

Non sea algae methanization

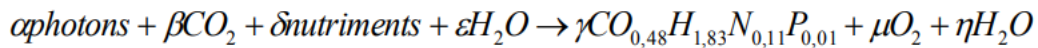
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1. Specific biomass to treat

There are plenty of different species of non algal biomass, in this study primarily focus in *Chlorella Sorokiniana* and *Tetraselmis Suecica*.

Microalgae are photosynthetic microorganisms that facilitate their growth by converting solar energy into chemical energy in the presence of inorganic elements (carbon dioxide, nitrogen, phosphorus, potassium, etc.), water, and light energy, according to the following reaction:



Inorganic carbon and water are transformed into algal biomass through this reaction.

Microalgal biomass, like of all living organisms, consists of three major elements: proteins, lipids, and carbohydrates. The average proportions of these elements are listed in Table 1, along with their calorific values (3). These proportions heavily depend on the species in question and the environmental conditions.

Table 1. Microalgae composition

	Proportion %	Calorific power (MJ/kg)
Proteins	6-52	15,5
Lipids	7-23	38,3
Carbohydrates	5-23	13
Ratio C: N : P	106:16:1	

Microalgae are autotrophic organisms and require inorganic elements for their growth.

The general equation for algal biomass, $\text{CO}_{0,48} \text{H}_{1,83} \text{N}_{0,11} \text{P}_{0,01}$ allows to estimate the quantity of nutrients necessary for the algae growth:

- Carbon: supplied in the form of carbon dioxide.
 - Requirement: 1.83 kg for 1 kg of algal biomass on a dry mass basis.
- Nitrogen: supplied as nitrate, ammonia, or urea.
 - Requirement: 0.07 kg for 1 kg of algal biomass on a dry mass basis.
- Phosphorus: supplied as phosphate.
 - Requirement: 0.01 kg for 1 kg of algal biomass on a dry mass basis.
- Other nutrients are essential in trace amounts: iron, magnesium, manganese, nickel, zinc, molybdenum, cobalt, boron, vanadium, and copper. However, if their concentrations are too high, these elements can be toxic to the culture.

The nutrient requirements can vary depending on the species of microalgae. However, nutrients need to be continuously supplied to optimize its growth.

For example, in wastewater as nutrients microalgae can use nitrates and phosphates present there, which also will reduce the level of these two elements in the water. They also have the ability to capture certain heavy metals.

1.1. Production of Biomethane by Anaerobic Digestion

Methanization is a natural process involving the breakdown of biodegradable matter in the absence of air (anaerobic transformation). It's a semi-controlled process in which bacteria convert biomass into methane and carbon dioxide. This digestion involves different stages (hydrolysis and acidification, acetogenesis, and methanogenesis), which will be explained in more detail later in the document.

Methanization is currently a mature process at an industrial scale and commercially available. The two products of this transformation are:

- Biogas: a gas mixture saturated with water composed of approximately 30% to 70% CH₄ and 20% to 40% CO₂, along with some trace gases (NH₃, N₂, H₂S).
- Digestate: a liquid or paste rich in organic matter that can be used as a fertilizer.

Before being utilized, biogas must undergo treatments to remove pollutants harmful to downstream facility operations. The level of treatment depends on the type of valorization; generally less intense treatments (desulfurization, dehydration) are used for heat and/or electricity production.

Early studies on anaerobic digestion of microalgae for energy purposes were conducted using microalgae species collected from open basins for water treatment. Studies indicate that digestion of microalgal biomass occurs within retention times ranging from 10 to 40 days, with methane production ranging from 0.1 to 0.4 L/gVSS.

A promising approach is to use rapidly growing species to ensure sufficient biomass. However, it's important to mention that the amount of biogas produced can differ significantly depending on the species, easily doubling or halving.

The production of biomethane from a culture of microalgae using both culture technologies is depicted in Table 2 and is compared to biomethane production associated with other conventional biomasses.

Table 2: Production of biomethane from microalgae

TYPE OF BIOMASS	Productivity (T DM/ha/yr)	Production of Biomethane (m ³ /T DM)	Productivity of Biomethane (m ³ /ha/yr)	Associated Energy Production (MWh/ha/yr)
PONDS	27	242	6.520	65
PHOTOBIOREACTORS	76	242	18.392	184
MAIZE	9 - 30	205 - 450	1 660 – 12 150	17 - 122
WHEAT	3,6 – 11,75	384 - 426	1 244 – 4 505	12 – 45

1.2. Integration of Third-Generation Biomethane Production Chain

Among the various pathways for energy valorization of microalgae, anaerobic digestion stands as the most mature and currently requires no major technological barriers. The production of biogas from microalgae through a methanization process, includes a subsequent conversion into biomethane.

