Algae-Based ph Indicators

How algae pigments can be used as pH indicators and applied in an environmental and commercial setting

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Section 1:

Brainstorming ideas:

-Algae Biofuels

-Algae pH indicators

-Life on mars

-Ocean pollution

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-Social media's effects on dopamine

Topic: Algae based pH indicators

Important questions:

-Is it possible to use algae pigments as pH indicators?

-Why are algae based pH indicators important?

-How do algae based pH indicators work?

-How can algae based pH indicators be applied in a commercial setting?

-How can algae based pH indicators be applied in an environmental setting

-What algae type is the best pH indicator?

Plan of Approach:

-Create and experiment where I can extract and test algae pigments

-Gain a better understanding of the importance of algae in our environment and why algae pigment will be a very useful and important ph indicator

-Do a lot of background research about algae pigments

-Create a visually appealing tri fold and organise my logbook

Basic research:

-Why do algae pigments differ between species?

-Why do algae pigments even change at different pH levels?

-How to extract algae pigments from algae powder?

-Why are natural ph indicators important for the environment

-What advantages do natural pH indicators provide

Brief Project Description:

For my project I will be testing the efficacy of algae pigments as pH indicators to determine if they would be useful environmental and commercial applications.

pH=potential of hydrogen

Section 2:

Research Question:

Can algae pigments be used as natural pH indicators, and how do they compare to traditional pH indicators like litmus paper and PH paper?

What's the purpose?

To determine if algae pigments can be used as natural pH indicators, how they would compare to traditional pH indicators like litmus paper and pH paper, and if there are any potential environmental or commercial applications.

Why I chose this topic:

This experiment combines my two favourite fields of science, chemistry and biology. This topic is environmentally friendly, sustainable and scientifically valuable. Natural Ph indicators such as algae are biodegradable and non-toxic, which makes them safer than traditional ph indicators like pH paper and litmus paper. They also have many real world applications like ecologically friendly water testing, food safety, and medicine. Algae pH indicators can be used in food safety to test if food is still fresh and edible. Algae can also help test ocean acidity levels, linking to climate science.

Some background research:

About Algae: Algae is a very interesting plant. It's so simple yet it's one of the most ecologically important species on the planet. Nearly all species of marine life rely on algae to stay alive, from shrimp to whales. Algae is one of the most critical parts at the base of the marine life food chain. Algae is a “blanket term” that describes a group of marine organisms that all have something in common, they are autotrophic. That means that algae can take very simple substances, like carbon dioxide, light, and bacteria and transform them into food. Algae are phototrophs so they utilise photosynthesis, like many other terrestrial plants. Because of this they can easily provide themselves with necessary nutrients by harnessing the power of natural light and Co2 while

providing us with one of the most important building blocks of life, oxygen.

Many forms of algae contain photosynthetic pigments. They can come in many different colours, ie Chlorophylls (green, absorbs red and blue light), Carotenoids (Yellow, Orange, Red, helps with light absorption and photoprotection. Fucoxanthin, a carotenoid pigment, gives it a brown-ish hue by absorbing blue-green light), and Phycobilins (Red, Blue, Purple, absorbs glue green light to give it a red-ish hue). Certain algae species such as chlorophylls, carotenoids, and phycobilins contain pigments that can change color in response to different pH levels because of adaptive advantages (to optimise photosynthesis), and chemical structure changes (shift between different molecular states at different ph levels that change how they absorb and reflect light). Because of this, algae can be used as a natural pH indicator that is abundant and reliable, much like beets. It is also very useful because it can be used to test out the health of an environment, contributing to natural restoration.

