**Original texts for demo edits**

**Demo Edit Unit 1:**

Immortality is an alluring concept. Some scientists believe that it will be possible to "upload" one's mind by recreating the circuitry of the brain in silico. Before we can upload brains, we first must reverse-engineer neural circuitry and begin by creating a circuit map.

Electron microscopy provides the only possible method through which we're able to clearly visualize synapses and follow neural processes. Volumetric reconstruction of neural tissue using electron microscopic resolution is necessary to map neural circuitry. Focused ion-beam scanning electron microscopy (Knott et al. 2008) gives excellent quality images, but fails to process tissue pieces larger than 40 microns in diameter. Thin sections imaged with transmission electron microscopy succumb to the damaging effects of manual handling and section distortion. Thus, it's most prudent to use a method that images the block-face directly and is capable of imaging large block-faces. Serial block-face scanning electron microscopy (SBEM; Denk and Horstmann 2004) provides both necessary components.

Using SBEM, Dr. Kevin Briggman and associates (Briggman, Helmstaedter, and Denk 2011) recently mapped the connections between starburst amacrine cells and bipolar ganglion cells in the mouse retina to better understand the wiring specificity, elucidating the cellular circuit between starburst amacrine cells and direction-selective bipolar retinal ganglion cells.

By staining a 200-micron piece of retina, which contained the entire arborization field of a starburst amacrine cell with an extracellular stain that could outline cells and neural processes in SBEM, Briggman was then able to reconstruct neural processes. Based on morphology, he assessed the locations and sizes of putative synapses on these processes.

Unfortunately, synapses were invisible within the data because the tissue was only stained with an extracellular, electron-dense stain and some synaptic features are intracellular. In an effort to address this ambiguity, Briggman then stained a second piece of tissue where synapse-associated features were stained and visible. He then correlated the extracellular morphology found at synapses between the first and second pieces of tissue.

This is the first example of relatively large neural circuit reconstruction and it solved controversy about exactly how starburst amacrine cells are wired to be directionally-selective. The next steps in whole-brain circuit reconstruction will be large sample preparation (Mikula, Binding, and Denk 2012) and imaging on a whole-brain SBEM for mapping the whole mouse brain as a first mammalian complete connectome (Seung 2011).

**Demo Edit Unit 2:**

Scientists are still looking for small size smart robots that can navigate in dynamic and unknown environments. This challenge inspired Tahmid Latif and Alper Bozkurt from North Carolina State University to use cockroaches as biobots (biological robots). They developed a wireless biological interface that uses an electronic interface to remotely steer cockroaches. This concept helps to create a mobile web of sensors that uses cockroaches to collect and transmit data, such as locating survivors in hard areas during earthquakes.

Cockroaches have antennas -called cerci- to sense: tactile, temperature and humidity. Researchers used these antennas to drive the cockroach by sending a series of electrical pulses to it. The system consists of: a microprocessor with Zigbee interface [1], electrodes and a battery. The user controls the microprocessor wirelessly using a Zigbee transceiver; the microprocessor sends electrical pulses to the cockroach’s antennas using electrodes and then the cockroach moves.

Tahmid Latif and Alper Bozkurt used Madagascar Hissing cockroach during their analysis because of: its larger size (~50-75mm), slow speed (~3cm/s), long life span (~2 years) and robustness. Before the experiment starts, they anesthetized the cockroach by cold-treatment (4C) for 45-60 minutes. They attached one side of the electrodes (5cm long stainless steel coated with 250um thick Teflon) to the antennas to serve as electronic reins, injecting small charges into the roach’s neural tissue. The charges trick the roach into thinking that the antennas are in contact with a physical barrier, which effectively steers them in the opposite direction. The researchers evaluated two microprocessors that control the electrodes: Microchip's PIC16F630 [2] and Texas Instrument’s CC2530 [3]. CC2530 was better because of its low weight (500mg), Zigbee module connectivity and the availability of 21 general purpose I/O. CC2530 gets its power from the 90mAh Li-Po battery.

Cockroaches followed an S-shaped trajectory drawn on the laboratory floor and spent 81 sec. with 10% success rate to complete the route. This finding opens the door to scientists to start using insects in biobots world but the system's overall weight is still a concern in this new field and needs more studies to reduce its size.

**Demo Edit Unit 3:**

Traditional methods for controlling biological signals in cells are a sledgehammer: they are global, slow, and often non-specific. The authors of this paper describe their new technique to generate local, fast, and targeted cell signaling in live cells that are genetically altered to have light-sensitive proteins. They engineered a cellular perturbation system applicable to many signaling proteins. The main requirement for the candidate signaling protein is to be naturally activated by interactions that re-localize it to the membrane.

