Muscle Glue
Calvin College Senior Design 2015-16 Team 20

Project Overview
Create an adhesive based on marine mussel bioadhesives to outperform traditional petroleum-based adhesives in wet environments.

The adhesive is produced using recombinant DNA in bacteria. A strand of DNA that encodes for a protein is spliced into E. coli DNA. The E. coli is allowed to reproduce, then the cells are lysed and the adhesive protein is separated, purified, and activated.

Mussel adhesive proteins work in water-rich environments, adhere to both organic and inorganic surfaces, and have high biocompatibility. This makes them ideal for use as surgical glues.

Current methods to produce mussel based adhesives rely on extraction of the protein straight from the organism itself. These inefficient techniques use 10,000 mussels to produce a single gram of protein.

Project Goals
An effective medial glue must be
- Biocompatible - it must not cause harm to the patients
- Functional in aqueous environments - must work in wet environments since the human body is 70% water.
- Affordable - can not be so expensive that nobody can use it.
- High Strength - must reliably prevent wound reopening.
- Safe and reproducible - The manufacturing process must not cause long term damage to the environment or workers.

Design Process
- Determine manufacturing method
  Extraction directly from mussels, and recombinant techniques in yeast, E. coli, and Chinese hamster ovary cells were considered. E. coli growth was the cheapest and most environmentally friendly method.
- Determine specific protein desired
  Many different proteins are involved in the adhesion process in mussels. MFPs (mussel foot proteins) 3 and 5 are the stickiest but hardest to separate, while MFP 1 is easy to separate but not sticky. The final protein used is mgfp-151RGD which is a modification of protein 5 by the addition of portions of the structure of protein 1 on either end. This results in high adhesion and easy separation.
- Develop process simulation base case
  A base case was developed using SuperPro designer.
- Economically optimize process
  Process variables such as reactor size were varied to make the process as economically efficient as possible while still producing the required amount of protein.

Economics
In order to meet the goal of 5% of the U.S. market, 370 kg of adhesive would need to be produced per year. Economic estimations for a process of this scale are
- Revenue from sales: ~$30 million / year
- Estimated operating costs: ~$13 million / year
- Estimated capital costs: < $60 million
- FDA approval / clinical studies: unknown

This leaves $17 million per year of profit, which will more than pay back the capital cost over the 15 year lifespan of the plant.

Activation
Final product is packaged and shipped to consumers

Centrifuge out water and other remaining products of tyrosinase reaction

Remove other products of reaction via diafiltration

Hydroxylate with tyrosinase to activate adhesive properties of protein

Purification
Centrifuge to remove non-protein cell remains and lysis buffer.

Extract with acetic acid, what is not soluble in acetic acid is left behind

Use affinity column to separate out tagged protein.

Separate protein from affinity column elution buffer with adsorption membrane

Freeze dry to remove excess water

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