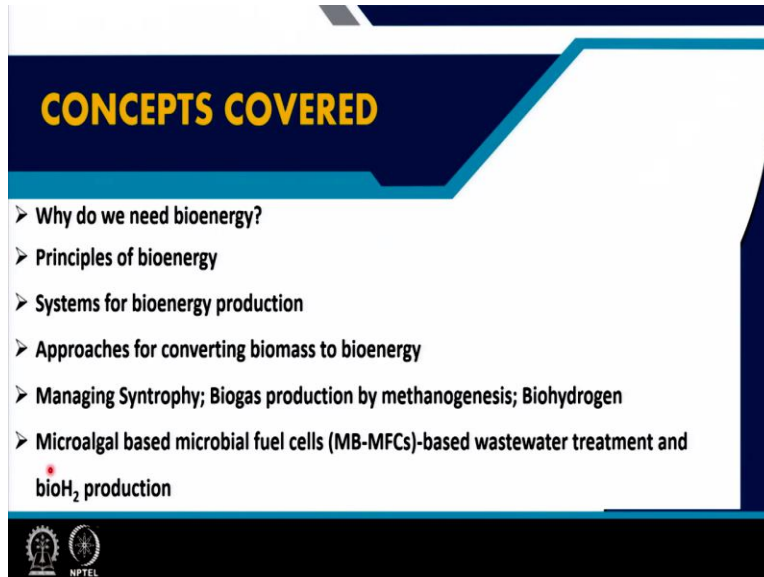


Environmental Biotechnology
Prof. Pinaki Sar
Department of Biotechnology
Indian Institute of Technology, Kharagpur


Lecture – 50
Bioenergy and Environmental Biotechnology

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CONCEPTS COVERED

- Why do we need bioenergy?
- Principles of bioenergy
- Systems for bioenergy production
- Approaches for converting biomass to bioenergy
- Managing Syntrophy; Biogas production by methanogenesis; Biohydrogen
- Microalgal based microbial fuel cells (MB-MFCs)-based wastewater treatment and bioH₂ production



Welcome to the next lecture of this course on bioenergy and environmental biotechnology and in this particular lecture the following concepts will be covered. We will talk about the need for bioenergy and the principles of bioenergy production. We will discuss in detail about the systems for bioenergy production and different approaches and particularly the the syntrophy and the how the syntrophic associations are managed to produce the bioenergy with emphasis on biogas production by methanogenous, methanogenesis, biohydrogen production.

And then micro algal based microbial fuel cell and bio hydrogen production are also will be discussed at the end we will talk about a case study.

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Bioenergy

Sun's energy captured in biomass and converted to energy forms, It is a form of renewable energy


Why do we need bioenergy?

Overuse of fossil fuels (13 terrawatts (TW) annually) poses three giant risks for the survival of human society

1. Depletion of fossil-fuel reserves
2. Geopolitical strife from competition for dwindling resources may lead to economic and energy disruptions, political turmoil, and war
3. Increase in atmospheric CO₂ concentration and global warming, leading to climate change

Eliminating the **third** risk by finding substitutes for fossil fuels should have a **happy** consequence of minimizing risks one and two

Biomass derived energy or bioenergy can be effective substitutes for fossil fuels and mitigate the climate change



So, bioenergy to introduce with bioenergy the Sun's energy is captured in biomass through photosynthesis and converted into energy forms and it is the form of a renewable energy. So, bioenergy basically represents a renewable form and why do we need this bioenergy? Overuse of fossil fuels poses three great risks for the survival of human society. And these risks include the depletion of the fossil fuel reserves geopolitical strife from competition for dwelling resources which may lead to economic and energy disruptions political turmoil and also perhaps war.

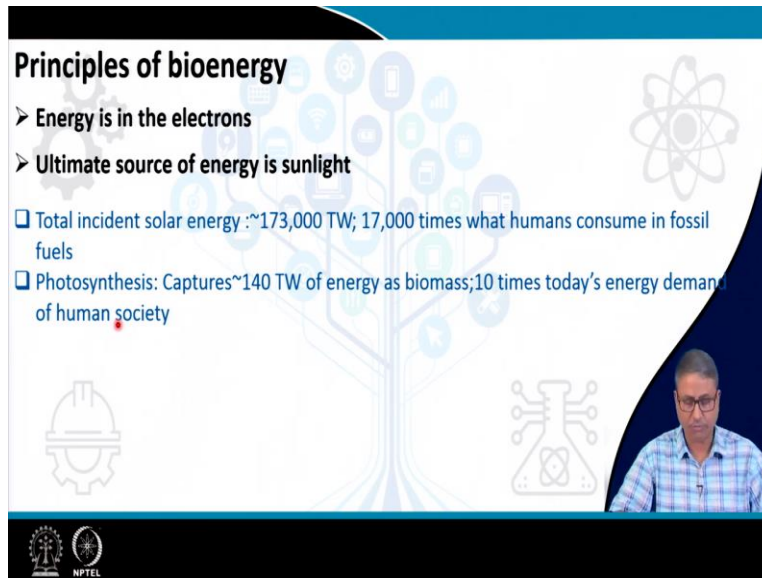
Increase in atmospheric CO₂ concentration and global warming leading to the climate changes. Now eliminating the third risk that is the enhanced release of carbon dioxide from the fossil fuel and enhancing the risk associated with the climate change and the global warming. So, eliminating this third risk by finding substitutes for fossil fuels should have the happy consequence for minimizing risk one and two naturally.

