast read out system based on fluorescence or some other, you know, tag based mechanism. So, you could actually read a base pair by base pair map of whatever DNA was incorporated.

We, in a detailed manner, looked into Nano pores and the way they behave in the presence of electrical fields by translating DNA across the pore and it would typically act as a, as an ion channel. We looked into the fabrication strategies of some of the inorganic Nano pores and how they would fabricate with things like SOI for example, silicon on insulator, so on, so forth.

We also discussed about mechanical cantilevers and how mass based sensing could be utilized for studying binding events. The famous example that was discussed was the very first AFM tip, which was developed at IBM and how that could be used for DNA hybridization array. We also talked little bit about how this could be used for even protein-protein binding. So, the mass based cantilever approach can be used for a protein-protein binding to find out or detect very small mass limits of, of protein molecules.

And then, finally, we detailed into the protein structure, the basics about what are the different kind of confirmations that the protein would have. We also looked at how the proteins would be produced within living cell by this huge Nano machinery, which is at work where a simple DNA would be transcripted at the first stage to a small messenger RNA, which would eventually lead to the formulation of a completely different translated sequence of molecules or amino acids, known as proteins. So, in, in a nut shell, we covered all these in lecture 20 to 22.

Thank you.