Sugaway: Using Synthetic Biology to Treat Diabetes
(Technical Report)

The Growing War between the Pharmaceutical Industry and Diabetic Patients
(STS Research Paper)

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Promotto Islam
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On my honor as a University Student, I have neither given nor received unauthorized aid on this assignment as defined by the Honor Guidelines for Thesis-Related Assignments

Signature: _____ Date: 5/13/2022

Promotto Islam

Approved: _____ Date: _____

Dr. Keith Kozminski, Department of Biology

Approved: _____ Date: _____

Dr. Joshua Earle, Department of Engineering and Society

General Research Problem

How can the treatment and prevention of diabetes be improved?

According to the Centers for Disease Control and Prevention (CDC), diabetes mellitus affects 34.2 million individuals in the US (CDC, 2020); by 2060, cases may exceed 60 million (Lin et al., 2018). Administration of synthetic insulin is the primary treatment for diabetes, but since the discovery of synthetic insulin in 1978, its price has risen faster than its cost of manufacture (Belluz, 2019). High prices limit access to insulin, jeopardizing the health of many diabetics. (Rajkumar, 2020).

Through synthetic biology, novel biological systems can be engineered, including genetically engineered bacteria. Recent advances in synthetic biology have harnessed the power of preexisting biological systems in order to engineer novel biological systems. Such bacteria may be used to treat phenylketonuria, a metabolic disease that prevents the digestion of phenylalanine, an important amino acid. (Durrer et al., 2017). This suggests that genetically modified bacteria may also serve as a treatment for diabetes, specifically as an insulin supplement.

Alternatives to Insulin for Treating Diabetes

How can genetically modified bacteria be used to supplement insulin use in diabetic patients?

Faculty Advisor: Dr. Keith Kozminski (Department of Biology)

Project Type: Capstone Project

Diabetes mellitus consists of a group of metabolic diseases which all share a common characteristic of inducing high blood glucose levels (Kerner & Brückel, 2014). This increase is often due to a dysregulation (type I diabetes) or dysfunction (type 2 diabetes) of native insulin

(Guthrie & Guthrie, 2004). Glucose is a carbohydrate that is naturally ingested in most diets, and to control blood glucose levels insulin is secreted to induce the conversion of glucose into a long-term storage form called glycogen. However, in diabetics this balance is disturbed and chronically elevated blood glucose levels can cause serious complications such as cardiovascular disease, nerve damage, or kidney damage. After a clinical diagnosis of diabetes, individuals must make careful lifestyle changes and continue a strict medication regimen. However, this is not always effective or doable, which contributes to the growing prevalence of diabetes in the United States. One method in countering this is through the production of cheaper and more effective medication, and the field of synthetic biology offers promising methods to finding a better treatment for diabetes.

The goal of this capstone project is to genetically modify bacteria with plasmids in order to uptake excess extracellular glucose and to safely convert it into glycogen. There are no unusual constraints for this project.

Current guidelines set by the American Diabetes Association (ADA) state that the primary treatment for diabetes should be a treatment plan of insulin (ADA, 2021). Other glucose lowering medications are also considered such as Metformin or Pramlintide depending on the patient's condition. For severe cases of type I diabetes, surgical transplantation of the pancreas is an option, but will require lifelong immunosuppressant medication and could trigger further complications. Insulin treatment can be effectively administered either by an injection or by an insulin pump, but are expensive to own and are not an option that is available to everyone (The BMJ, 2017). The aforementioned medications have also been shown to have similar benefits to insulin treatment (Mercurio, 2017), but not all medications are easily accessible due to their price and many come with unfavorable side effects. Besides medication guidelines, there are also

lifestyle changes such as getting more exercise and dietary restrictions, but these are often not enough to help individuals with severe diabetes or a genetic basis for diabetes.

The common factor in both the medical and lifestyle treatments described are their accessibility. Many medications are not easily accessible due to their costs, and lifestyle changes may not be feasible for individuals who live in food deserts and lack access to affordable, healthy, food. Therefore, there is a need for an effective and cheap treatment to diabetes which can be readily accessible by all diabetic patients.

To investigate the feasibility of an insulin supplement, bacteria will have to be genetically modified using plasmids. The initial plasmid will be a glycogen synthesis plasmid that will contain instructions to increase the metabolic conversion of glucose to glycogen. After transformation, expression of proteins will be verified by a Western Blot and protein functionality will be tested with a glycogen concentration assay. Once verified that the expressed proteins are working as expected, the sustainability and toxicity of transformed bacteria will be measured. This will ensure the bacteria can safely convert glucose to glycogen without causing harm to other surrounding microorganisms.

The proposed deliverable from this project is a genetically modified bacteria which mimics the functionality of synthetic insulin. This represents the basis for a probiotic which would be taken as an oral drug, which would be ingested and supplement the typical synthetic insulin taken by diabetics. This probiotic is cheaper and easier to administer compared to current treatments, and will reduce the dependency on synthetic insulin.