Moreover, it smoothly fits into the entire process of utilizing microalgae, as illustrated in the next figure:



Figure 1. Methanization process

There is two integration modes of the methanization unit into the microalgae production process:

1. Direct energy valorization of microalgae. A portion of the microalgal biomass is allocated for the production of feed, food, etc. Meanwhile, the remaining portion of this biomass is utilized for energy through the process of methanization. The allocation for methanization depends on the quality of the obtained microalgal biomass and also meets production needs.
2. Energetic valorization of the residue after extracting compounds of interest from microalgae. The valorization of microalgal biomass into high-value products in the short term and into liquid biofuel in the long term requires a step of extracting compounds of interest. The residue resulting from this process possesses a significant energy value that can be utilized. Anaerobic digestion constitutes a highly efficient solution for the utilization of these residues

Depending on the location of the microalgae unit (industrial cluster, agricultural operation, etc.), various uses can revolve around microalgae cultivation and methanization. Certain compounds present in liquid and gaseous effluents, problematic for industries (CO₂, NO_x, N, P, etc.), could be absorbed by microalgae. Organic waste and effluents can be co-digested with microalgae.

The digestate resulting from methanization can also be utilized as a fertilizer or thermally in specific sectors such as incineration.

2. Methanisation process

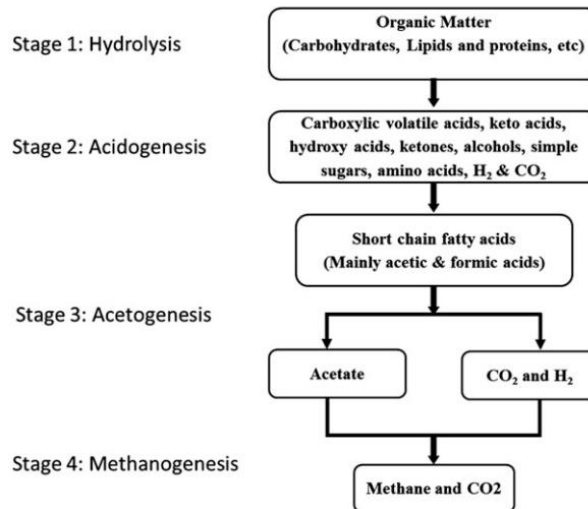


Fig. 1 Stages in Biomethanation Process

2.1 Hydrolysis

Hydrolysis is the first and main step in the biotransformation of various complex organic raw materials.

The hydrolysis step is essential because anaerobic bacteria can only use dissolved organic substances that can penetrate the cell wall.

Organic matter including lipids, carbohydrates, and proteins will be converted into amino acids, monosaccharides, and long-chain fatty acids (LCFA)

Extracellular enzymes participate in the hydrolysis of these complex organic materials.

Carbohydrate hydrolysis occurs within a few hours while protein and lipid hydrolysis takes several days, which makes hydrolysis a limiting step.

Since algae do not contain lignin, cellulose and hemicellulose (because it is difficult for anaerobic microorganisms to decompose them), algae are preferred as raw materials for biological methanization.

2.2 Acidogenesis

This is the second stage of the process in which two groups of microorganisms decompose soluble organic molecules such as monosaccharides and amino acids through facultative bacteria.

Alcohol, hydrogen, acetic acid, formic acid, and carbon dioxide are produced once this reaction is complete.

Few end products such as formic acid, acetic acid and hydrogen are directly consumed by methanogenic organisms during methanogenesis.

Compared to hydrolysis, the kinetics of this step is faster.

2.3 Acetogenesis

The third step of the process converts alcohol, butyric acid, propionic acid, valeric acid, etc. into hydrogen, carbon dioxide, acetic acid through acetogenic bacteria.

A very low partial pressure of hydrogen is required to form acetate from propionic acid, valeric acid, or butyric acid.

This shows that acetogenic bacteria live in close symbiosis with methanogenic bacteria.

The acetate generation and methanation processes operate simultaneously until excessive acidification occurs.

The overall response is:

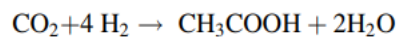


Table 3 shows the acetogenic reaction of different substrates.

Table 3. Examples of acetogenic reactions

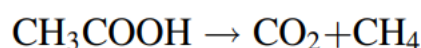
S. No.	Substrate	Reaction
1.	Propionic acid	$\text{CH}_3(\text{CH}_2)\text{COOH} + 2\text{H}_2\text{O} \rightarrow \text{CH}_3\text{COOH} + 3\text{H}_2$
2.	Butyric acid	$\text{CH}_3(\text{CH}_2)\text{COO}^- + 2\text{H}_2\text{O} \rightarrow 2\text{CH}_3\text{COO}^- + \text{H}^+ + 2\text{H}_2$
3.	Valeric acid	$\text{CH}_3(\text{CH}_2)_3\text{COOH} + 2\text{H}_2\text{O} \rightarrow \text{CH}_3\text{COO}^- + \text{CH}_3\text{CH}_2\text{COOH} + \text{H}^+ + 2\text{H}_2$
4.	Lactic acid	$\text{CH}_3\text{CHOHCOO}^- + 2\text{H}_2\text{O} \rightarrow \text{CH}_3\text{COO}^- + \text{HCO}_3^- + \text{H}^+ + 2\text{H}_2$
5.	Ethanol	$\text{CH}_3(\text{CH}_2)\text{OH} + \text{H}_2\text{O} \rightarrow \text{CH}_3\text{COOH} + 2\text{H}_2$

2.4 Methanogenesis

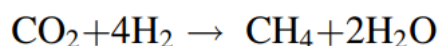
This stage is the final step that summarizes the methanogenesis process.