So, the fossil fuel is going to be depleted one day. So, on the other hand the way the carbon dioxide is increasing. So, we need to switch to some technologies which will reduce carbon dioxide emission. So, the bioenergy provides the options to combat both the challenges together one way it helps us to mitigate the climate change global warming related issues by reducing the carbon emissions.

And the other way it provides us the alternative to the fossil fuel reserve that is the sustainable

and renewable energy sources and also minimize the conflicts that is rising out of the natural resources of this conventional energy resources. Now biomass derived energy or the bioenergy can be an effective substitute for fossil fuel and will eventually help in mitigating the climate change.

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Principles of bioenergy

- Energy is in the electrons
- Ultimate source of energy is sunlight
- ☐ Total incident solar energy :~173,000 TW; 17,000 times what humans consume in fossil fuels
- ☐ Photosynthesis: Captures~140 TW of energy as biomass;10 times today's energy demand of human society

The slide features a background graphic of a tree with various icons (gears, Wi-Fi, smartphone, etc.) on its branches. There are also icons of a hard hat, a chemical flask, and an atom. A small video inset in the bottom right corner shows a man in a plaid shirt speaking. The NPTEL logo is visible in the bottom left corner.

Now what are the principles of bioenergy? This energy that we harvest through bioenergy process is basically the energy is relying within the electrons. And it is considered to be the ultimately the solar energy of the light energy which is the ultimate source of the energy in most of this bioenergy resources. And if we look at the data that the total incident solar energy versus the photosynthetically captured energy we will realize the importance of this bioenergy.

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Principles of bioenergy

- Energy is in the electrons
- Ultimate source of energy is sunlight

Diverting food crops to biofuels compete with food production and their bioenergy output is limited

C-neutral source, photosynthetic energy capture must be expanded to produce some "new biomass"

Residual biomass from human activities, agriculture, food-producing industry, municipal and industrial wastewaters can easily meet world's energy demand

The slide features a blue header, a white background with faint icons of a gear, a tree, and a molecular structure, and a black footer with the NPTEL logo. A presenter is visible in a small window on the right side of the slide.

Now diverting the fruit food crops to biofuel compete with food production and their bio energy output is therefore limited. So, although in the initial days of bioenergy research. There was some kind of discussion and work going on towards utilizing the crops which might be very useful bio fuel resource but because they compete with the food production and use of water etcetera. So, the scope was found to be limited. Carbon neutral source photosynthetically mediated energy capture therefore considered to be must be expanded to produce some new biomass.

And residual biomass also from human activities including the agriculture food producing industry municipal and industrial wastewater can easily meet the worlds energy demand.

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Systems for bioenergy production

- Microbial communities must have high energy yield, robustness and self-stabilization
- Microbial communities must be resilient to fluctuations in environmental conditions, variations in nutrient and energy inputs and intrusion by microbial invaders that might consume the desired energy product
- System robustness (in terms of energy output) demands a high level of self-stabilization of the function of the community diverse community with substantial functional redundancy is needed
- Complex and mixed communities perform the desired function of bioenergy generation
- Understanding key community members, allow us to track communities and their metabolic reactions, the rate of process development and the reliability of the processes

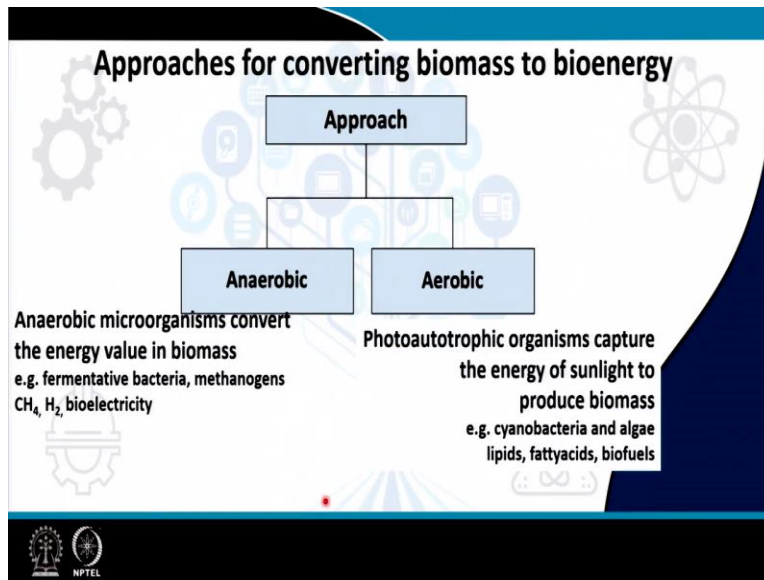
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Now in this respect when we think of converting energy which is naturally available within the different biological resources including the wastewater also. We need to have a better understanding and better appreciation of the microbial communities which are actually working towards converting this different energy forms to usable energy form. Now the microbial communities responsible for catalytic these events must have the high energy yield robustness and self stabilization.