The Growing War between Pharmaceutical Industry and Diabetic Patients

In the United States, why do many diabetics distrust or resent pharmaceutical companies?

High prices and consequent access inequities have been matters of contention between diabetic patients and the pharmaceutical companies. From 2012 to 2018, synthetic insulin prices rose an average of 14 percent a year. For one diabetic, the annual expenses for insulin are now close to $6,000. About a quarter of diabetics cannot cover the costs of their insulin; more than half rely on public assistance for their medical expenses. If recent trends continue, the annual per-patient cost for insulin may double to $12,000 by 2024 (Hayes & Farmer, 2020). High prices have forced some diabetics to ration or used expired insulin, causing some deaths in the past 5 years (RCA, 2021). Advocates have cited these deaths in their fight against the pharmaceutical giants that produce and price synthetic insulin.

The insulin production pipeline is complex, incurring costs at each stage (fig. 1). The

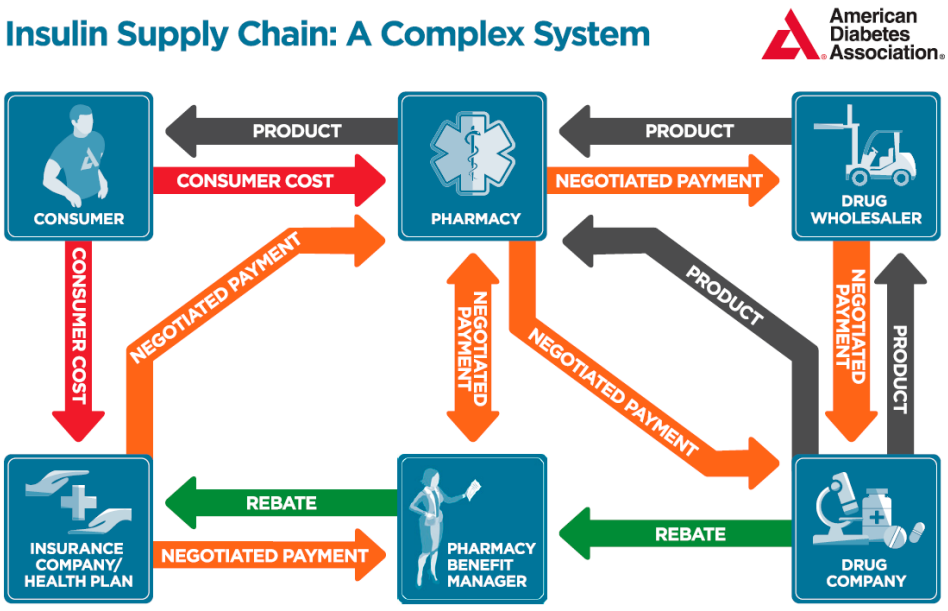

Figure 1. Schematic of Insulin Supply Chain (Cefalu et al., 2018)

complexities obscure cash flows, while negotiated payments and rebates elevate prices.

This complex system is unique to the United States, which has the most privatized healthcare sector among developed countries. No federal agency centrally establishes or regulates drug prices. Instead, multiple pharmaceutical companies and insurers negotiate prices. The system affords companies opportunities for price gouging, which has inflated prices for EpiPens, opioids, and insulin (Belluz, 2019). Pharmaceutical companies justify high prices by citing the costs of drug development and clinical trials, as stated specifically by Johnson and Johnson in their *2017 Janssen U.S. Transparency Report* (JP, 2018). Yet the costs of newer insulin variants may not outweigh the benefits that pharmaceutical companies advertise (Luo et al., 2019).

In this struggle, the advocacies that represent diabetic compete with the trade associations that represent pharmaceutical companies Insurers are also engaged. The Right Care Alliance (RCA, n.d.) claims it strives "to make health care institutions accountable to their communities" by prioritizing patient care. One of their campaigns is to increase the affordability of insulin treatment, and they have organized multiple protests since 2018 against pharmaceutical company price gouging. The Pharmaceutical Research and Manufacturers of America (PhRMA, n.d.) is a trade association representing pharmaceutical companies. It claims to promote "effective advocacy for public policies that encourage discovery of important, new medicines." In 2020, PhRMA sued the Minnesota Board of Pharmacy, claiming that a state law the board used to regulate insulin prices was unconstitutional (MMA, 2021). Some researchers who are committed to finding a cheaper way to produce insulin call themselves Biohackers. The Open Insulin Project is a group of volunteer researchers developing "an open-source (freely available) model for insulin production" (OIP, n.d.). Applying synthetic biology, OPI is genetically modifying yeast cells to produce human insulin, which they hope can be harvested for us by diabetics.

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