During this phase, compounds such as hydrogen, carbon dioxide and acetic acid are converted into methane and carbon dioxide through methanogens. The two types of bacteria involved are strict anaerobes that decompose acetic acid and are called acetoclastic methanogens, while those that decompose hydrogen are called hydrogenotrophic methanogens. The first pathway constitutes the main pathway of methane formation, i.e. the second pathway is about 70% and 30%.

The reaction to form methane from acetate is as follows:



while from hydrogen, the reaction is



This chemical reaction has a dual function in biological methanation because it not only produces methane but also removes hydrogen gas. High ammonia concentrations also limit the activity of methanogens in the digestion of algal substrates .

3. Different types of cultivation systems

There are two main cultivation systems used for the production of algal biomass: open ponds and closed photobioreactors.



Table 4. Different types of bioreactors.

Open ponds	<ul style="list-style-type: none"> • The average volumetric productivity of this system is 0.06 to 0.42 g/L per day. • The effectiveness of the system will depend on the composition of the pond and the type of algae growing. • Open reservoirs or naturally existing bodies of water (ponds, lakes, lagoons, etc.) are commonly known as open ponds and are easy to construct and operate. These ponds are kept shallow to allow solar radiation to penetrate easily. Water and nutrients circulate continuously in the crop. • Pond productivity is measured by calculating the biomass produced per unit area per day. • The area of the round pond and the road can be about 1 hectare and the area of the large pond can be about 200 ha.
Unmixed open ponds	<ul style="list-style-type: none"> • This system has no control over the factors involved in the reproductive process.
Circular pond	<ul style="list-style-type: none"> • This system stands out as the first design to be used commercially for growing algae. The main disadvantage of this system is that its scale is limited to a radius of about 1000 m² • Voltages in this range will cause the main rotary mixer to become uncontrollable.
Open Raceway Pond	<ul style="list-style-type: none"> • In the early 1950s, Oswald and his colleagues introduced this system, known as the HRAP (high rate algae pond). This system is mainly used to treat wastewater by maintaining a symbiotic relationship between aerobic bacteria and algae. • With the help of paddles, the circulation of broth and nutrients is carried out in loop-shaped channels. • The pond is made of concrete, PVC or clay and is about 0.2 to 0.5 m deep, allowing sunlight to penetrate deeply. • Although these systems are well developed, they still present problems with infestation and unwanted bird species.
Closed photobioreactors	<ul style="list-style-type: none"> • Photobioreactor (PBR) system yields range from 0.02 g/L to 3.22 g/L/day • are used to grow algae under controlled conditions. • The effectiveness of this system can be evaluated taking into account the fact that it does not allow exposure of the birds to the external environment.
Tubular PBR	<ul style="list-style-type: none"> • Tubular PBR is made by installing linear plastic or glass tubes. This

	<p>configuration can be easily manipulated to form PBR-like structures such as spiral tubes and fences.</p> <ul style="list-style-type: none"> • The ability of this composition to expose crops to maximum sunlight makes them suitable for use in outdoor cultivation. • <u>The problem with tubular PBRs is algae build-up at the bottom of the tube.</u> • However, sinking can be avoided by using lifting propellers to maintain a highly turbulent flow. • Tubular PBR is the largest size, with an area of up to 750 m³.
Flat-Plate PBR	<ul style="list-style-type: none"> • Made using a thin rectangular container made of transparent material. • These cylinders are tilted at a certain angle to block as much sunlight as possible. • The density of photoautotrophic cells grown in this PBR is high (>80 g/L). • They are more suitable for cultivation because they have less accumulation of dissolved oxygen and photosynthesis is more efficient. • However, there are issues such as temperature control and algae adhesion to the reactor walls.
Column Photobioreactors	<ul style="list-style-type: none"> • This configuration provides the best mixing, the best controlled culture conditions, and the highest volumetric mass transfer rate. • It is economical and relatively easy to install. • The PBR columns are vented at the base and the transparent walls maximize light exposure. • You can also turn on the interior lights.
Continuously Stirred Tank Reactors	<ul style="list-style-type: none"> • The shape of the tank is wide and hollow, allowing it to work both inside and outside a closed cylindrical channel. • The risk of contamination of the culture is very low. • From above, mixing and lighting fixtures are included. Drainage systems and gas injectors are located in the lower and middle sections. • Constant turbulence promotes algae growth and prevents contamination of the culture. • Compared to open pond systems, algae cultures in PBR are effectively protected from all types of pollution and losses due to low evaporation. • Culture parameters (nutrients, temperature, pH, etc.) can be effectively controlled.

It also can be find the Hybrid Production System , which is a two-stage culture method that uses both open ponds and PBR for different growth stages. The first culture step is completed in a photobioreactor where continuous cell growth takes place in a pollution-free environment under controlled conditions. The second stage of cultivation takes place in open ponds and is designed to expose the cultures to environmental and nutritional stresses. This improves the production of necessary lipid products.