And microbial communities must be resilient to fluctuations in environmental conditions variations in nutrients and energy input intrusion by microbial invaders that might consume and desired energy products. So, in any system where we are planning to develop bioenergy process using microorganisms we need to understand these situations. And also the system robustness that is in terms of the energy output which demands a high level of self stabilization of the function of the community diverse community members and substantial functional redundancy which is needed.

So, complex and mixed communities are found to perform the desired function of bioenergy generation and understanding the key community members which are responsible for such reactions allow us to track the communities and their metabolic reactions the rate of process development and the reliability of the different processes.

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So, the approaches for converting biomass and bioenergy are quite well defined as you can see that we can have anaerobic or aerobic approaches. So, within the anaerobic approach is the anaerobic micro organisms are used. And these organisms which are in complex communities they convert the energy value of the biomass through fermentative reactions and methanogenic reactions and they enable the production of methane hydrogen bioelectricity etcetera.

On the other hand the aerobic approaches where the photo autotrophic organisms are involved they capture the energy of sunlight into biomass for example the cyanobacteria and algae and then that biomass bound energy can be converted to usable form like the lipids fatty acids and other biofuel molecules.


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Approaches for converting biomass to bioenergy

Approach

Benefits

- High specific growth rates, year round harvesting and homogeneity,
- Produce larger (by 100-fold or more) biomass-based energy stocks than plants
- Does not compete with food production
- Generation of renewable energy
- Minimization of environmental pollution
- Global warming mitigation





Now the benefits of these approaches are high specific growth rates year round harvesting and homogeneity produce larger like hundred fold or more biomass based energy stock than plants does not compete with food production generation of renewable energy minimization of environmental pollution and also it helps in mitigating the global climate change problem, global warming.

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Managing Syntrophy


- Self-stabilizing communities involved in biomass conversion to CH_4 , H_2 or electricity involve essential syntrophies
- To produce CH_4 , methanogens use only acetate or H_2 and CO_2 , and all of the electron flow must be funnelled through H_2 and acetate
- Likewise, oxidation of H_2 to form CH_4 by methanogens must be suppressed when H_2 production is the goal

In a microbial fuel cell (MFC), build-up of H_2 could lead to two undesired outcomes:

- slowed fermentation
- diversion of electron flow to CH_4

Balanced syntrophy between H_2 -producing fermenting bacteria and H_2 -oxidizing methanogens is the key to reliable methanogenesis and biohydrogen production

Synergy between phototrophs and an array of heterotrophic bacteria is essential for the operation of a microbial photobioenergy system



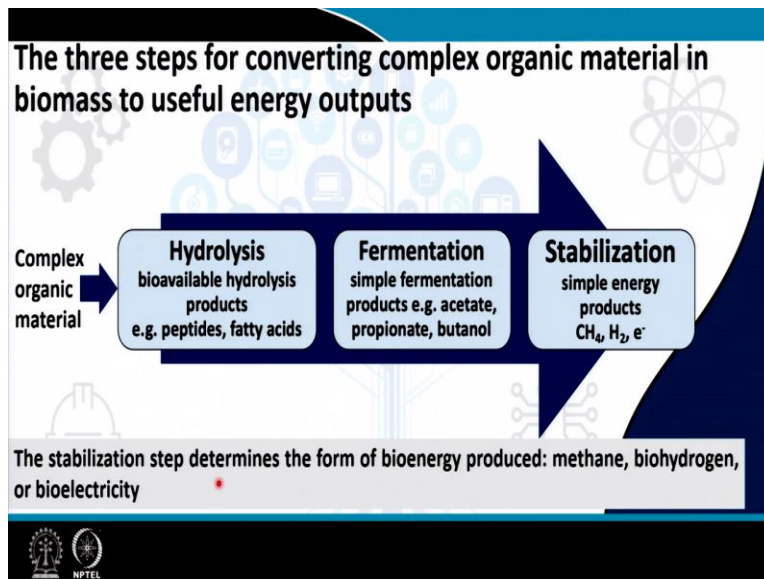
Now managing the syntrophy which is found to be the close association between different group of microorganisms which drive this production of this energy resources are found to be an important component. Now, self stabilizing communities with participation of multiple members involved in biomass conversion to methane and hydrogen or electricity that are actually

catalyzed by different members of the syntrophic association to produce methane.

Methanogens they use only acetate or hydrogen and carbon dioxide and all of the electrons must be funneled through hydrogen and acetate likewise the oxidation of hydrogen to form methane by methanogens must be suppressed when hydrogen production is the goal. In a microbial fuel cell for example buildup of hydrogen could lead to two undesired outcomes one is the slowing down of the fermentation and diversion of electron flow to methane.

Essentially the balanced syntrophy between hydrogen producing fermenting bacteria mostly the secondary fermenters and hydrogen oxidizing methanogens is found to be a key component for the reliable methanogenesis and bio hydrogen production. And the synergy between the phototrophs and an array of heterotrophic bacteria is also essential for the operation of the microbial photobioenergy system.

