AIP Conference Proceedings

RESEARCH ARTICLE | FEBRUARY 20 2024

A short commentary: Biological pre-treatment and its enhancement-is a primitive concept? FREE

Sonam Kumari; Shweta Shah; Vivek Shit; Joginder Singh; Manoj Kumar

Check for updates

AIP Conf. Proc. 2986, 030159 (2024)
https://doi.org/10.1063/5.0194871

View Online
Export Citation
CrossMark

24 February 2024 17:48:20

AIP Advances
Why Publish With Us?
25 DAYS average time to 1st decision
740+ DOWNLOADS average per article
INCLUSIVE scope
Learn More
AIP Publishing

AIP Publishing

A Short Commentary: Biological Pre-Treatment And Its Enhancement-Is A Primitive Concept?

Sonam Kumari[1, a)], Shweta Shah[1, b)], Vivek Shit[1, c)] Joginder Singh[2, d)] and Manoj Kumar[1, e)]

[1]*Department of Life Sciences, School of Natural Sciences, Central University of Jharkhand, Ranchi – 835205, Jharkhand, India.*
[2]*Department of Biotechnology, School of Bioengineering and Biosciences, Lovely Professional University, Phagwara- 144411 Punjab, India.*

[a)] singh.sonam14@gmail.com
[b)] shweta.shah.shweta@gmail.com
[c)] vivekshit67@gmail.com
[d)] joginder.15005@lpu.co.in
[e)] Corresponding author: manoj@cuj.ac.in

24 February 2024 17:48:20

Abstract. This review provides an overview of some of the issues surrounding the existing implication of renewable energy technologies (RETs) and background rural knowledge to unfold the modern energy harvesting system. Owing the alarming call by nature about environmental pollution and continuous hike in petroleum prices, a better fuel option is still a mission worldwide.

Keywords: RETs, lignocellulosic biomass, saccharification, consortia

INTRODUCTION

Asian developing countries are being jumbled of such renewable energy source where 'ecological stress' is a pre valent term [1]. Since last five decades, second-generation biofuel production systems are being practiced for the conversion of non-comestible parts of food crops like straw, seed husk and stalks, bagasse etc, waste products from food, forest, fruit and wood industry, municipal wastes etc. Numerous research papers have been cited regarding those wastages as rich source of lignocellulosic biomass i.e., lignin, cellulose, hemicellulose and pectin.

Recycling Process

At the same time, recycling process is still poorly managed particularly in developing countries. Disseminated knowledge on such renewable biomasses are confined in research, which could be a case study at rural level[2–4]. Researchers have already fabricated the components of plant secondary cell wall (lignin, cellulose and hemi-cellulose) that are tightly interlinked through various bonds (covalent, non-covalent and chemical bonds) to provide rigidity and defence against enzymatic and pathogenic degradation[5]. Various mechanical or pre-treatment approaches as discussed in fig 1 are either obsolete or comparable with scientific output of pre-existing traditional approaches.

3rd International Conference on Functional Materials, Manufacturing, and Performances
AIP Conf. Proc. 2986, 030159-1–030159-6; https://doi.org/10.1063/5.0194871
Published by AIP Publishing. 978-0-7354-4840-7/$30.00