Levskaya et al. built this membrane recruitment system using photosensitive proteins named Phytochromes. These proteins from plants detect red and near-infrared light through the photoisomerization of a bound chromophore. This light detection changes the Phytochrome’s conformation between a state under red light that binds directly to a phytochrome interacting factor (PIF) and a state under infrared light that doesn’t bind to PIF. The scientist added a membrane-localization part to the Phytochrome, and attached a signaling protein to the PIF to complete their system. A cell illuminated with infrared light under the microscope will have inactive, free-floating, PIF-attached signaling proteins. When the scientist points a red laser in the phytochrome-rich membrane, the PIF-attached proteins are forced to stay close to the membrane; effectively increasing the activity of the signaling proteins. Turning off the red laser frees the proteins and turns off the cellular signal.

To demonstrate the feasibility of this new technique they focused on the signaling proteins Tiam and intersectin, precursors of the Rho-GTPases Rac1 and Cdc42 that have crucial role in the organization of actin cytoskeleton during cell movement. They performed three main experiments: The first experiment tested if membrane recruitment of a small part of intersectin (ITSN-DH-PH) that regulates Cdc42, was effectively inducing transient increases of local protein activity. They shown images of local enrichment of biosensors responsive to Cdc42 activity in the membrane that disappeared few seconds after turning off the red laser. The second experiment tested if membrane recruitment of a part of Tiam (Tiam DH-PH domain) was sufficient to induce changes in the shape of NIH3T3 cells. They illuminated the whole cell with red light for 20 minutes and inmediatly after counted the percentage of cells that made new lamellipodia (actin cytoskeletal projection on the mobile edge of the cell). The result was that almost 80% of cells made new lamellipodia under red-light treatment, compared with a 10% of control populations. To make things even more interesting, in a third experiment they pointed a red laser dot on the edge of one cell and gradually moved it outward, slowly extending this red-targeted region from the cell body. They show in movies that they effectively guided the direction followed by the new lamellopodium-- the first reported control of cell movement in real time using light-sensitive proteins!

**Demo Edit Unit 4:**

In a recent work on ‘Interactions with Big Data Analytics’, authors Danyel Fisher et.al. talk about interesting developments in the world of analyzing data. Authors define analytics as a term that refers to any data driven decision. An example of application of analytics is Zynga, an online games company that studies how its audience plays the game and uses that data effectively to modify the games.

The paper reports the state of practice by interviewing sixteen pioneering analysts in this field. The paper discusses about the definition of big data, contemporary ways of analyzing data, challenges peculiar to big data, and proposes a five step workflow type of an approach to analyzing big data. In our digital lives (interactions through information technology devices) we generate huge amounts of data: social relationships, purchasing behavior, watching of videos, etc. Big Data Analytics aims to construct the big picture from the minutia of our digital lives.

The authors draw a refreshing parallel to the old age mainframe computing where the work would be submitted to massive systems and the results would be obtained after a period of time. Big data analytics, argue the authors, is very similar: that it involves hypothesis and needs huge computing power, that it is often submitted and results are available after a period of time, and that the end user computers are only used for viewing the results and not for processing.

Pivotal contribution of the paper is the generalization of how big data analytics can be approached. Acquiring data, choosing the right architecture for analyzing the acquired data, fitting the data for the chosen architecture, coding and debugging, and fine tuning are the five steps suggested by the authors. This five step process repeats itself as many times as necessary until meaningful results are obtained. The paper cautions the skill gap in bringing the right proportion of scientific flavor in models created by business users.

Of immediate significance, is the potential to apply big data analytics to design more user friendly interfaces, enrich customer experience by analyzing the ways customer uses the product, understand healthcare spending, etc. The limitation is only our human ability to think creatively and harness the exploding world of data.

**Demo Edit Unit 5:**

Reactive oxygen species (ROS) are highly reactive chemicals often associated with escalating warfare between pathogens and their hosts. For example, ROS are integral to biological defenses, such as the respiratory burst in phagocytes of animals and programmed cell death in plants, to ward off microbial infections. In a landmark study, published in the journal Plant Cell, Tanaka and colleagues recently uncovered an additional role for ROS as “regulators of symbiosis.”

The team studied a symbiosis between perennial rygrass (Lolium perenne) and a fungus (Epichloë festucae) that lives endophytically (i.e., inside) the grass. The mycelium of this fungus—composed of cells called “hyphae”—colonizes all leaves of the plant, but the hyphae sprout only sparsely in tissues, never breach cell walls or membranes, and grow in perfect synchrony with the leaves of its plant host. This exquisite harmonization of fungal and plant growth directs resources to the production of fungal toxins that protect the symbiosis from herbivores. But how this harmonization is achieved and what its underlying mechanisms are have remained a mystery.