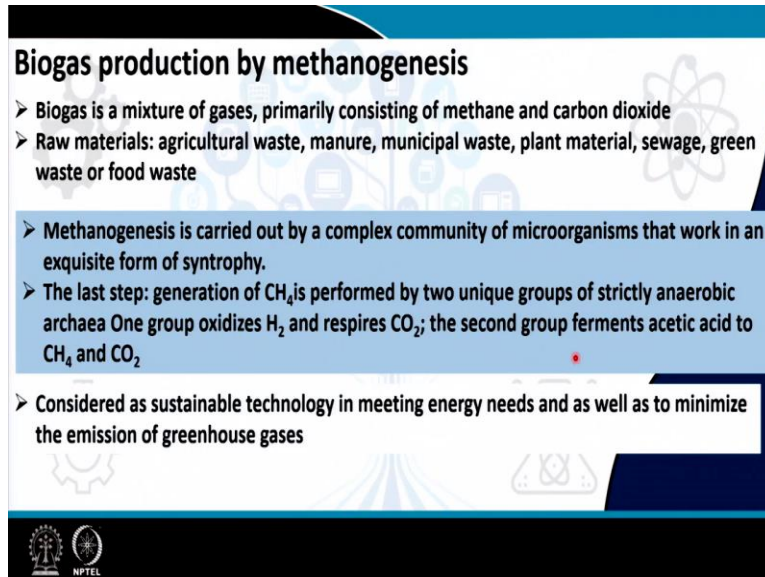
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Now here are the three steps for converting the complex organic material which are present in a biomass or in other materials into useful energy inputs. One is the hydrolysis where the bioavailable hydrolysis products. So, make them bioavailable for example the peptide peptides fatty acid etcetera then the fermentation whether simple fermentation products for example acetate propionate butanol etcetera are produced.

And then finally the stabilization where the simple energy products like methane hydrogen electrons are produced. The stabilization step determines the form of bioenergy produced like methane biohydrogen or bioelectricity.

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Biogas production by methanogenesis

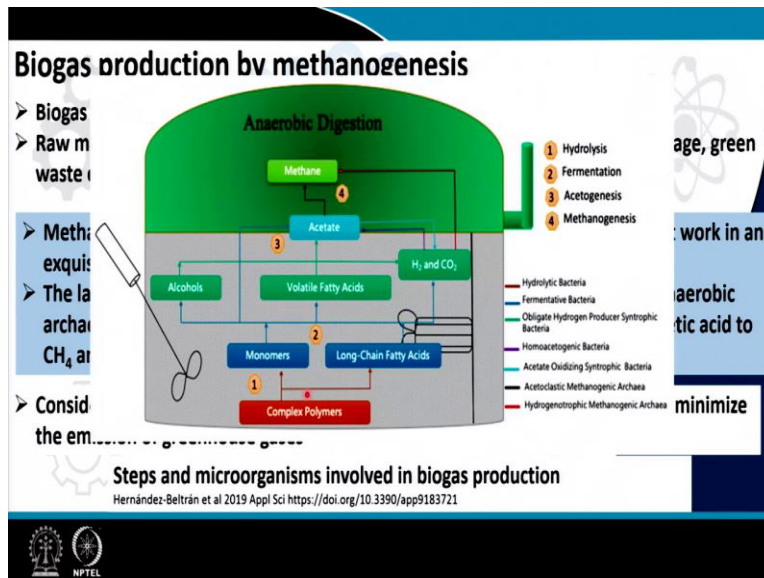
- Biogas is a mixture of gases, primarily consisting of methane and carbon dioxide
- Raw materials: agricultural waste, manure, municipal waste, plant material, sewage, green waste or food waste
- Methanogenesis is carried out by a complex community of microorganisms that work in an exquisite form of syntrophy.
- The last step: generation of CH_4 is performed by two unique groups of strictly anaerobic archaea. One group oxidizes H_2 and respire CO_2 ; the second group ferments acetic acid to CH_4 and CO_2
- Considered as sustainable technology in meeting energy needs and as well as to minimize the emission of greenhouse gases

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Now next we will discuss the biogas production by methanogenesis which is of course a part of the syntrophy. Now biogas is a mixture of gases primarily consist of methane and carbon dioxide and the various raw materials including the agricultural waste manure municipal waste, plant material, sewage, green waste or food waste are utilized to produce the methanol biogas. Methanogenesis as we have understood earlier that is carried out by complex community of microorganisms particularly the archaea as a part of the syntrophic association.

And it is generally the last step that is followed by the primary oxidation and the secondary fermentation the generation of methane is performed by strictly anaerobic archaea and we have the hydrogen utilizing archaea and the acetate utilizing archaea who are responsible for converting this substrate into methane. And this is considered as a sustainable technology in meeting the energy needs and as well as to minimize the emission of the greenhouse gases.

