

Reducing Coconut Husk Pollution Using Cellulose-Degrading Bacteria to Make Self-Illuminating Ceiling Tiles

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The study of Engineering is always exciting! Did you ever know that even Biology can be engineered? Well, it can, and is done presently. The fusion of engineering and biology is a breakthrough as seen by many scientists and is termed Synthetic Biology. Synthetic Biology, according to the National Human Genome Research Institute, is a field of science that involves redesigning organisms for useful purposes by engineering them to have new abilities [1] Amazing, right? Organisms can do the extraordinary based on the instructions (DNA or gene) you place inside it. Because of these mind-blowing possibilities, companies worldwide are harnessing the power of nature through engineering to solve problems in medicine, manufacturing, agriculture, and many other aspects of life. Conservation is also another critical area in which synthetic biology has been much appreciated, termed as Bioremediation where microorganisms break down and consume pollutants. Consider this scenario; applying synthetic biology principles in biodegrading coconut husks to reduce its arising pollution and further incorporating this achievement into building self-illuminating ceilings. Other applications include self-illuminating recreational facilities, such as pathways in communities using biological parts from specified self-illuminating bacteria. It is also imperative to note that, Synthetic Biological Engineers care about you and the environment; thus, safety is of utmost importance to them and they ensure that their designs cause no harm to neither you nor our world.

Unfortunately, pollution has become a major global issue. Governments, individuals, and organizations have taken initiatives to help reduce pollution since various types of pollution (water pollution, land pollution, air pollution etc.) have revealed numerous life-threatening manifestations. Some of these indicators include climate change and the spread of air and

water-borne diseases such as , cholera and typhoid fever leading to the loss of lives. Pollution has led to environmental degradation and has been observed to have begun since the industrial revolution [2]. Land pollution is among the first three types of pollution negatively affecting the environment [3]. The husks of coconut in Ghana is a major contributing factor to land pollution in the country [4]. Coconut is a drupe fruit that primarily grows in tropical regions (e.g., in Ghana) and is locally consumed while some are exported. Currently, annual production in Ghana is 224 million coconut fruits, and smallholder farmers produce 179 million of the total production as of 2019, according to the Coconut Producers and Exporters Association [5]. Due to very high temperatures in the country and the medicinal benefits of the nutrients for consuming coconut, there has been active patronization of coconut; thus, lots of waste is being produced daily. The husks of these coconuts have been seen to be disposed in gutters, deserted lands, in water bodies, and dumping sites and have raised significant concerns from locals and sanitation organizations since these pollutants have become catalysts for natural disasters like floods. Even though many individuals and some inclined conservative organizations have tried to recycle these husks and the fibres into other useful products like coconut fibre mats, coconut fibreboard, coconut fibre seedling pots, coconut fibre dish scrub pad, etc. there is still massive pollution of coconut husks. [6]



Fig. 1 Image of coconut juice, mesocarp, epicarp and endocarp



Fig. 2 Coconut husk disposed on land sites waste indiscriminately

So, how do we give these bacteria a specific role to play? *Escherichia coli* (*E. coli*) is a commonly used bacteria for bioengineering experimental purposes. Most strains of *E. coli* are harmless and able to metabolize glucose in both aerobic and anaerobic circumstances. The engineered bacteria (chassis) with antibiotic resistance, in this case *E. coli*, can be modified by inserting a sequence of DNA into it, facilitating the production of an enzyme to perform a task, for instance, breaking down cellulose found in a coconut husk. Bioremediation has become very useful in our society because organic wastes are biologically degraded under controlled conditions to an innocuous state or below the concentration limits established by regulatory authority [7]. The process also reduces the quantity of pollution and emissions produced compared to the traditional methods of getting rid of biological pollutants such as coconut husk. Traditional methods such as burning the coconut husk release soot, an impure carbon particle that pollutes the environment and harmful gases that harm life directly or contribute to the greenhouse effect. Hence, fostering global warming.

Even though Bioremediation involves using microorganisms to break down and consume pollutants, modifications through the power of synthetic biology can be made to these organisms to possess other features that can be useful to the environment. Coconut contains cellulose nanofibrils as a part of its chemical composition. Hence, the genetic makeup of a cellulose digesting bacteria, for example, the *Fibrobacter succinogenes* S85 found in the rumen of herbivores [8], can be modified to digest the cellulose nanofibrils in coconut. Interestingly, more than one modification can be made to an engineered bacterium. For instance, the genetic makeup of the coconut husk degrading bacteria can also be further modified such that it can produce luminescence during the night for aesthetics purposes or to enhance visibility. ‘Does this call for electricity?’ you may ask; surprisingly, not at all. It is simple. For example, lux operon (BBa_K1725352) obtained from the *Vibrio fischeri*, a rod-shaped bacterium found globally in marine environments has bioluminescence properties and

could be used for such purpose and has proved itself to be accurate in its functionalities as seen from the image below. [9].



