Annual Report for AOARD Grant FA2386-11-1-0001 "**Research Title**" Study of opto-electronic properties of a single microtubule in the microwave regime,

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Abstract: For the first time in the world we have isolated single brain microtubule and measured its electronic properties using four probe technique. One of the greatest challenges of the AC measurement on the nanowires is that the measuring probes communicate among themselves and sends signal to the measuring instrument without sample under measurement. Therefore, this is a typical environment where AC signal flows continuously through the air and identifying the signal from samples to the noise is nearly impossible. In this situation, we are developing a number of technologies so that during AC measurement, we can significantly reduce the AC contribution through Air. Here in this years report, we describe the technological challenges in this respect.

Introduction: We started working on the brain microtubule way back in 2008, since, I understood that in the brain, neurons separated by 6 inches, synchronize, get phase and frequency locked and that is the source of enormous computability of the brain. However, no experimental evidence existed at that point of time. We became the first group in the world to study single microtubule electronics reliably and identify resonance band of these biological architectures, which was very essential to understand synchrony that would lead to phase and frequency lock conditions in the entire brain. Another problem bothered us for a long time; if we want to compute similar level of complex problems, we need 1000s of megawatts of energy; how brain does that using only 25 watt? Finally, that I desperately wanted to confirm was the potential of synchrony in brain computing, does brain execute non-linear frequency pulling to introduce simultaneity and drastically reduce the computing time?

Therefore, we need AC source in the brain to make our result practically feasible. While it is extremely difficult to build GigaHartz sources by biological cells, we often observe that they produce kilohertz signals like heart beat and neurotransmissions. We found experimentally that proteins can generate megahertz signals even under DC current flow through the protein structure. This experiment we have attempted to repeat in the most reliable way possible. One of the biggest challenge of the four probe measurement is that the probes are naked in the environment when it contacts with the chip during measurement. These problems are universal, however, for microtubule, this is even bigger because of the reason that unlike conventional materials, microtubules are purely insulators, and this makes microtubule more vulnerable to the external signals, at the pico-ampere level. Therefore, obviously, we have taken outmost care to deal with this particular problem.

Experiment: Porcine's brain, Tree, Algae extracted tubulins (Cytoskeleton, Denver, CO), were preserved at -80o C. To polymerize tubulin into microtubules, Microtubule cushion buffer (60 % v/v glycerol, 80 mM PIPES pH 6.8, 1 mM EGTA, 1 mM MgCl2) was added to General Tubulin Buffer (GTB, 80 mM PIPES pH 7, 1 mM EGTA, 2 mM MgCl2) and/or GTP solution. The mixture is added to 1 mg of tubulin. At 37o C, 10 mM tubulin leads to an

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14. ABSTRACT
This is the first annual report of a three-year project. In progress to date, the research group has isolated a single brain microtubule and measured its electronic properties using a four probe technique.

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uncontrolled growth of microtubule >100μm, we maintain this tubulin concentration for all experiments. GTP, TX (Taxol), VB (Vinblastine), K352 (Pironetin), N-termini, tau, CH (Colchicine) were optionally added to this solution individually or as a combination, produced microtubules were isolated for single nanowire measurement of resonance levels. Ultrasound power varied between pico-watt to femto-watt, if Mg2+ is not added, effect of ultrasound pumping is not observed, so Mg2+ is essential, GTP is not. Phase coherent signals were measured for all combinations of tubulin mixtures when placed in a heat-bath for pure ac pumping. The heat bath of Fig. 2a was placed inside the Raman measurement chamber and growth profile was measured with 532nm laser light from basic Raman spectrum switching between protein and microtubule nanowire shows convergence of Raman vibrations. We have ruled out possibilities of other physical processes detailed arguments in online text B. Rejection arguments: Rejection of DEP, rapid crystallization, ion-induced growth, self-assembly in the collapse of matter. For consistency, all figures in this paper are produced from porcine brain tubulin.

We have measured all possible electronic properties of a single microtubule nano-wire. Using a four probe system, we send current from the outer two probes and measure voltage drop in the two electrodes between. During AC measurement, we follow two different protocols, one, we either send AC signal to the central two electrodes or send signal at the external two probes. In both ways, we try to generate resonant oscillations in the microtubule, and try to build up vibrations as largely as possible.

Results and Discussion: Here we describe the experimental results. Following ac pumping, we take out the chip, identify single microtubules of different lengths as shown in Figures 1a, and deposit four gold electrodes on single microtubules as shown in Figure 1b. This allows us to stimulate and record response from separate pairs of electrodes on a single microtubule. We vary the length L from 200nm to 25μm; for any length of microtubule, current-voltage characteristic, resistance variation as a function of injection current, power transmission; positions of resonance peaks are so identical that for all cases it appears to be a single plot (Fig. 1c,d). The finding is significant since, if one takes a cluster of 300 proteins and increases the number of protein molecules to the condensate at a time, until it reaches 40000 protein clusters, there is no change in any electronic property.