To address this question, Tanaka and coworkers generated random mutations in the E. festucae genome. They used a method called insertional mutagenesis, in which DNA pieces are randomly inserted into the fungal genome in the hope of disrupting a gene resulting in observable growth changes in symbioses with these fungal mutants. They indeed found a mutant showing a highly unusual growth pattern: unlike the synchronous growth of the wild-type fungus, the mutant hyphae showed profuse and abundant proliferation throughout the grass. Moreover, plants infected by this mutant showed poor growth and often died.

This set the stage for the next step: finding the genetic change that had caused these aberrations. Using molecular genetics tools, the researchers homed in on the gene the DNA insertion had disrupted. Surprisingly, only a single integration event had caused this abnormal growth mutant—it had disrupted a fungal gene the researchers named noxA.

To get an idea of what the encoded protein (NoxA) does, the team first compared its sequence with those of enzymes with known activities. They noticed that NoxA was very similar to NADPH oxidases, enzymes that are often involved in generating ROS in cells. Indeed, when the researchers next looked at ROS production in the plant, they observed that ROS accumulated only in plants infected by the wild-type fungus and not in those infected by the noxA-disrupted mutant fungus. This confirmed that NoxA is involved in ROS production required for proper functioning of the symbiosis.

The study by Tanaka and colleagues raises tantalizing questions. Chief among them is what the mechanism is through which ROS help maintain a functional symbiosis. The researchers suggest that ROS could be involved in establishing physical connections between the cell walls of the plant and fungus. Alternatively, ROS may play a role in symbiotic signaling: their short half-life predisposes them for cellular communication, perhaps facilitating an interspecies “Morse code” that helps maintain the symbiosis. If so, identifying the plant sensor and signaling pathways involved could provide deeper insights into how plants recognize and interact with beneficial symbionts and can distinguish them from pathogens.

**Demo Edit Unit 6:**

Sixty percent of world population is afraid somehow to go to the dentist, but fifteen to twenty percent suffer odontophobia. The World Health Organization recognizes odontophobia as a severe fear of the dentistry and of receiving dental care. This fear often leads people to avoid going to the dentist until emergencies happen, requiring invasive treatment which can reinforce their fear of dentistry – the avoidance cycle- . Odontophobia symptoms are hyperventilation, tachycardia, hypertension, sudden drop in blood pressure, transpiration, insomnia, mouth dryness, nausea and vomits among others.

Odontophobia can be treated in combination of behavioral and pharmacological approaches. The use of pleasant odors (like fresh bread or coffee), warm colors of the place, pleasant music, or beautiful paintings on the walls are effective techniques that dentists around the world are using. A recent study from the National Italian Union of Dental Industry showed that sensorial stimuli are decisive from the psychological perspective of patients and can be preferable than the use of anxiolytics. Intraligamentary anesthetics, computer-controlled injections and nitrous oxide sedatives are pharmacological ways that offer no-pain solutions.

The study concluded that dentists can manage odontophobia by taking some simple steps: Transmit serenity and friendliness by taking time to chat with the patient; tell and show what is going to be done (which is especially effective with kids);  offer earphones to isolate them from external sound and at the same time to provide music therapy; adjust the chair to the patient dimensions; provide an image of quality and professional demeanor; offer a well-illuminated place with plenty of space, and offer no-pain anesthetics.

By now, oral hygiene continues to be the best way to avoid going to the dentist, but that doesn’t solve the phobia. Researchers are now focusing on the use of internet as a way to confront anxiety, suggesting the effectiveness of online support communities on helping odontophobics to successfully receive dental care.

**Demo Edit Unit 7:**

Bunch compactness is a major factor affecting the quality of wine and table grapes. Compact bunches show favourable conditions for the development of different grape pests and diseases, such as the moth Lobesia botrana (1,2) or the rot fungi Aspergillus spp. (3,4) and, especially, Botrytis cinerea (5-11). The presence of these phytopathogens reduces crop yield and grape and wine quality, thus dropping economic profits (12). Among the reasons given to the major incidence of these organisms in compact bunches, some authors have pointed out the poor air circulation and sun exposure of the inner parts of the bunches (5, 13), as well as different changes in the epicuticular wax layer development in the areas where berries are in contact (14-15). Moreover, berries may burst due to high pressure inside compact bunches (13), providing water and nutrients for the growth of these organisms. On the other hand, the number of interior berries increases with bunch compactness (5). These berries may not receive the sun irradiation needed to achieve an adequate phenolic maturity, leading to a heterogeneous ripeness of the bunch. Consequently, consumers, food industry and winemakers prefer grape bunches with certain values of compactness considered of higher quality (16-17).

