Undergraduate Thesis Prospectus

Reaction-Diffusion Model of the Centromere-Signaling Network

(technical research project in Biomedical Engineering)

Human Gene Editing: With Great Power Comes Great Responsibility

(STS research project)

by

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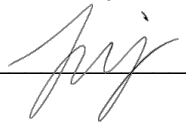
signed:  date: 10/30/19

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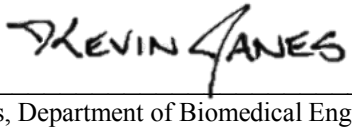
approved:  date: 11/07/19
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General Research Problem

How can genetic science improve disease treatments?

According to a 1959 estimate, 7% of people carry a genetic disease (Stevenson, 1959), but when diseases with tangentially genetic causes and late onset are included, the estimate can be as high as 50% (Baird et al., 1988). Although the patients' quality of life varies depending on their genetic disease (Cohen & Biesecker, 2010), depression and reduced functional capacity are associated with life-threatening genetic diseases such as Huntington's disease (Ho et al., 2009).

Reaction-Diffusion Model of the Centromere-Signaling Network

How can a reaction-diffusion model of the centromere-signaling network be constructed?

Significance and State-of-the-Art

Approximately 5% of the US population lives with cancer (Howlader et al., 2018). According to the National Cancer Institute (NCI), 1.7 million new people are diagnosed with cancer and half a million people die from cancer every year (NCI, 2018). Cancer patients are affected by fear for the future, strains on family life and daily activity, and debilitating pain (Schipper et al., 1984). The average annual healthcare expenditure is $150 billion (NCI, 2018), and there are innumerable research laboratories scattered across the world dedicated to researching treatments and the underlying mechanisms of this deeply impactful disease. In cancer biology, aneuploidy is widely viewed to be a hallmark of many cancers, but since chromosomal abnormalities are irregular and highly variable, it is difficult to quantify what types and percentages of cancers are related to aneuploidy

(Gordon et al., 2012; Thompson et al., 2010; Thompson & Compton, 2011). Many computational modeling and systems biology approaches exist for cancer, including modeling of tumor cell proliferation, metastasis, and angiogenesis (Araujo et al., 2014; Kansal et al., 2000; Stefanini et al., 2012); predictive patient-specific modeling based on relevant genetic, transcriptomic, and physiological data (Lee et al., 2019); models to predict patient-specific response to certain therapies (Powathil et al., 2013; Vainas et al., 2012); and reaction-diffusion models of certain signaling pathways (Bianconi et al., 2012; Pappalardo et al., 2016).

Approach

The centromere-signaling network (CSN) is a signaling cascade that occurs during metaphase of mitosis (and meiosis) to regulate the spindle assembly checkpoint and also proper cohesin-mediated apposition of sister chromatids (Trivedi & Stukenberg, 2016). Overexpression of certain proteins (e.g. the chromosomal passenger complex [CPC], Mps1, Bub1, Sgo1, and Haspin) in the CSN is a characteristic of many aneuploid and cancerous cells (Kim et al., 2017; Ling et al., 2014; Mu et al., 2019; Ricke et al., 2011; Xia et al., 2015). It is hypothesized that these protein imbalances lead to mis-segregation of chromosomes and subsequent aneuploidy, which are believed to be directly related to cancerous mutations in cells. No bedside tool exists that can intake patient-specific protein concentrations, pinpoint mitotic pathology at the single-cell level, and predict future or detect current malignancies. A model like this could elucidate the causes of cancer, identify new chemotherapeutic targets, and allow exquisite control of conditions not possible *in vivo* or even *in vitro*. The specific aims of our project include the following:

1) Mine the literature for physicochemical parameters of the CSN

2) Develop a computational model of the CSN

3) Utilize the computational model to make predictions about the probability of merotelic attachment formation, aneuploidy, and tumorigenesis

We will use the computational modeling software Virtual Cell (VCell), because it is specifically designed for cellular reaction-diffusion modeling. By the end of the project, we hope to have a predictive model that intakes CSN protein concentrations for either healthy or cancerous human cells to determine the likelihood of CPC recruitment and merotelic attachment formation at the inner centromere.

Human Gene Editing: With Great Power Comes Great Responsibility

How do scientists, patients, regulatory agencies, and members of the general public compete to distinguish responsible from reckless applications of human gene editing?

Significance and State-of-the-Art

Gene editing was born in the 1970s in genetic engineering companies such as Genentech (Russo, 2003). Diverse editing approaches arose over time; perhaps the most famous approach is the CRISPR-Cas9 technique, which followed Jinek et al. in 2012. The Food and Drug Administration (FDA) attempts to regulate clinical applications of gene editing in the United States. While other countries also have medical regulatory bodies, some researchers violate regulations or cross purported ethical boundaries (Cyranoski, 2019a, 2019b).