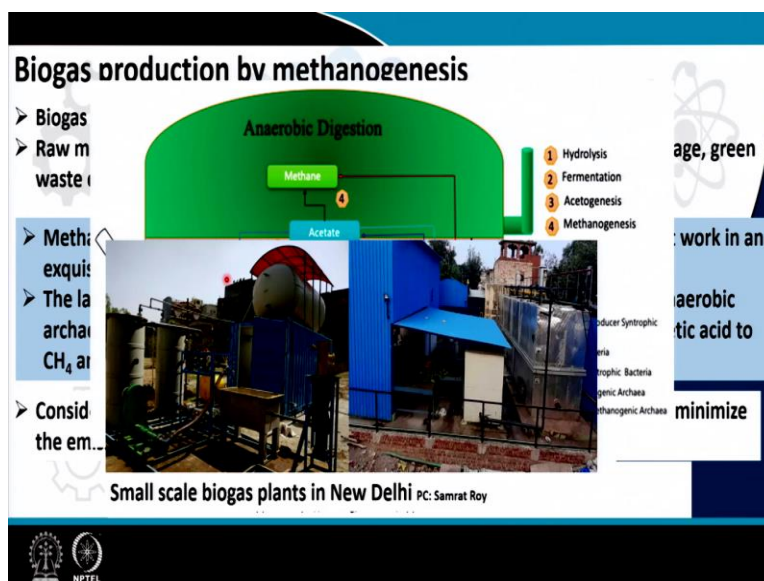
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For example if we look at the steps of microorganisms involved in the biogas production we can see that the anaerobic digester where we have the complex polymers which are first broken down into monomer and long chain fatty acids through hydrolysis and then they are subjected to fermentation. And followed by fermentation the alcohols and volatile fatty acids hydrogen and carbon dioxides are produced through acidogenesis.

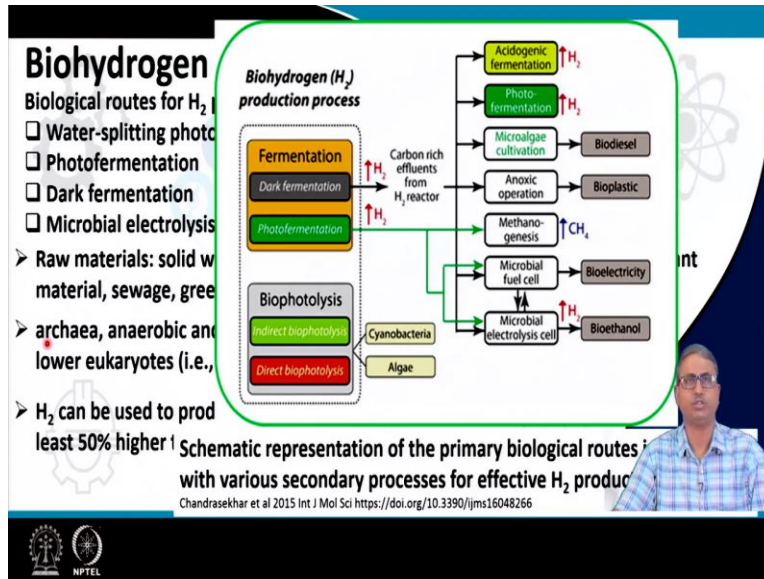
And then the methanogenesis they use this substrate either the acetate or the hydrogen carbon dioxide to produce the methane.

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And here is a picture of a small scale biogas plant which is located in New Delhi.

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Now next is the biohydrogen, biological routes for hydrogen production are considered to be very important and efficient process and are categorized into four groups like water splitting photosynthesis photo fermentation dark fermentation and microbial electrolysis processing raw materials like solid waste wastewater agricultural waste municipal waste plant material sewage and diverse materials can be used to facilitate these reactions.



And organisms like archaea anaerobic and facultative aerobic bacteria cyanobacteria and lower eukaryotes like green algae and protist they can produce hydrogen. And hydrogen can be used to produce pollution free electrical energy with an efficiency least at least 50% higher with a combustion steam turbine turbine approach. So, here you can see a systematic approach or schematic approach of the primary biological routes of hydrogen production from different substrate and different reactions are also mentioned and eventually we get the the hydrogen produced through these steps.

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Microalgal based microbial fuel cells (MB-MFCs)-based wastewater treatment and bioH₂ production

- Integration of MB-MFCs system with microalgal based bioH₂ production are a cost-effective approach for wastewater treatment and bioH₂ production
- Wastewater is used as nutrient rich substrates for bacterial growth, the bacteria oxidize the substrates and generates H⁺ and e⁻, the e⁻ moves towards the anode, and is then transferred to the cathode where electron flow generates a bioelectric current
- Then, H⁺ moves towards cathode through the proton membrane exchanger and reacts with O₂ (which are produced during microalgal respiration) and forms H₂O
- In algal cells, direct photolysis occurs, which produces bioH₂, furthermore, produced algal biomass can be utilized for this production.

Mehariya et al 2021 Energies <https://doi.org/10.3390/en14082282>

The micro algal based microbial fuel cells MB-MFCs are also getting increased attention because they are allowing wastewater treatment and production of the biohydrogen where integration of the micro algal based micro fuel cell microbial fuel cell system with we have a micro algal based hydrogen production our cost effective approach for waste water treatment because the waste is also treated and the carbon and electron which are there are converted to basically the electrons are converted to the in the top they are they are used for the production of the hydrogen.

Waste water is used as a nutrient rich substrate for bacterial growth the bacteria oxidize the substrate and generate the proton and electron and the electron moves towards the anode and then transferred into the cathode where electron flow generates a bio electric current now. Then the hydrogen or the proton moves towards the cathode through the proton membrane exchanger and reacts with the oxygen which are produced during the micro algal respiration and from the water.

In algal cells direct photolysis occurs which produces biohydrogen furthermore the produced algal biomass can be utilized for this production.