Although bunch compactness is a trait with a large agronomic and commercial relevance, little is known about its genetic basis. Some reasons might be its multifactorial nature and the difficulty to obtain objective and quantitative data for this trait, needed for an accurate phenotyping. Many studies (18-23) estimate it according to the visual descriptor proposed by the International Organization of Vine and Wine (24), while other authors have developed specific visual rating systems for its evaluation (24). Thus, the lack of a globally accepted criterion and the subjectivity linked to a visual system makes it difficult to compare results between different studies. Trying to solve it, and looking for a quantitative evaluation of bunch compactness, some authors have indirectly evaluated this trait through the determination of other characteristics of the grape bunch that vary with compactness. For instance, studying the degree of compression between the berries, measuring the force required to create a certain gap between two contiguous berries (5,6) or the suppleness of the bunches, determining the bending angle of the bunch (25).

On the other hand, several studies have proposed various relationships based on metrics of components of the grape bunch for the estimation of bunch compactness (26-30). Thus, this trait has been indirectly estimated (I) volumetrically, evaluating the empty spaces that appear in bunches as their compactness decreases (31,32); (II) by the number, weight or volume of the berries per centimetre of rachis (33-37); and (III) by the relationship between the weight of the bunch and its morphological volume (28), ratio that can be considered as the average density of the bunch. These estimations have been published in literature in the form of indexes, and they seem to be the most interesting system for the indirect evaluation of bunch compactness, mainly because of their simplicity, their potential applicability to different grape varieties, and by not requiring complex measuring devices and large cost investments for its evaluation.

In this sense, the aim of this study was to evaluate the usefulness of several indexes, either previously published in literature or newly designed, for an objective and quantitative estimation of bunch compactness that was useful for intervarietal studies of this trait.

**Demo Edit Unit 8:**

Ovarian cancer is the deadliest gynecologic cancer with a high mortality rate that has remained unchanged in the past four decades. The dismal prognosis of ovarian cancer is in large part due to the acquired resistance to chemotherapy. Epithelial ovarian cancer, the most common type of ovarian cancer, is initially responsive to cisplatin therapy. The recurrent disease, however, is often refractory to treatment and leads to mortality. New strategies to overcome drug resistance are urgently needed in order to reduce the mortality rate of ovarian cancer.

The discovery of small interfering RNA (siRNA) by Fire and Mello in 1998 has provided new avenues of combating resistant cancers. Silencing genes that are involved in drug resistance using RNA interference (RNAi) can allow for reversing cisplatin resistance in ovarian cancer. Successful treatment of ovarian cancer cells with multidrug resistance (MDR) gene-silencing siRNAs and cisplatin requires the development of novel vehicles that can specifically and effectively deliver cisplatin to cell nuclei and siRNAs to cell cytoplasms, respectively. We report here the first use of nanoscale metal-organic frameworks (NMOFs) for the co-delivery of cisplatin and pooled siRNAs to overcome drug re-sistance in ovarian cancer cells.

MOFs are an emerging class of self-assembled, porous materials whose properties can be readily tuned by varying the molecular building blocks. When scaled down to the nano-regime, NMOFs serve as efficient nanocarriers for the delivery of imaging contrast agents and chemotherapeutics. We surmised that NMOFs represent a unique nanocarrier platform by virtue of their high porosity and controllable surface functionalities: the large pores of NMOFs can be used to load chemotherapeutics while the metal ions on the NMOF surfaces can be used to bind siRNAs. The simultaneous and efficient delivery of cisplatin and pooled siRNAs to ovarian cancer cells can allow for enhanced anticancer efficacy by blocking multiple drug resistance pathways. In this work, cisplatin and siRNA were sequentially loaded into UiO NMOFs by covalent attachment to bridging ligands inside the NMOFs and coordinating to metal sites on the NMOF surfaces, respectively. UiO NMOFs protect siRNAs from nuclease degradation, enhance siRNA cellular uptake, and promote siRNA escape from endosomes to silence MDR genes in cisplatin-resistant ovarian cancer cells. As a result, co-delivery of cisplatin and siRNAs with NMOFs led to an order of magnitude enhancement in chemotherapeutic efficacy in vitro, as indicated by cell viability assay, DNA laddering, and Annexin V staining.