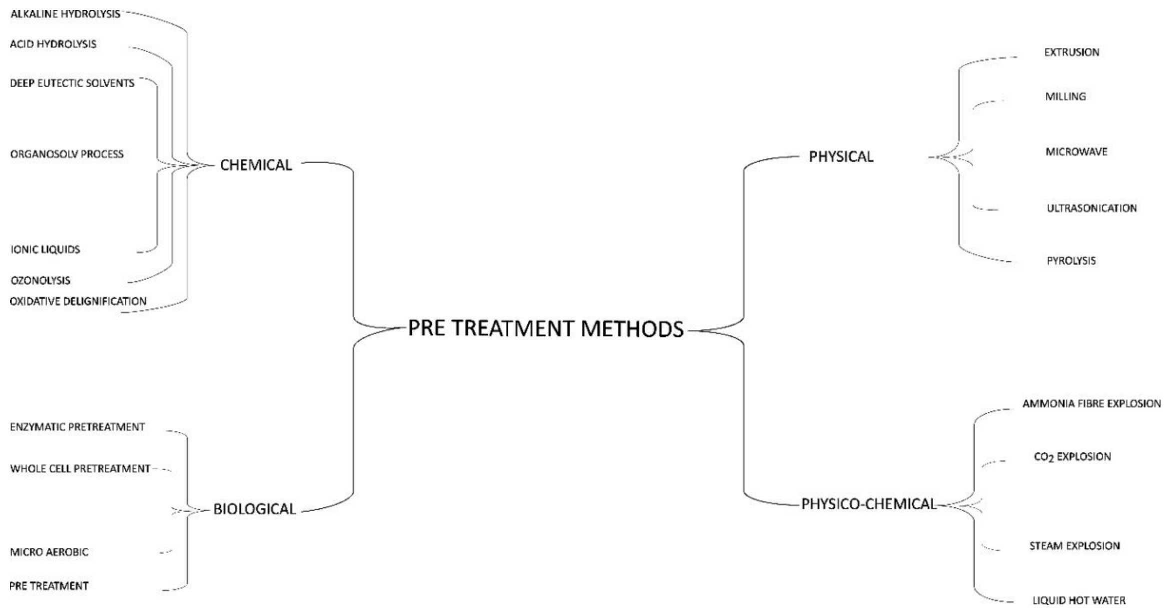


FIGURE 1: Lignocellulosic Biomass Pretreatment Methods

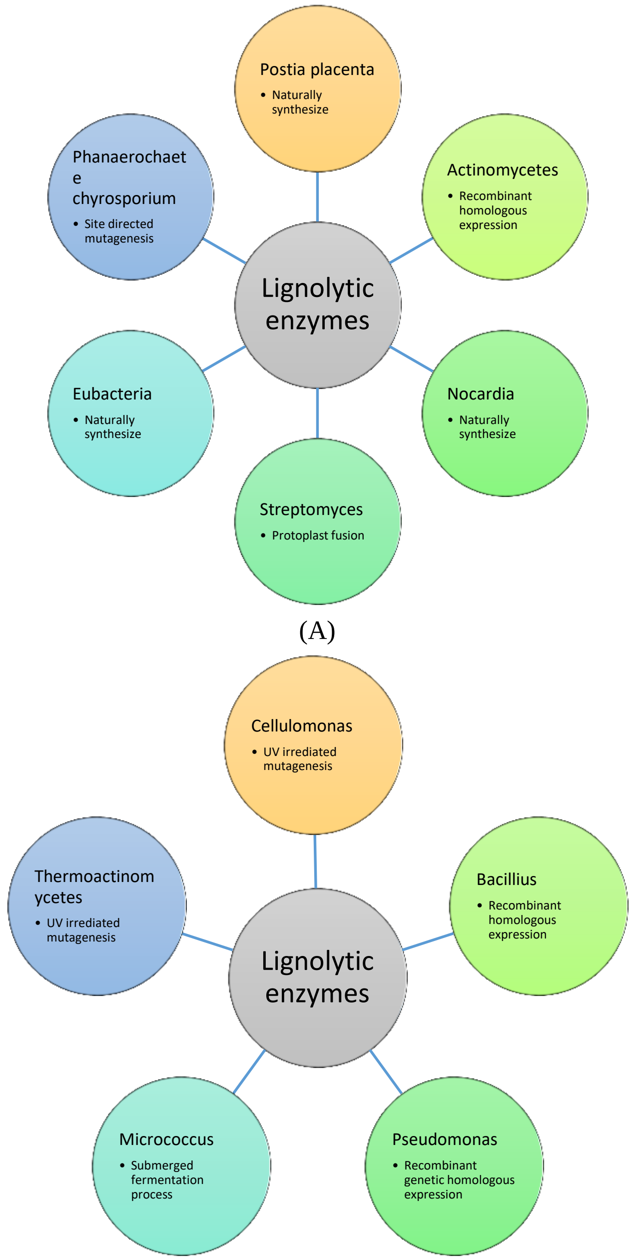
The fact that conventional fuels cannot bring into being a range of contemporary energy requisite system, for instance mechanical power and electricity confines their capacity to advance other aspects of life, as well as education and employment. As a result, they need extensive period and attempt to collect, and as local reserve stocks are being reduced, they increasingly have to be sourced from further afield which is mesmerized between nature and society. This considerably reduces the time available for prolific activities and further enhance the renewability of natural wastages. If managed unsuccessfully, such resources can also degrade the environment and generate jumble effects in other sectors. Given the cultural practices in many rural areas, these impacts are often most felt by women and children[6].