Table 5: Comparison of two of the microalgae production systems

TECHNOLOGY	 POND	 PHOTOBIOREACTOR
PRINCIPLE	<p>The ponds are large open basins, with the majority being of the 'racecourse' type.</p> <p>The circulation and ventilation of the crop are carried out mechanically.</p>	<p>The photobioreactors are closed systems where the conditions of mixing and material transfer are optimized</p>
ALGAL BIOMASS CONCENTRATION (DM=dry matter)	0,1 – 0,5 g DM/L	2 – 8 g DM/L
AVERAGE SURFACE YIELDS	10 – 50 T/ha/yr	30 – 150 T/ha/yr
CURRENT PRODUCTIVITY	27 T DM/ha/yr	76 T DM/ha/yr
PRODUCTIVITY ESTIMATED 2050	56 T DM/ha/yr	116 T DM/ha/yr
COSTS	10 - 40 €/m ²	100 - 300 €/m ²
ADVANTAGES	<ul style="list-style-type: none"> → Easy to build and operate → Low cost → System suitable for mass production of microalgae → Low energy consumption 	<ul style="list-style-type: none"> → High surface productivity → Better control of cultivation conditions → Optimization of material and light transfers
DISADVANTAGES	<ul style="list-style-type: none"> → High risk of contamination → Low surface productivity → Limited optimization of material transfers → Evaporation 	<ul style="list-style-type: none"> → High cost → Accumulation of O₂ in the reactor → Temperature regulation required → Generally high energy consumption

Currently, the most efficient ways to grow algae are ponds and pipe-type PBRs. Open ponds are a much cheaper way to grow algae. Installation and maintenance are also easier.

Open ponds require less energy compared to PBR. However, open ponds are less efficient than PBR. Soiling, evaporative losses, temperature fluctuations, poor mixing, limited light, etc. are some of the disadvantages associated with open pond systems.

4. Operating Parameters

4.1 Temperature

At a temperature range of 30 to 35°C, thermophilic methane-producing bacteria are highly active, and at a temperature range of 50 to 60°C, thermophilic methane-producing bacteria are highly active. The most favorable temperature for biogas production is 35°C.

However, methane production can occur over a wide temperature range. Temperatures below 32°C may increase the volatile acid/alkali ratio. When temperatures exceed 32°C, the rate of decomposition of volatile solids and methane production is higher.

However, the formation of recalcitrant and/or inhibitory complexes reduces biomass digestibility when the temperature rises above 180° C .

To improve the energy demand and increase the profitability of the process, is required low temperature pretreatment as anaerobic digestion of microalgae.

Table 6. Effect of low temperature pretreatment of microalgae

References	Microalgae	Pretreatment conditions	Anaerobic digestion conditions	Result
Chen and Oswald (1998)	Microalgal biomass grown in wastewater	100°C, 8 h	Batch	CH ₄ production increased by 33% ^a
Gonzalez-Fernández et al. (2012a)	<i>Scenedesmus</i> biomass	90°C, 3 h	Batch 35°C	CH ₄ production increased by 220% ^a
Gonzalez-Fernández et al. (2012b)	<i>Scenedesmus</i> biomass	80°C, 25 min	Batch 35°C	CH ₄ production increased by 57% ^a
Alzate et al. (2012)	<i>Scenedesmus</i> and <i>Chlamydomonas</i> biomass	55°C, 12 and 24 h	Batch 35°C	CH ₄ production decreased by 4–8% ^a
Alzate et al. (2012)	<i>Acutodesmus obliquus</i> and <i>Oocystis</i> sp. Biomass	55°C, 12 and 24 h	Batch 35°C	CH ₄ production decreased by 3–13% ^a
Alzate et al. (2012)	<i>Microspora</i> biomass	55°C, 12 and 24 h	Batch 35°C	CH ₄ production increased by 4–5% ^a

^aCompared to control

4.2 pH

Anaerobic microorganisms can be divided into acid-producing microorganisms and methanogenic microorganisms.

The optimal pH ranges for acid-producing and methanogenic bacteria are 5.5-6.5 and 7.8-8.2, respectively.

The optimum pH range for both crops ranges from 6.8 to 7.4. Methanogenesis is the most important step, so the pH should be close to neutral.

4.3 Hydraulic Retention

HRT refers to the time spent by the wastewater or sludge in the digester.

This is the main parameter of the biomethane formation process. The HRT must be high enough to stimulate a dynamic population of microorganisms, especially methanogens, functioning in the reactor.

Figure 2 illustrates the efficiency of soluble COD removal at different temperatures in anaerobic methane digesters using continuous reactions.