The 1975 Asilomar Conference occurred after the introduction of recombinant DNA technology, and similarly, ethics conferences followed CRISPR-Cas9's introduction. According to the National Academies of Sciences, Engineering, and Medicine (NASEM), the First International Summit on Human Genome Editing was held in 2015; a second followed in 2018 (NASEM, 2018). Gene editing can treat a vast array of diseases, from Huntington's disease to

hearing loss and beyond (Chien, 2018; Ekman et al., 2019). Two constraints hinder gene editing: *i*) off-target gene edits can affect unintended genes and *ii*) off-target or on-target gene edits may have undesirable effects. Targeting efficiency is improving (Liang et al., 2015, p. 201; Shen et al., 2014), but comprehensive longitudinal studies to evaluate the unforeseen effects of editing all genes will take time. The most controversial technique is germline gene editing (editing the embryonic or sex cells of an organism so that all of the organism's and their progeny's cells contain the edited gene), as opposed to somatic gene editing (editing only some of the body's non-sex cells in a non-heritable manner).

Documentary Research

Scientists

Some scientists claim that we need to evaluate safety comprehensively before using gene editing in humans. The Association for Responsible Research in Genome Editing (ARRIGE) contends that "human genome edited technologies should not be permitted nor authorized until deemed safe and effective for human beings" (ARRIGE, 2018). A minority of rogue scientists has pushed forward with controversial experiments. In November 2018, Chinese researcher He Jiankui announced that he edited the embryos of twin girls by inactivating the *CCR5* gene. The girls' father was HIV-positive, and the edit aimed to give the girls resistance to HIV/AIDS. After the babies were born without obvious health defects, he stated that he was proud to have brought "children as healthy as any other babies" into the world (Jiankui, 2018).

Other scientists push the boundaries of regulation while refraining from the most controversial steps of editing human embryos: implantation into the womb and birth. Russian biologist Denis Rebrikov is one such researcher who claims that he will "not transfer an edited embryo without the permission" of the Russian Ministry of Health. However, he maintains that

as “soon as we demonstrate the safety of” gene editing, the laws “will change” to permit implantation of edited embryos (Cyranoski, 2019b).

Distinct from scientists who say that we need to halt further gene editing until safety and efficacy are proven, bioethicists question whether gene editing (especially germline) should ever be ethically permissible. Koplin et al. (2019) recommends abiding by the precautionary principle: since the risks are unknown, we should restrict gene editing preemptively until we learn more about them.

Certain bioethicists distinguish gene editing for “therapy” from for “enhancement” (Sparrow, 2019). Therapy is treatment of a disease, whereas enhancement is physical augmentation of attributes to be more “desirable” (e.g. altering height genes to permit a patient to grow taller). Enhancement editing, the creation of “designer babies,” has been very controversial (Segers et al., 2019; Sparrow, 2019). Germline enhancement gene editing may lead to genetic obsolescence. According to Sparrow (2019), those who had gene editing done in the past will have obsolete genes compared to those edited in the present.

Patients

Patients with genetic diseases agree that scientists should perform experimental gene editing therapies despite safety risks. Malakkar Vohryzek, a patient with an unknown skin condition that insertion of the *Dsup* gene might cure (Hashimoto et al., 2016), claims that if he dies “because of an experimental treatment, it will at least help science” (Begley, 2019). John Sabine is a patient with a mutation in the *HTT* gene causing his Huntington’s disease, which may also be treatable with CRISPR-Cas9 (Ekman et al., 2019). He said that patients with genetic diseases will not think “there is any moral issue” in using gene editing (Hayden, 2016).

Regulatory Agencies

Regulatory agencies are particularly important players, since they control what research and clinical uses are permissible by law. The consensus among the World Health Organization (WHO), many federal governments, and national medical regulatory agencies (e.g. the U.S. FDA) is that germline gene editing should be restricted for now (Cohen & Adashi, 2016; Cyranoski, 2019b; Dobrovidova, 2019). However, despite the position of the Russian Ministry of Health (Dobrovidova, 2019), researchers such as Denis Rebrikov continue to contest national policy (Cyranoski, 2019b). Under a recent Chinese law, researchers who implant edited germline embryos are responsible for any adverse events from the edited genes (Cyranoski, 2019a).

General Public and Biohackers

Riggan et al. (2019) examined the general public's attitudes towards gene editing. Many contended that gene editing is a "slippery slope," since the line between enhancement editing and therapeutic editing can be blurry (e.g. increasing muscularity can also decrease susceptibility to type 2 diabetes). Biohackers favor the widespread dissemination of gene editing technology out of the ivory tower of academia and into the general public. Most have no professional training in biological sciences, but they conduct do-it-yourself (DIY) biological experiments (Nature, 2010). Biohacker Josiah Zayner claims that if gene editing "is just in the hands of a few people, and nobody else is using it, then it can't have the power that it really needs to have." Zayner sparked controversy in 2017 when he injected himself with edited genes. Zayner is also the CEO of a company known as The ODIN, which sells DIY CRISPR-Cas9 kits for editing human cells among others (Zayner, 2017). The FDA has stated that DIY human gene editing kits are illegal (FDA, 2017), but The ODIN continues to sell them.

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