Fig.3 The Lux light generator expressed in a medium

Therefore, putting genetic parts such as promoters, Ribosome Binding Sites (RBS), Coding Sequences (CDS), Terminators, and even repressors lead to the proper functioning of an engineered bacteria. All these genetic parts come in different forms and have specific roles they play when they come together. Hence, these parts are explicitly chosen according to their desired function required in the chassis (host bacteria). There are also different ways of putting the desired gene into a chassis. One of these ways is called the BioBrick Assembly using the Restriction Enzyme Cloning or Digest method, where the gene is being placed in a plasmid by the use of restriction enzymes (e.g., PstI, EcoRI, etc.). Then, this new plasmid (recombinant plasmid) is placed in the bacteria. Here is an analogy of the cloning process; consider making a BBQ beef brisket sandwich which would be your engineered bacteria. The sandwich bread is the bacteria to be engineered and the sandwich filling consisting of beef, vegetables, spices and cheese, which collectively determine the taste of sandwich are the cloned plasmids which determine the functionality of the engineered bacteria

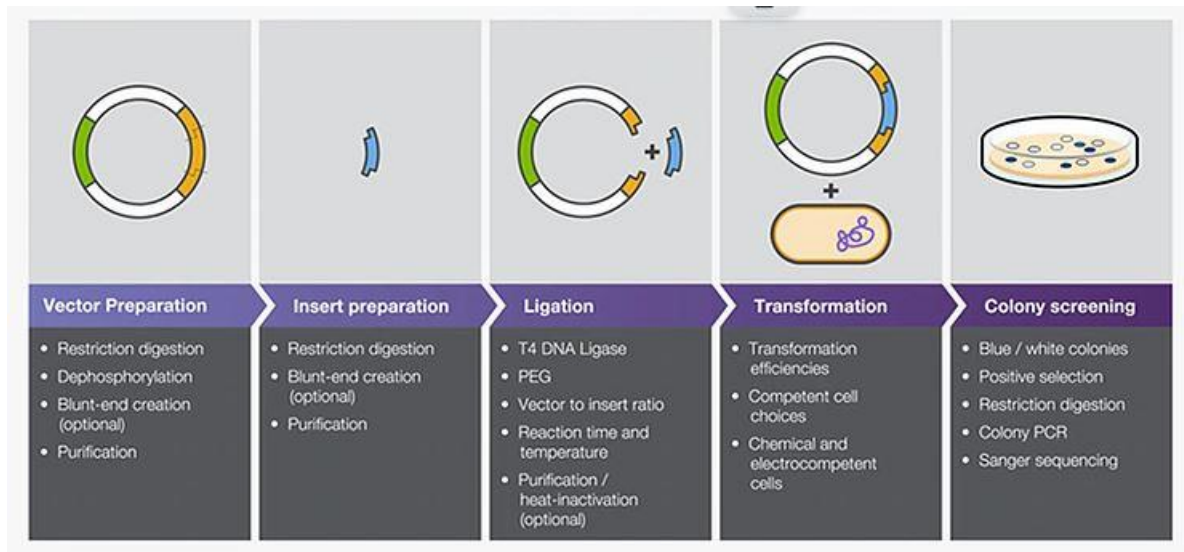
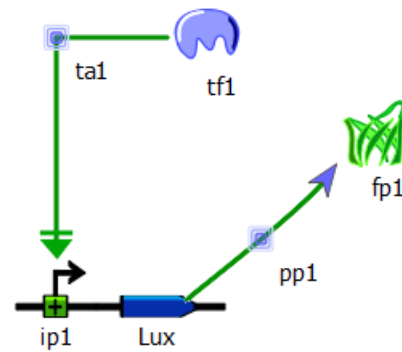
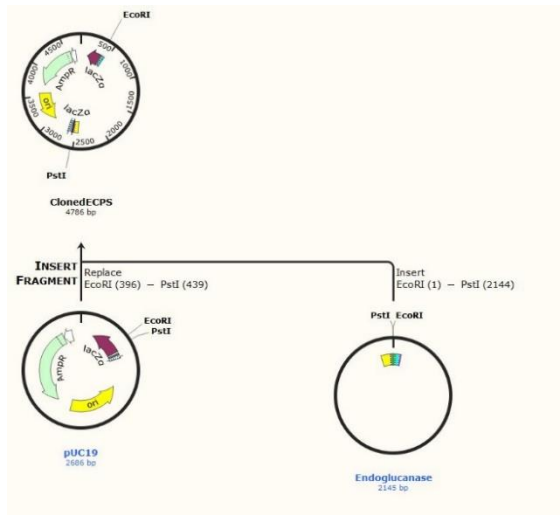


Fig.4 Summary of the Restriction Cloning process

Referring to the above example of an engineered bacteria digesting cellulose and being self-illuminating as well, this is how our bacteria was engineered. Using the Restriction Enzyme Cloning method, we came up with two recombinant plasmids (one for cellulose digestion and one for the self-illumination). From our obtained results, both DNAs were successfully transferred into the plasmid, which will further be placed in *E.coli* bacteria cells to perform both functionalities at the same time. These results were developed and simulated using some Synthetic Biology Software – used in designing our required plasmids – SnapGene and TinkerCell. This approach is an advantage of Synthetic Biology because designing plasmids and engineering bacteria is not only done in a Biology Lab but can be first simulated virtually and later experimented physically in the lab.

Fig. 6 Genetic circuitry of the LUX in Tinkercell showing the expression of the fluorescent protein (fp1)

Fig. 5 Image shows a successful cloning of the puc19 plasmid with the endoglucanase (cellulose) gene to form a recombinant plasmid



Indeed, synthetic biology is a field that has brought about vast improvements and groundbreaking results. Even though this field of science has not been explored to its full potential, it is inspiring what the future holds.

It is always a great day for science!

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