Microbial Technology

To harness most of these naturally available resources, saccharification is obligatory. Biological pre-treatment strategies, however, outcompete other pre-treatments due to the application of milder conditions, and lower by-product formation and corrosiveness. The variety of applied techniques comprises micro-aerobic treatments, ensiling or composting, separation of digestion stages, and pre-treatments using various fungi. Fungal pre-treatments have achieved particular success using various white, brown, and soft rot fungi, or a combination of these. Digestive enzymes of white rot fungi from the genera *Ceripoioposis*, *Phanerochaete*, *Fusarium*, *Trametes*, *Polyporus*, and *Pleurotus* target cellulose as well as lignin, allowing the use of recalcitrant, second-generation substrates for biogas production. Several strains of *Trichoderma reesei*are high cellulase producers, nevertheless, after genome sequencing, 10 cellulase encoding sequences were reported, but, only 4 were produced by the fungi in satisfactory amounts. Also, they had poor repertoire of genes for hemicellulases and the industrial strains have reached enzyme titers upto 100g/L[7,8]. Also,this digestion can be achieved by application of various microbial enzymes for biomass degradation[9–11]. An enzyme family known as CAZymes family (Carbohydrate Active enZymes family) act on these polymers and reduce them by acting as enzyme cocktail[12]. Breakdown of lignin is obligatory for the release of interlinked cellulose and hemicellulose to facilitate further enzymatic action. Lignin peroxidases, cellobiose dehydrogenases and laccases are few enzymes that are known for lignin modification[13–15]. Some anaerobic fungi like white rot fungi (*Phanaerochaete chyrosporium*) and brown rot fungi (*Postia placenta*) can digest lignocellulose naturally by secreting ligninases, lignin peroxidases and glycosyl hydrolases “Figure 2”[16,17]. White rot fungi are capable of degrading all three components of lignocellulose and brown rot fungi mostly affects cellulose and hemicellulose. They are the major wood decomposers in forests ecosystem [18]. As mentioned by [19] about 14,000 different species of fungi were discovered by the year 1976 with the capability of digesting the cellulosic

24 February 2024 17:48:20

components. The cellulosic component of cell wall can be digested by different cellulolytic enzymes. Diverse microbes like fungi and bacteria are capable of producing these enzymes [20]. Endoglucanases and Exoglucanases belongs to the class of cellulases enzyme produced by microbes that are capable of hydrolysing and degrading the cellulosic biomass [21].Endo-hemicellulases, exo-hemicellulases and accessory or debranching enzymes are type of hemicellulases that cleaves the polysaccharide to provide monomer sugars from hemicellulose[22].Different bacteria like *Thermoactinomycetes*, *Cellulomonas*, *Bacillus*, *Pseudomonas* and *Micrococcus* species are known to produces cellulase enzymes that digests the cellulosic component. *Streptomyces*, *Actinomycetes*, *Eubacteria* and *Nocardia* are few bacterial species known for lignin degradation “Fig. 2” [18,23].



(B)

FIGURE 2: Illustrates microbes producing lignocellulolytic enzymes either utilizing naturally available, bioprocessed or through rDNA technology implemented enzymes. In Fig “A” two fungal species has shown to produce lignin degrading enzymes in orange and four bacterial spp. in blue dot. Fig “B” shows all bacterial spp. (blue dot) producing cellulolytic enzymes for biomass degradation.

24 February 2024 17:48:20

Microbial Domain Through rDNA Technology

Very few bacteria are known for lignocellulosic digestion but are preferred over fungi since they have faster growth rate, wider availability, better adaptability to extreme environment and show heterologous expression of prokaryotic genes in an efficient way[24]. Due to the enzymatic activity, cellulose is converted into glucose which is then utilised by fungus for fermentation. [25] Reported about an anaerobic, spore forming thermophilic bacteria *Clostridium thermocellum* which was discovered in 1983 that produces both cellulases and hemi-cellulases as cell-bound enzyme and multi-protein complex combinedly called as cellulosomes. The efficiency of this method is less, and the process time is high but genetic modification of microorganisms can enhance the yield. Thus, the biological pre-treatment using microbes provides mild environment for depletion of lignocellulosic biomass that results in production of no harmful chemicals and the product monomers can be directly used for fermentation for biofuel production.[26] Suggested that the fermentation process for biofuel production could work out in three distinct strategies:

Separate Hydrolysis and Fermentation (SHF)

After hydrolysis of biomass, fermentation takes place separately, facilitating maintenance of different temperatures and optimal conditions required for each step. But it often witnesses low enzymatic efficiency due to inhibitory effect of accumulated end products.

