# Research Proposal

# On

# A STEP TOWARDS PLASTIC FREE WORLD BY PRODUCING MASS BIO-DEGRADABLE FUNGI THROUGH REPLICATING HYDROLASE ENZYME.

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## Research Question:

Is it possible to replicate the hydrolase enzyme in a fungus which does not consist of it, which will help in degrading plastic following bioremediation?

## Background:

In the present world, one of the most harmful substance for the environment and a threat to our ecosystem is plastic. Plastics are a wide range of synthetic or semi-synthetic that use polymers as a main ingredient. However, earth heals itself and so it is found in a research work done by The Yale University in 2011 that some fungi are capable of degrading plastic. [1].Fungus is a member of the eukaryotic organisms including yeasts, molds. When we speak of fungi, we have to speak of fungal bioremediation [2]. which is an eco-friendly approach of fungi to mitigate environmental pollutants. Some endophytic fungi. [3]. such as *Pestalotiopsis microspore, Aspergillus niger* with the help of their mycelia which reach their chosen and releasing required enzymes.[4]. In this case with the help of hydrolase enzyme such as proteases, laccase and esterase, fungi are said to degrade plastic.[5]. The goal is to replicate such hydrolase enzyme in other fungi which will eventually help in achieving the goal of degrading plastic.

## Literature Review:

There have been no such direct research to replicate the key enzyme of plastic degradable fungi in other fungi. However, there have been much research on the key factors of plastic degradation: 1.How Fungi degrade plastic? 2. Which Fungi are more appropriate in plastic degradation? 3. Where to cultivate such Fungi? 4. How to replicate the genome consisting of hydrolase enzyme? Our key goal is to replicate the very gene in other fungi with the help of gene sequencing and plasmid transformation. This approach is not seen previously as in most other researches, the researchers were working on the genome of the plastic degrading fungi only. They did not try or thought of replicating it on other fungi. Although, it is a new approach it could be essential to open a new horizon in eliminating plastic from our environment. Previous researches have shown how *Pestalotiopsis microspore* can degrade the synthetic PUR. But our goal is to try doing that with a very different fungus. Previously, the genome was also identified and the primers were also identified. [6]. This is mostly about previous researches which spiked the interest for this one.

## Research Methodologies:

The research is divided into the following steps:

1) Identifying the perfect Fungus for replication: Most appropriate to select in this case should be a healthy sample of *Pestalotiopsis* which was previously able to degrade plastic in a lab test.
2) Extraction of Hydrolase enzyme: The fungal enzyme can be extracted using ureal solutions in a controlled environment of a lab.
3) Identifying the genome: In this case the specified genes are Glycoside hydrolases, Polysaccharide lyases and carbohydrate esterases.

4) Gene cloning: Using gene extraction, PCR amplification and cloning vector. We will replicate the gene.
5) Transformation into host cells: The extracted gene will be transformed into a suitable host and we need to find a proper recombinant plasmid.
6) Protein expression: we need to induce protein expression under optimized conditions and further follow cell lysis and purification techniques.
7) Sustainability of the fungus with the new gene: We need to observe the fungus
8) which will carry the new gene for any further complications.

These are all the steps required for the process.

## Project Practicalities :

The complete research needs to be conducted in the controlled environment of a research facility. The total budget should be stretchable likewise in a genome extraction research. The time for the research is within a year. The whole research will be conducted under all set of laws, rules and codes provided by The National Research Institute and the sovereign country.

## Roadblocks and potential limitations:

The potential complication might be the identification of a proper sample. Afterwards, the budget and the permissions in this regard can be a complication as well. Moreover, without proper technology for gene transfer and replication, this research cannot be conducted. Again, there can be risk of biohazards as it might produce a fungus with possibilities of spreading fungal infections. There can be an ethical side to consider about when we are working with gene replication or altercation. However, these risks can be mitigated if the experiments are done with caution.

## Post-program plan:

The post program plan if the results are in favor is to publish the paper in a reputed journal like “Youth Research Journal” in order to spread mass awareness regarding the prospect of degrading plastic. Afterwards, we will mass replicate this research so that industries can use them for reducing plastic emission and control pollutants. Eventually, it will save the planet.

However, if the results are not in favor, we will find other methods to enhance the progress of fungi in degrading plastics. We will continue further researches until we find our desired results. We will continue our works in order to build a safer world without pollutants.

## LITERATURES CITED AS REFERENCES:


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