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Microalgal based microbial fuel cells (MB-MFCs)-based wastewater treatment and bioH₂ production

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Mehariya et al 2021 Energies <https://doi.org/10.3390/en14082282>

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So, you can see here a kind of a schematic diagram is presented here where the waste water can be treated using the micro algal fuel microbial fuel cell to produce the hydrogen.

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Bioelectricity generation by microbial fuel cells (MFCs)

CO₂

Exoelectrogen Microorganisms

Electron donor (Organic - inorganic substrate)

Anode

Anaerobic Chamber

Proton Exchange Membrane

Cathode

Aerobic Chamber

Schematic representation of a two-chambered MFC

Ucar et al 2017, Front Microbiol <https://doi.org/10.3389/fmicb.2017.00643>

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Similarly the bio electricity can also be generated by microbial fuel cell where we see that the bacterial cells are used to reproduce the required electrons and then electrons are allowed to flow through it.

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Bioelectricity generation by microbial fuel cells (MFCs)

- MFC is a special form of a fuel cell in which microorganisms grow as a biofilm on the anode
- The bacteria catalyze the oxidation of organic compounds either in or derived from biomass (after it has gone through fermentation), and the bacteria then transfer the electrons to the anode
- The electrons move through an electrical circuit to the cathode to an electron acceptor, normally oxygen (O₂)
- The energy value of the electrons is harvested in the circuit as electrical power, again an electron sink that naturally leaves the water
- Microorganisms utilized in MFC: *Geobacter*, *Rhodobacter*, *Rhodospirillum*, *Nostoc*, *Acidiphilium*, *Haloferax*
- Generate combustion-less, pollution-free bioelectricity directly from the organic matter present in biomass



Now MFC's are kind of a special form of fuel cell in which microorganisms grow as biofilm on the anode and the bacteria they catalyze the oxidation of organic compounds either in or derived from the biomass after it has gone through fermentation and the bacteria then transfer the electrons to the anode. The electrons then move through the electrical circuit to the cathode to an electrical electron acceptor and normally the oxygen.

The energy value of the electron is harvested in the circuit as electrical power again an electron sink that naturally leaves the water. And these following microorganisms like geo bacter rhodobacter rhodo spirulium non stock acidophylum haloferox are commonly used in MFCs and this they generate combustion less pollution free bioelectricity directly from the organic matter present in the biomass.

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Biodiesel
Photosynthetic microorganisms that capture sunlight energy and concentrate it in lipids that can be used to produce biodiesel

Biodiesel can be produced from three sources

- Residual fats from food preparation
- Lipids extracted from high-oil plants, soybeans, sunflowers, and Jatropha.
- Lipids extracted from photosynthetic microorganisms, algae and cyanobacteria

Photosynthetic microorganisms (algae or cyanobacteria) offer many significant advantages

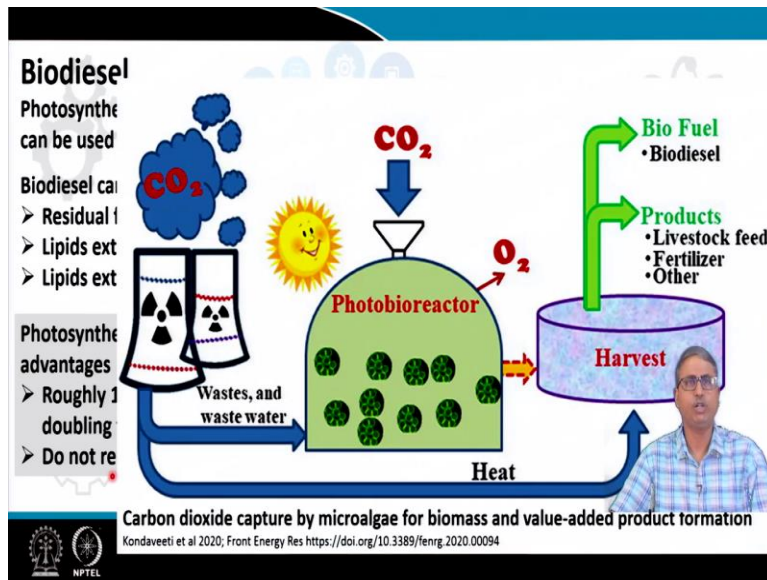
- Roughly 100-fold higher yield of lipids per hectare from other sources much shorter doubling time, continuous harvesting, and a more homogeneous physical structure
- Do not require arable land and need not compete with food production

The slide features a video inset of a man in a blue plaid shirt speaking. The background includes a stylized atom symbol and a network of nodes. Logos for IIT Bombay and NPTEL are visible at the bottom left.

Similarly the biodiesel is found to be an another very important form of bioenergy where photosynthetic microorganisms are used that can capture the sunlight energy and concentrate it in lipids that can be used to produce the different forms of the biodiesel. The biodiesel can be produced from this source by as a residual fat from food preparation lipid extracted from high oil plants like soya beans, sunflowers, jetrofa. Lipids extracted from photosynthetic microorganisms, algae, cyanobacteria etcetera.