Simultaneous Saccharification and Fermentation (SSF)

Since, both processes are carried out in same reactor, cost of production is reduced and also inhibitory effect of end products and contamination is reduced. It is the most used strategy for bioethanol production. However, in the presence of microbes like *Saccharomyces cerevisiae* which use hexoses efficiently, the pentose fermentation is laid back leading to suboptimal biofuel production.

Consolidated Bioprocessing (CBP)

In this, both the processes occur in the same bioreactor, however, there is no need for introduction of enzymes into the vessels unlike the previous ways, as they are synthesized there itself. This requires the employment of efficient microbe which can simultaneously produce a complete cellulase system and high concentrations of biofuels. Microbial consortia comprising of *Clostridium thermocellum, Neurospora crassa, Fusarium oxysporum, S. cerevisiae,* and *Paecilomyces sp.* have been used.

The fermentation end product is a good alternative over fossil fuels as it is cost-efficient and environment friendly[18]. Therefore, biological pre-treatment strategies offer great potential to improve the digestibility of different biogas substrates; however, detailed investigations of the mode of action, application of different substrates, full-scale implementation, and possible by-product formation are still needed.

Methods to Enhance Biological Degradation

Microbial Consortia

To enhance the efficiency of biological degradation process, microbial consortia can be used “Figure 3”. This concept is based on interaction of microorganisms in natural environment where they synergistically help each other to degrade macromolecular substrates. A consortium reduces the duration of pre-treatment process and also enhances conversion of substrate into product when compared to action of single bacterial strain. A bacterial-bacterial, fungal-fungal or bacterial-fungal consortium can be prepared for biomass degradation. The fungal enzymes are responsible for degrading the lignin component and conversion of lignocellulose into soluble forms on which bacterial enzymes act and converts them into monomers [19,27].

24 February 2024 17:48:20

Genetic Modification

Microbes can also be genetically modified "Fig. 3" for higher yield. For this purpose, different approaches such as heterologous gene expression in competent host, insertion, deletion and silencing of genes can be considered.Various genetic recombination techniques like CRISPR-cas 9 can be used to introduce a particular gene of interest encoding for an efficient lingo-cellulolytic enzyme into model organisms like *S. cerevisiae* and *E. coli* [26,28,29].