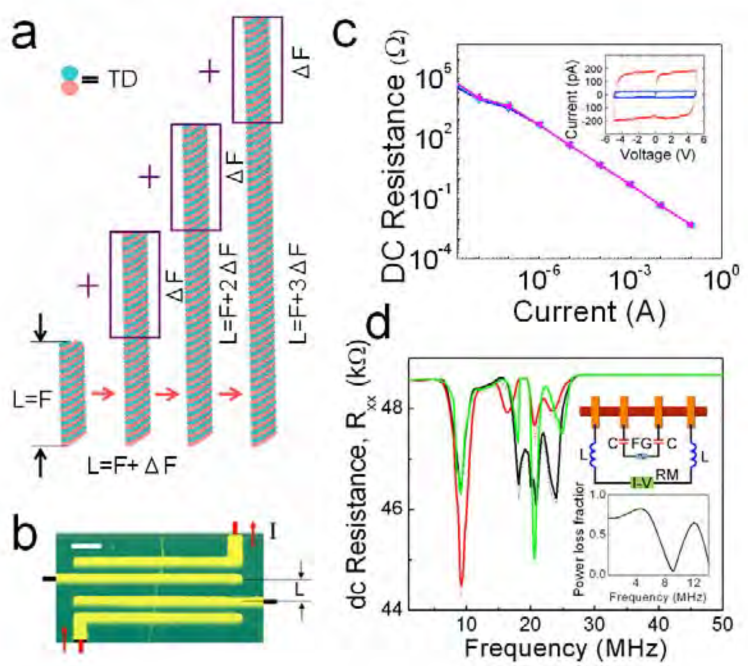

Figure 4. Alleviation of size effect in a condensate: a. We vary L=200nm (F) to 2μm, at a

gap of 200nm (ΔF) made by 25 rings (top); electronic properties for these L MTs are plotted in C,D. **b.** AFM image of a four-probe MT-device, scale bar~300nm. I is current direction, voltage drop is measured between inner two electrodes (gap L). **c**. Change in resistance for the source current 1pA-1nA for all L. Current voltage characteristics of all MT devices at 300K (inset, red→without ground, blue→ground). **d**. In a four-probe MT-device, decrease in dc resistance (RM) that accounts degree of resonance at two outer probes is plotted with the ac frequency applied between inner two-probes (FG) for L=600nm (black), 1μm (red), 1.5μm (green). We find identical 8 peaks for all L (Fig. 4a), though intensity differs. Power loss spectra for single MT as function of frequency for all L are superimposed (inset).

Thus, similar to atoms in Bose condensate, quantity of proteins become irrelevant, only one state with one frequency and one phase represents entire microtubule but changing one tubulin state does not change 40000 others, so they are not entangled. Fröhlich or Gibb's condensates20-27 do not have such provision, which justifies assigning microtubule as X-class bio-condensate. Figure 1d shows that intensities of resonance peaks vary as a function of L, however, statistically, resonance peaks strictly occur at seven frequencies in the high-radio-frequency domain and one in the very low radiofrequency domain. Thus, during dynamic instability & transport, microtubules interact varying the intensities of resonance peaks--while all other properties are kept constant.

List of Publications:

1. Synchrony induced ultra-fast growth of microtubule and its length independent electronic properties S. Sahu, S. Ghosh, K. Hirata, D. Fujita, A. Bandyopadhyay Nature Materials (in press).

2. Computational myths and mysteries that have grown around microtubule in the last half a century and their possible verification S. Sahu, S. Ghosh, D. Fujita, A. Bandyopadhyay Journal of Computational and Theoretical Nanoscience (Special Issue) 8, 1-7 (2011), also selected for cover page

Lectures given in the international conferences and in the universities.

1. Remarkable electronic properties of a single Microtubule Google Mountain view campus, workshop on quantum biology 22 October 2010
http://www.youtube.com/watch?v=VQngptkPYE8

2. Practical computing with organic molecules Design and synthesis of a 3D nano brain, International symposium for Young Organic Chemists, Tsukuba, Japan March 1-3, (2011)

3. Quantum aspects of microtubule: Direct experimental evidence for the existence of quantum states in microtubule, Towards a science of consciousness May 2-8 (2011), Sweden

4. Electromagnetic energy of cells and microtubule: how microtubule research will revolutionize the human technologies, Czech Republic 1-3 July 2011

Lecture given in University of Arizona
http://streaming.biocom.arizona.edu/categories/?id=143

DD882: The invention disclosure process is underway. We hope by the end of the next year, we will be able to submit the invention disclosure with significant details.

This document may be as long or as short as needed to give a fair account of the work performed during the period of performance. There will be variations depending on the scope of the work. As such, there is no length or formatting constraints for the final report. Include as many charts and figures as required to explain the work. A final report submission very similar to a full length journal article will be sufficient in most cases.