Photosynthetic microorganisms like algae and cyanobacteria offer many advantages like roughly hundred fold higher high yield of lipid per hectare from other sources much shorter doubling time continuous harvesting process and a more homogeneous physical structure is available. And they do not require arable land and need not compete with food production.

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For example the carbon dioxide captured by microalgae for biomass and formation of value added product formation is shown in the diagram. Where you can see that the waste materials are used in the photobioreactor where further carbon dioxide is captured and then the biomass is produced which is converted into a kind of a livestock feed fertilizer and also the biodiesel molecules.

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India's first 2nd generation (2G)-alcohol plant

- India's first 2G-alcohol demonstration plant built by India Glycols Lt. at their Kashipur site
- Based on indigenous and unique DBT-ICT 2G-Alcohol Technology developed by the DBT-ICT Centre for Energy Biosciences at Institute of Chemical Technology, Mumbai, produces ethanol from agriculture residues
- Potential to significantly reduce emissions from the transportation and agricultural sectors in India
- Converts biomass feed to alcohol within 24 hours
- Ethanol production of 1 ton/day scale during Phase 1 at the India Glycols site
- Scaled up to a demonstration scale plant of 750,000 litres annual alcohol capacity

Now at the last we will talk about case study for bioenergy which is the India's first second generation or two g alcohol plant. So, this is India's first two g alcohol demonstration plant built in India Glycols limited at the Kashipoo site based on the indigenous and unique DBT ICT 2G alcohol technology developed by the DBT ICT center for energy bioscience at Institute Of

Chemical Technology, Mumbai they can produce ethanol from the agricultural residues.

And it has shown potential to significantly reduce the emissions from the transportation and agricultural sectors in India. And it readily converts the biomass agricultural waste biomass into alcohol within 24 hours. And ethanol production of one ton per day scale during phase one at the India Glycol site was at further scaled up demonstration plant with 750000 liters annual alcohol capacity.

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India's first 2nd generation (2G)-alcohol plant

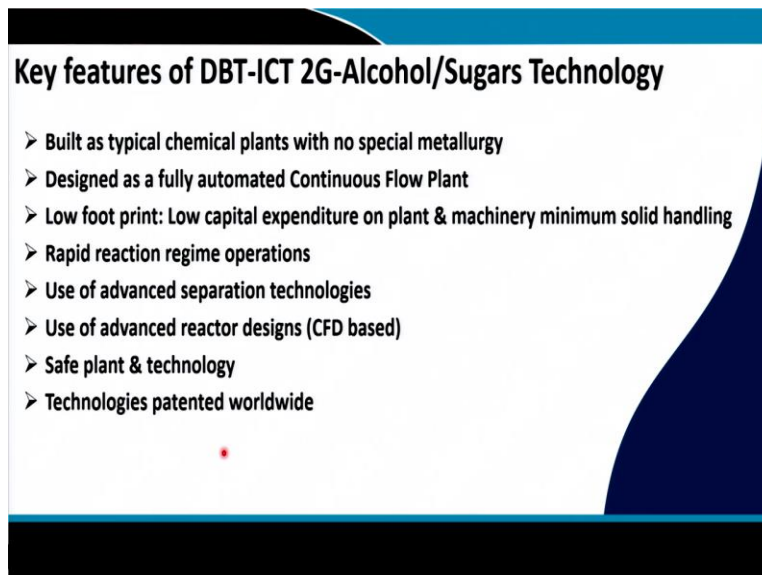
- India's first 2G-alcohol demonstration plant built by India Glycols Lt. at their Kashipur site
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https://ec.europa.eu/energy/sites/ener/files/documents/32_arvind_lali-dbt-ict.pdf

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So, here is the picture of the plant you can see.

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Key features of DBT-ICT 2G-Alcohol/Sugars Technology

- Built as typical chemical plants with no special metallurgy
- Designed as a fully automated Continuous Flow Plant
- Low foot print: Low capital expenditure on plant & machinery minimum solid handling
- Rapid reaction regime operations
- Use of advanced separation technologies
- Use of advanced reactor designs (CFD based)
- Safe plant & technology
- Technologies patented worldwide

And the key feature of this strategy is or the technology is the built as a typical chemical plant with no special metallurgy. Designed as a fully automated continuous flow plant low footprint low capital expenditure on the plant and machinery minimum solid handling rapid reactor regime operation and safe plant and technology and technological technologies are patented worldwide.

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Key features of DBT-ICT 2G-Alcohol/Sugars Technology

- Built as typical chemical plants with no special metallurgy
- Designed as a fully automated Continuous Flow Plant
- Low footprint: Low capital expenditure on plant & machinery minimum solid handling
- Rapid reaction regime
- Use of advanced separation technologies
- Use of advanced reactor technologies
- Safe plant & technology
- Technologies patented

Timeline of plant development:

- Lab scale process (1 kg/day): 2009- present
- Pilot Plant (1 ton/day): 2010-2013
- Pre-commercial scale (10 ton/day): March 2016

https://ec.europa.eu/energy/sites/ener/files/documents/32_arvind_lali-dbt-ict.pdf

So, and you can see here the events of progress with lab scale 1 kg per day waste conversion in 2009 to 1 ton and finally 10 ton waste per day which is being done at in this facility in March 2016.