"sCOD" likely refers to soluble Chemical Oxygen Demand. Chemical Oxygen Demand (COD) is a measure of the amount of oxygen required to chemically oxidize both organic and inorganic substances in water. "sCOD" specifies that the measurement is focused on the soluble fraction

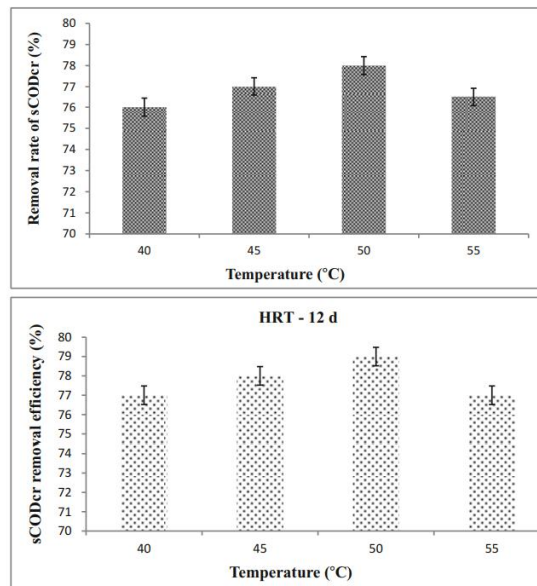


Fig. 2 Temperature-dependent sCOD removal efficiencies of different anaerobic methane digesters by continuous reaction (a) HRT=10 d (b) HRT=12d (Source: Kim et al. 2006)

4.4 Loading Rate

The optimal loading rate depends on the composition and biological composition of the algae, which allows efficient conversion of organic matter.

5. Large-Scale Production of Algal Biomass: Photobioreactors.

Photobioreactors provide a controlled environment for cultivating microalgae, allowing researchers and industries to optimize growth conditions for specific species. There are different species which can grow inside of them, one of the most common used are:

- *Chlorella sorokiniana*: is a popular choice for photobioreactors due to its rapid growth and high lipid content. The controlled conditions in photobioreactors allow for precise regulation of factors such as light intensity, temperature, and nutrient availability, which can enhance *Chlorella*'s productivity.
- *Tetraselmis Suecica*: is another genus of microalgae that can be grown in photobioreactors. Like *Chlorella*, *Tetraselmis* is often considered for various applications, including biofuel production. The ability to customize environmental parameters in photobioreactors can support the cultivation of *Tetraselmis* under optimized conditions.

The photobioreactor design that will be explained it is based on the cultivation of *Chlorella sorokiniana*, which is one of the most popular algae species to be cultivated

4.1. Reactor design

According to literature, the main parameters that need to be controlled for the successful cultivation of ***Chlorella sorokiniana*** in a photobioreactor include:

1. Light intensity: The light intensity should be measured and maintained at a specific level using a PAR meter. In the study, the light intensity was set at $800 \mu\text{mol photons m}^{-2} \text{ s}^{-1}$.
2. Temperature: The temperature inside the vessel should be controlled using an automatic cooling water control valve and a heating blanket. In the study, the temperature was maintained at 30°C .
6. Aeration: Aeration should be provided using a compressor and a sterile filter with a PTFE membrane. In the study, the aeration rate was set at 1.3 L min^{-1} .
7. CO₂ supply: The CO₂ supply should be measured and controlled using a mass flow controller/meter and a gas analyzer. In the study, the final supply of 2.0% CO₂ was reached.
8. Foam suppression: A sterile foam suppressant should be added to prevent foaming. In the study, sterile 2% foam suppressant (Antifoam B, Sigma-Aldrich, St. Louis, MO, USA) was used.
9. pH control: The pH should be measured and maintained at a specific level using an electrode and automatic addition of sterile solutions. In the study, the pH was maintained at 7.0 ± 0.05 with the automatic addition of a sterile 2% HCl or 8% NaOH solution.

The parameters should be adjusted according to the specific needs of the microalgal strain being cultivated and the experimental conditions. The machine used to determine these parameters in the study was a PAR meter for light intensity, a temperature control valve and

heating blanket for temperature, a compressor and gas analyzer for CO₂ supply, and an electrode for pH measurement.

4.2. Growth efficiency

The same study found several key findings regarding the growth efficiency of *Chlorella sorokiniana* in synthetic media and unsterilized domestic wastewater:

1. In synthetic media:

- Increasing the NH₄⁺ (N) concentration to 360 mg/L and adding extra PO₄⁻³ (P) and SO₄⁻² (S) contributed to an increase in the total biomass levels during the cultivation of *C. sorokiniana* in synthetic media .
- Under these conditions, the maximum concentrations of chlorophylls and carotenoids were 180 ± 7.5 and 26 ± 1.4 mg/L, respectively.

2. In unsterilized domestic wastewater:

- Only one type of wastewater contributed to the productive growth of *C. sorokiniana*, but all wastewaters stimulated an increased accumulation of protein.
- When growing in optimal unsterilized wastewater, *C. sorokiniana* showed a maximum specific growth rate of 0.73 /day, a biomass productivity of 0.21 g/L/day, and 100% NH₄⁺ (N) removal.

These results demonstrate the adaptability of *Chlorella sorokiniana* to changes in the composition of the growth medium and its ability to accumulate high levels of protein in systems with poor-quality water.

4.3. Reactor design

In the same study it was also provided the step-by-step instructions for building a reactor to cultivate *Chlorella sorokiniana*:

1. Inoculum Preparation:

- a. Isolate *Chlorella sorokiniana* from a water reservoir and maintain it on a standard growth medium.
- b. Add antibiotics to the growth medium to reduce the risk of bacterial contamination.
- c. Cultivate the isolated *Chlorella sorokiniana* in sterile conditions using a shaker at specific temperature and light conditions.

