



CALIFORNIA STATE SCIENCE FAIR 2016 PROJECT SUMMARY

Name(s) Kylie M. Huch	Project Number 36475
Project Title Quantifying the Extent to Which Traits of Extinct Species Can be Revived via the Bioengineering Method of De-extinction	
Objectives/Goals The objective of this experiment is to quantify the capacity the bioengineering method of de-extinction holds to restore the expressed traits of an extinct species. It aims to quantitatively assess the degree to which replicating the structure of a gene's encoded protein restores its expressed trait. Abstract Methods/Materials I used the bioinformatics platform, Geneious, to design two variants of the band-tailed pigeon AMY2A gene modified to carry mutations from the passenger pigeon AMY2A gene structurally significant to the encoded protein. The first variant contains all structurally significant mutations, representing the ideal application of the bioengineering method, the second variant contains only the two most structurally significant mutations, representing a minimal application of the bioengineering method. I then designed CRISPR systems to create a third gene variant by editing the band-tailed AMY2A gene invitro to recreate experimental variant two. Also using Geneious, I created delivery vectors carrying each gene variant and one carrying my CRISPR systems for expression in E.coli HB101. I will isolate the expressed proteins (variants of the a-amylase starch digestion enzyme) via column chromatography and assess their functions via a standard amylase activity assay and an acorn starch digestion. Results Between the 1608 bp band-tailed and passenger pigeon AMY2A genes there are 50 nucleotide differences (3.1%), 30 of which result in a difference in one of the 535 encoded amino acids (5.6%). Out of the 30 aa differences, 27 affect the structure of the encoded protein to some degree and two drastically alter it. I'm in the process of acquiring the funding to synthesize my delivery plasmids and thus have yet to express my gene variants. Conclusions/Discussion Experimental variant one represents the maximum capacity the bioengineering method holds to revive an extinct species. If its encoded a-amylase is identical to passenger pigeon a-amylase in function, it would indicate the bioengineering method holds the capacity to completely restore expressed traits of an extinct species. Experimental variant two represents a minimalist application of the bioengineering method and the extent to which the function of its encoded a-amylase replicates that of the passenger pigeon a-amylase will provide a measure of the lower bound of the capacity this method holds to restore expressed traits of an extinct species.	
Summary Statement I devised a way to quantify the capacity the bioengineering method of de-extinction holds to restore the expressed traits of an extinct species.	
Help Received Ben Novack, based out of the USCS Paleogenomics Lab, provided me with the passenger pigeon and band-tailed pigeon AMY2A gene sequences and explained the processes of de-extinction. My biotechnology teacher taught me to use the equipment required for transformations and protein isolation.	