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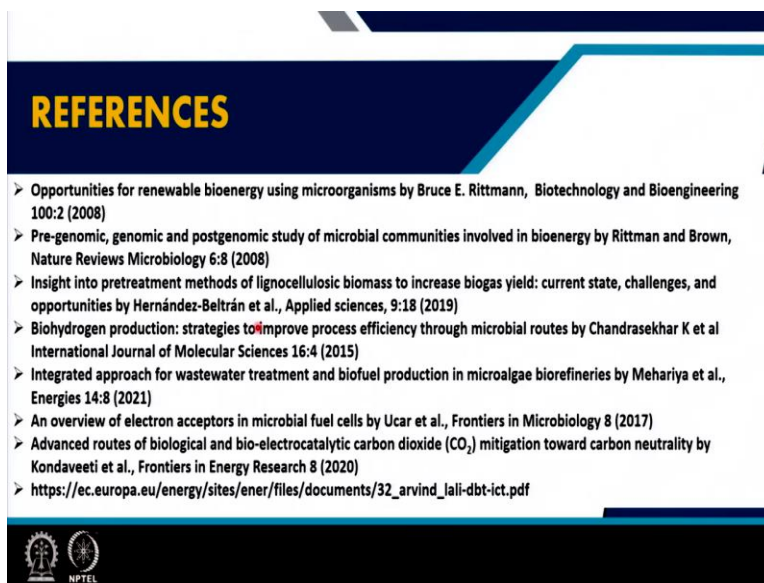
Technology performance achieved

- Continuous flow plant: from pretreatment to fermentation
- Designed for handling all agricultural residues: bagasse; rice straw; wheat straw; bamboo; cotton stalk; corn stover; wood chips etc.
- Fastest conversion of all feedstock to ethanol in < 24 hours
- Chemicals and enzymes separated, recycled & reused: low opex
- Zero discharge technology
- Value addition from mineral fertilizer, silica (rice straw), and lignin (cotton stalk)
- Separate streams of C6 and C5 sugars in ~90% purity
- Enzyme use: 1kg/ton Biomass
- Fermentation yield: 0.43g/g combined sugars
- Choice between combined or separate fermentations of sugars
- Cost of production (excl. biomass cost) < 0.4USD/L ethanol

And there are a large number of technological performance which are achieved and which

include the continuous flow plant from pre treatment to fermentation. Designed for handling all different kind of agricultural residues, fastest conversion of all feedstock to ethanol that is less than 24 hours chemicals and enzymes are separated recycled and reused. So, zero discharge technology value addition from mineral fertilizer separates streams of C6 and C5 sugars in 90% purity enzymes use one kg per ton biomass etcetera.

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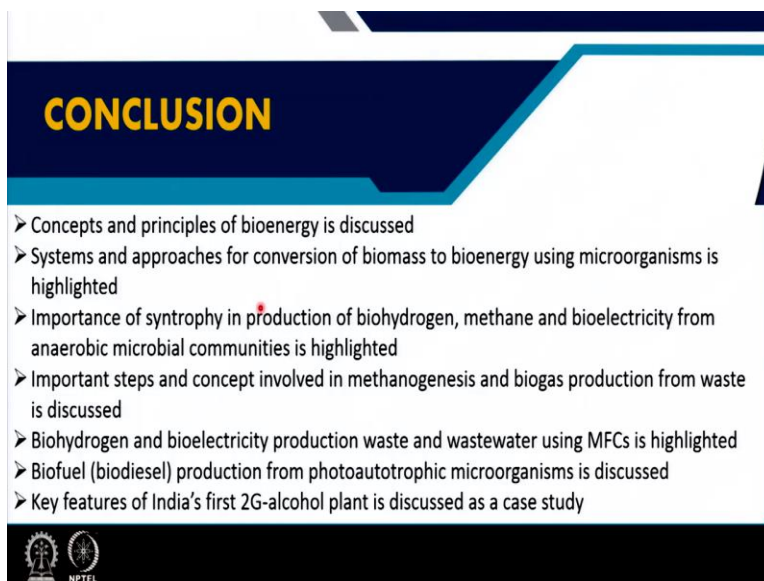


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CONCLUSION

- Concepts and principles of bioenergy is discussed
- Systems and approaches for conversion of biomass to bioenergy using microorganisms is highlighted
- Importance of syntrophy in production of biohydrogen, methane and bioelectricity from anaerobic microbial communities is highlighted
- Important steps and concept involved in methanogenesis and biogas production from waste is discussed
- Biohydrogen and bioelectricity production waste and wastewater using MFCs is highlighted
- Biofuel (biodiesel) production from photoautotrophic microorganisms is discussed
- Key features of India's first 2G-alcohol plant is discussed as a case study

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So for these details the following references can be used and in conclusion the concept and principle of bioenergy with relevance to environmental biotechnology is discussed, systems and approaches for conversion of biomass to bioenergy using microorganisms is highlighted

importance of syntrophy in producing bioenergy like biohydrogen, methane and also electricity from anaerobic microbial communities highlighted.

Important steps and concept involved in methanogenesis and biogas production from waste discussed and biohydrogen, bio electricity production from waste water including MFCs and bio fuel production from photo autotrophic algae is highlighted. And finally the India's first 2G alcohol plant is discussed as a case study and thank you so much.