2. Cultivation Vessel:

- a. Use a 6.8 L glass culture vessel with a working volume of 5.0 L.
- b. Install LED lamps as a light source, ensuring even distribution vertically around the vessel.
- c. Measure and maintain the light intensity using a PAR (Photosynthetically Active Radiation) meter.
- d. Control the temperature inside the vessel using an automatic cooling water control valve and a heating blanket.

- e. Provide aeration using a compressor and a sterile filter with a PTFE membrane.
 - f. Measure and control the CO₂ supply using a mass flow controller/meter and a gas analyzer .
 - g. Add a sterile foam suppressant to prevent foaming.
 - h. Measure and maintain the pH using an electrode and automatic addition of sterile solutions.
3. Experimental Conditions:
- a. Optimize the parameters for the photoautotrophic growth of *Chlorella sorokiniana*.
 - b. Cultivate the microalgae in various modes, including photoautotrophic growth regimens.
 - c. Adjust the nutrient medium to achieve specific concentrations of nitrogen, phosphorus, and sulfur.
 - d. Monitor the growth characteristics, biomass yield, and biochemical composition of the cells under different experimental conditions.

6. Large-Scale Production of Algal Biomass: Raceway Ponds.

Raceway ponds are often used for cultivating microalgae, and various species can be employed depending on the specific goals of the cultivation. Some common microalgae species grown in raceway ponds for water treatment and biodiesel production include:

- **Chlorella:** Chlorella is a genus of green algae known for its rapid growth and high lipid content, making it suitable for biodiesel production.
- **Scenedesmus:** Scenedesmus is another green algae genus with potential for biofuel production. It is known for its adaptability and ability to thrive in various environments.
- **Nannochloropsis:** Nannochloropsis is a genus of microalgae that is often considered for biodiesel production due to its high lipid content.

The choice of algae species depends on factors such as lipid content, growth rate, adaptability to environmental conditions, and the specific requirements of the intended application, whether it's water treatment, biofuel production, or other purposes.

6.1. Reactor design

To design a reactor for large-scale production of algal biomass in raceway ponds requires careful consideration of culture flow, mixing efficiency, carbon dioxide supply, and energy consumption, among other factors. Managing pH, carbon supply, and mixing efficiency are critical for achieving high biomass productivity while optimizing energy efficiency.

- **Light Intensity:** Light intensity is a critical factor in algal growth and biomass productivity. The specific growth rate of a microalga in a pond varies with depth because the light intensity declines with depth. The literature provides equations for estimating local

irradiance at different depths in the pond, which can help to optimize light exposure for maximum biomass productivity.

- Carbon Dioxide Supply: A supply of carbon dioxide is necessary to avert carbon limitation and attain high biomass productivity. Carbon dioxide can be effectively supplied in response to a pH signal, and microporous gas diffusers are used to provide carbon dioxide in the form of fine bubbles.
- Mixing Efficiency: Efficient mixing is crucial to prevent the accumulation of dissolved oxygen to far above the air saturation concentration, which can adversely affect productivity. The literature notes that poor mixing can result in inadequate oxygen removal during periods of rapid photosynthesis and an accumulation of dissolved oxygen, affecting productivity.
- Temperature: Temperature is an important factor in algal growth and biomass productivity. The article notes that up to 25% of the biomass produced by the end of a daylight period may be consumed during the following night through respiration, and the magnitude of this respiratory loss depends on the irradiance level during growth, the daytime temperature of growth, and the temperature during the night.
- Culture Flow: The flow in a raceway conduit needs to be turbulent to keep the cells in suspension, enhance vertical mixing, prevent thermal stratification, and facilitate oxygen removal generated by photosynthesis.

6.2. Reactor sizing

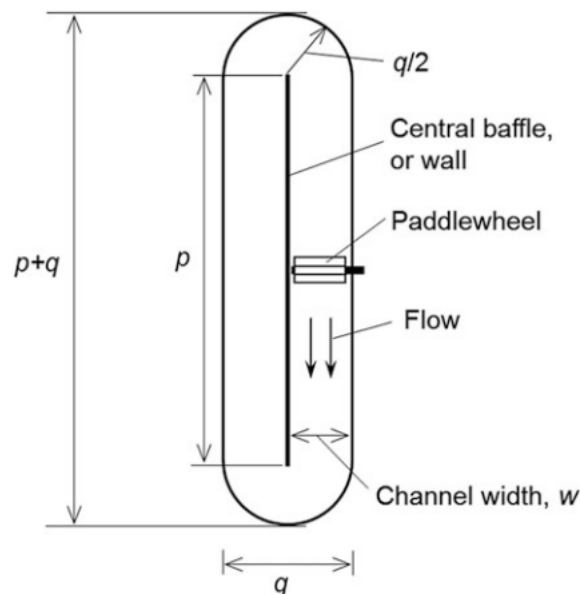


Figure 3. Raceway pond schema

A raceway pond is a closed-loop flow channel with a typical culture depth of about 0.25–0.30 m (Fig. 1). A paddlewheel continuously mixes and circulates the algal broth in the channel (Fig. 1). An algal biomass production facility will typically have many ponds. The surface area of a single pond does not usually exceed 0.5 ha, but can be larger.