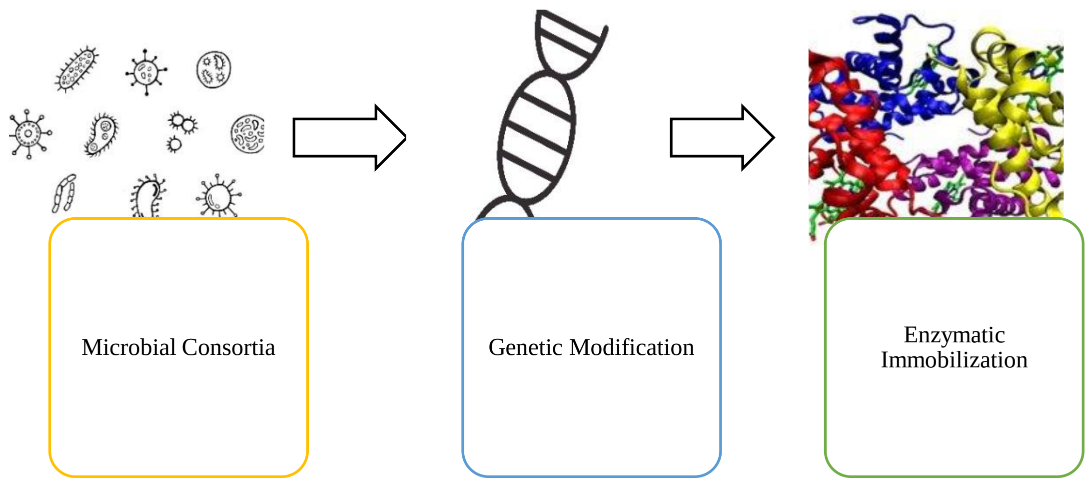


FIGURE 3: Illustrating methods for biomass bio-degradation

Enzymatic Immobilization

Nanoparticles such as mesoporous silica can be used for mobilization of enzyme "Fig. 3" of interest as they provide larger external area and wide-ranging pH for enzymatic activity [30].

CONCLUSION

Use of biofuels is the demand of present world but due to high production cost the expectations are not yet fulfilled. The biological pre-treatment process can be used for the same. This process is not only environment friendly but can also reduce the cost of production. Microbial enzyme production can be enhanced using different methods such as consortia preparation, heterologous gene expression and polymeric matrix support for enzyme expression. Many approaches for genetic manipulation have failed to produce desirable enzyme production. Also, consortia preparation can be an intimidating task as the microbes share different niche and genetic makeup. However, if these obstacles can be cleared, a sustainable process can be developed for cost effective biofuel production.

REFERENCES

1. M. Kumar and A. Varma, Sci Intl, 1-11 (2016).
2. E. M. Aro, Ambio **45**, 24–31 (2016).
3. K. Dutta, and A. Daverey, Renew **69,** 114–122 (2014).
4. K. Janda, and L. Kristoufek, Agric Econ **58,** 372–386 (2012).
5. J. Pérez, and J. Muñoz-Dorado, Int Microbiol **5,** 53–63 (2002).
6. D. F. Barnes, and W. M. Floor, AnnuRev Energy **21,** 497–530 (2003).
7. A.V. Gusakov, Trends Biotechnol **29**, 419–425 (2011).
8. S. Ellilä, and L. Fonseca, Biotechnol Biofuels **10**, 1–17 (2017).
9. A. K. Kumar and S. Sharma, Bioresour Bioprocess **4,** (2017).
10. L. Canilha, and K. A. Chandel, Journal of Biomed Biotechnol, (2012).

24 February 2024 17:48:20

11. Y. Zheng, and Z. Pan, Int J Agric Biol Engineering **2**, (2009).
12. D. Chettri, and A. K. Verma, Biotecnol Rep **28**, (2020).
13. L. Pollegioni and F. Tonin, FEBS Journal **282**, 1190–1213 (2015).
14. M. E. Scharf, and A. Tartar, Biofpr **2**, 540–552 (2008).
15. W. F. Anderson, and D. E. Akin, J Ind Microbiol Biotechnol **35**, 355–366 (2008).
16. D. Martinez, and L. F. Larrondo, Nat Biotechnol **22**, 695-700 (2004).
17. D. Martinez, and J. Challacombe, PNAS **106**, 1954-1959 (2009).
18. B. Tsegaye and C. Balomajumder, Bull Natl Res Cent **43**, (2019).
19. M. Dashtban and H. Schraft, Int J Biol Sci **5**, 578-595 (2019).
20. Y. Zeng and M. E. Himmel, Biotechnol Biofuels **10**, 1-16 (2017).
21. F. Rasul, and A. Afroz, Int J Biosci, 230-238 (2015).
22. W. R. de Souza, InTechOpen, (2013).
23. L. Munk, and A. K. Sitarz, Biotechnol Adv **33**, 13-24 (2015).
24. D. N. Georgiadou, and P. Avramidis, Heliyon **7**, 7 (2021).
25. M. L. Maki, and L. Armstrong, Bioengineered **4**, 15-20 (2013).
26. M. F. Adegboye and O. B. Ojuederie, Biotechnol Biofuels **14**, 1-21 (2021).
27. H. K. Sharma and C. Xu, Waste Biomass Valorization **10**, 235-251 (2019).
28. R. G. de Paula and A. C. C. Antoniêto, Biotechnol Adv **37**, 107-347 (2019).
29. U. Azimov and V. Okoro, Energies **14**, 6011-14 (2021).
30. S. Dutta, and K.C. Wu, Green Chem **16**, 4615-4626 (2014).

24 February 2024 17:48:20