Raceways generally have a flat bottom and vertical walls. If the thickness of the central dividing wall (Fig. 1) is neglected, the surface area A of a raceway such as shown in Fig. 1, can be estimated using the following equation:

$$A = \frac{\pi * q^2}{4} + p * q$$

where p and q are the length and width of the pond, respectively. The p/q ratio can be 10 or larger. If this ratio is too small, the flow in the straight parts of the raceway channel begins to be affected by the disturbances caused by the bends at the ends of the channel. The working volume V_L is related to the surface area and the depth h of the culture broth, as follows:

$$V = A * h$$

where h is the depth of the culture broth.

The surface-to-volume ratio is always $1/h$. A lower depth increases the surface-to-volume ratio and this improves light penetration, but in a large pond the depth cannot be much less than 0.25 m. A compacted earth construction lined with a 1–2 mm thick plastic membrane may be used for the pond, but this relatively cheap setup is uncommon for biomass production. Ponds used to produce high-value biomass are often made of concrete block walls and dividers lined with a plastic membrane to prevent seepage. Membranes made of ultraviolet resistant polyvinyl chloride (PVC), polyethylene, and polypropylene are generally used and can last for up to 20 years

The next figure shows the irradiance variation with depth in a 0.3 m deep raceway at various concentrations of the algal biomass in the broth. The local irradiance profiles were calculated for an alga with a K_a value of $2.632 \mu E/m^2/s$ and an incident irradiance of $2000 \mu E/m^2/s$ at the surface of the raceway.

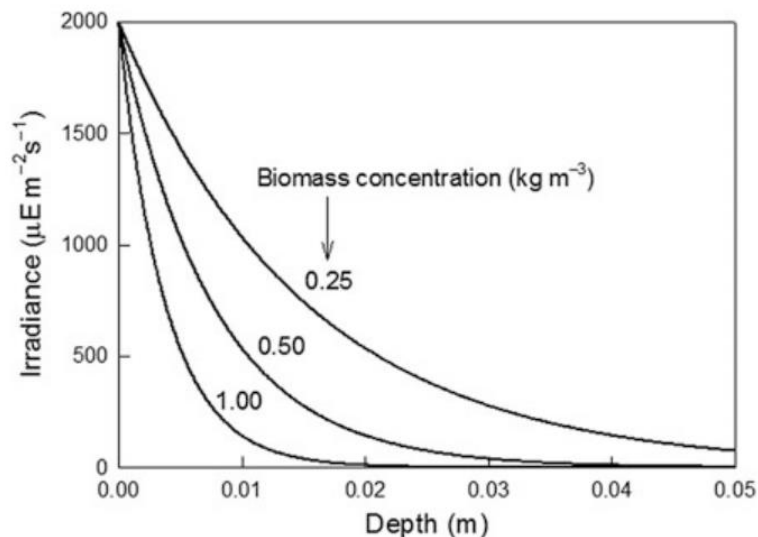


Fig 4. Irradiance variation with depth in a 0.3 m deep raceway.

6.3. Optimizing Reactor Design and Operation:

- Paddle Wheel: Paddlewheels are generally believed to be the most effective and inexpensive means of producing flow in raceways. The design and configuration of paddlewheels can impact mixing efficiency and power consumption.
- Energy Consumption: Computer simulations of pond fluid dynamics have resulted in design recommendations for minimizing energy consumption while achieving sufficient mixing to prevent sedimentation and dead zones. Lowering the channel flow velocity at night can greatly reduce power consumption.

6.4. Possible contamination culture.

The literature discusses the issue of culture contamination in the context of large-scale production of algal biomass in raceway ponds. It highlights several potential sources of contamination and the challenges associated with managing and preventing contamination. Here are the key points regarding culture contamination:

1. Contamination Sources: open ponds, such as raceway ponds, are exposed to various sources of contamination, including rain, dust, and other debris. Additionally, contamination issues may arise from infestations of predators feeding on algae, viral infections, and contamination by unwanted microalgae, fungi, and bacteria.
2. Contamination Management: It is important to manage practices which reduce the frequency of culture contamination and failure. Filtration of water may help reduce the frequency of certain types of infestations, but on the other hand, that filtration is expensive and may not prevent all forms of contamination, such as viral infections.
3. Predator Control: Predator control in raceways is potentially possible. It references studies that indicate the potential for controlling predators in raceway ponds, highlighting the importance of addressing this aspect of contamination management.
4. Bacterial Contamination: contamination with heterotrophic bacteria is inevitable and not necessarily harmful, but may necessitate the implementation of specific controls depending on the final application of the alga being grown.

6.5. Advantages and Disadvantages of Raceway Ponds:

- Advantages: Raceway ponds are cost-effective and relatively simple to construct and operate. They offer a large surface area for light exposure and can be used for the production of various algal species.
- Disadvantages: Challenges include the need for efficient mixing to prevent sedimentation and dead zones, as well as the management of pH and carbon supply to support high biomass productivity.

7. Bio fuel production in Raceway Ponds real example.

There is a project which talks about pilot systems operated at a full-scale raceway wastewater treatment plant in Delhi, California, with the aim of producing biofuel with microalgae. It involves the use of pilot-scale raceway ponds to investigate the optimization of biomass production and treatment.

It focuses on the production of biofuel from algal biomass using hydrothermal liquefaction (HTL). The goal is to prepare for the scale-up of 2500 gal/ac-yr of biofuel intermediates via hydrothermal liquefaction (HTL) of microalgae (*Chlorella Sorokiniana*) grown at 7 acres of existing raceways at a wastewater treatment facility.

In terms of characterizing the reactor and designing it, the parameters taken into account include:

- Productivity
- Hydraulics
- Nutrient recycling
- Conversion of algae carbon to fuel
- Achieving at least 25% lower cost than conventional wastewater treatment

The characteristics of the biofuel obtained include a yield of 3,400 gal/acre-yr and the cost range of \$4 - \$9 per gallon for the biofuel intermediates.

It also achieved significant numbers in terms of biomass yield and biofuel production. Here are the key accomplishments:

1. Biomass Yield: The project achieved an annual average biomass yield of 33 grams per square meter per day. This high biomass yield was obtained through the cultivation of microalgae using treated wastewater as a nutrient source.
2. Biofuel Intermediates: The project obtained biofuel intermediates through the hydrothermal liquefaction (HTL) process. The HTL conversion rate achieved was 0.35 grams of oil per gram of algae. This process resulted in the production of 4,100 gallons of biofuel intermediates per acre per year.
3. Nutrient Recycling: The project successfully demonstrated the recycling of nutrients from HTL wastewater without inhibition at dilution factors that meet nitrogen requirements.

Additionally, the project aims to demonstrate and optimize the conversion of wet algal biomass to fuel intermediates, which will be characterized by mass (C, N, P, S, H, and O).

8. Bibliography:

Comité de pilotage. (2013). *BIOMETHANE DE MICROALGUES: ÉVALUATION DU POTENTIEL DE PRODUCTION EN FRANCE AUX HORIZONS 2020 ET 2050: RAPPORT FINAL – FEVRIER 2013*. CRIGEN de GDF SUEZ. https://france-biomethane.fr/wp-content/uploads/2016/03/2013-Etude_du_potentiel_biomethane_microalgue.pdf

Gupta, S.K., Malik, A., and Bux, F. (eds) 2017, *Algal Biofuels: Recent Advances and Future Prospects*, Springer International Publishing, DOI: 10.1007/978-3-319-51010-1

Majid, M., Shafqat, S., Inam, H., Hashmi, U., & Kazi, A.G. (2014). Production of Algal Biomass. En K.R. Hakeem et al. (eds.), *Biomass and Bioenergy: Processing and Properties*, p. 207. Springer International Publishing. DOI: 10.1007/978-3-319-07641-6_13 https://www.researchgate.net/publication/266389121_Production_of_Algal_Biomass

Chisti, Y. (2016). Large-Scale Production of Algal Biomass: Raceway Ponds. En *Algae Biotechnology* (p. número de página del capítulo). DOI: 10.1007/978-3-319-12334-9_2 https://www.researchgate.net/publication/309698148_Large-Scale_Production_of_Algal_Biomass_Raceway_Ponds

Pruvost, J., Cornet, J-F., & Pilon, L. (2016). Large-Scale Production of Algal Biomass: Photobioreactors. En *Algae Biotechnology*. DOI: 10.1007/978-3-319-12334-9_3 https://www.researchgate.net/publication/309698143_Large-Scale_Production_of_Algal_Biomass_Photobioreactors

Pereira, H., Páramo, J., Silva, J., Marques, A., Barros, A., Maurício, D., Santos, T., Schulze, P., Barros, R., Gouveia, L., Barreira, L., & Varela, J. (2018). "Scale-up and large-scale production of *Tetraselmis* sp. CTP4 (Chlorophyta) for CO₂ mitigation: from an agar plate to 100-m³ industrial photobioreactors." *Scientific Reports*, 1. <https://doi.org/10.1038/s41598-018-23340-3>

Spierling, R., Crowe, B., Adler, N., Poole, K., Hutton, M., Huesemann, M., Lane, T., Poorey, K., Anderson, D., Benemann, J., & Lundquist, T. (2018). *Scale-Up of Algal Biofuel Production Using Waste Nutrients* (Informe WBS: 9.5.1.5, Número de premio: DE-EE0006317). California Polytechnic State University. San Luis Obispo, CA. <https://www.osti.gov/servlets/purl/1475450>

Serri, N. A., et al. "Preliminary study on the growth of *Tetraselmis suecica* in centred-light photobioreactor (CLPBR)." *IOP Conf. Ser.: Mater. Sci. Eng.* 716 (2020): 012008. <https://iopscience.iop.org/article/10.1088/1757-899X/716/1/012008>

Bulynina, S. S.; Ziganshina, E. E.; Ziganshin, A. M. (2023). "Growth Efficiency of *Chlorella sorokiniana* in Synthetic Media and Unsterilized Domestic Wastewater." *BioTech*, 12(3), 53. <https://doi.org/10.3390/biotech12030053>