Commercial Applications:

Using algae pigments is important and can be used in many different ways. Determining if algae pigments can be used as reliable ph indicators is necessary because algae pH indicators are natural and edible (algae is actually superfood!) they are completely safe to consume and add to different food products. This means that algae pigments could be added to foods like milk, meat, cheese, and various other food products that are harmful when they expire. Generally when food tends to decompose (rot) fungi and bacteria produce acidic (or alkaline) byproducts, shifting the pH. A change in pH could also be caused by fermentation (especially in dairy and fruit) as they spoil, producing acids that will lower the pH. Two other factors that could possibly change the pH of different food products are oxidation (which could cause fats and proteins breaking down altering their acidity or alkalinity), and protein breakdown (meats and seafood have proteins that breakdown into ammonia and amines making the pH more alkaline). The algae pH indicators could possibly detect that and change colour. So if you see changes in colour of food that has been covered in algae pigment, you might want to check the expiry date before you eat it!

Also, algae pigment pH indicators can be used in cosmetology. They are completely safe to put on your skin. Because of that, they could be added to various make-up and skincare products to again detect when they are expired because of the changes in pH caused by the breakdown of ingredients, microbial growth, and oxidation. Over long periods of time, preservatives, pH stabilizers, and emulsifiers in makeup degrade, which leads to shifts in the pH. Bacteria and fungus thrive in expired makeup products and produce acidic or basic byproducts that cause a shift in the PH. Finally, prolonged exposure to air causes chemical reactions,

especially in products with oils, that make the products more acidic or alkaline. The addition of algae pigment into these products could save a lot of people from skin irritation and infections by spotting a different colour in their product.

Algae Based ph indicators are also very affordable compared to traditional pH indicators and can be used for low-cost field based testing. They can be used for on-site water testing remote areas because of its portability in areas where traditional laboratory equipment is unavailable. This could benefit developing regions or general citizen science projects and monitoring local water sources like reservoirs, rivers, and lakes.

Finally, algae pH indicators are very environmentally friendly and sustainable. They are biodegradable, renewable, and safer for the researchers and ecosystems. They are very useful for environmental monitoring because many algae species are highly sensitive to pH changes that are generally caused by different forms of water pollution (acid rain, industrial waste, etc…). Using algae Pigment pH indicators can help scientists detect shifts in river, lake, pond, and ocean pH before even more severe ecological damage could occur. Ocean acidification is a huge problem today. It refers to the lowering of the pH in the ocean caused by the huge addition of Co2 in our atmosphere. The ocean absorbs about 30% of the total amount of Co2 released into the atmosphere. When the levels of Co2 in the atmosphere, the levels of Co2 in the ocean increase. They are in direct correlation with each other. The changes in the ocean pH severely impact marine life and how predators can detect prey and visa versa. They also affect calcifying organisms such as coral reefs, the most biodiverse places on earth. With that problem, the entire marine food web is at risk. Algae pigments from red-algae (phycobilins), and chlorophyll-based pigments change colour in response to acidification, making them vital for tracking long-term changes in pH. Algae pH indicators can also detect various different types of industrial and agricultural pollution like pesticides, fertilizers, and chemical spills. These severely alter the water's pH and lead to harmful algae blooms (HABs) and marine life deaths.

What People are doing with algae pigments:

Currently, scientists are using algae as biological indicators to monitor aquatic ecosystems. The government and the public are becoming quite concerned about maintaining the health of aquatic resources. Algae are ideal for water quality assessment because of fast reproduction rates, short lifespans, and high effectiveness. This makes them very valuable for assessing short-term environmental impacts. Periphyton (a community of organisms, including algae, fungi, bacteria, etc…) are one of the most important forms of algae that are associated with substratum in aquatic environments. They have been highly used as a tool for monitoring water quality.

Why this project matters:

-Algae is food safe, so it can be used in different food and cosmetic products! When these products expire, they can change in Ph. When food decomposes (rots), bacteria and fungi breakdown proteins, fats, and carbohydrates. The process often produces acidic or alkaline byproducts, it depends on the type of microbes that are present.

-If algae pigments can be proven as a reliable Ph tester, they could be used as Ph tests for scientists that are working remotely. They won't have to worry about accidentally bringing harmful chemicals into the environment or leaving anything dangerous behind.

-Algae pigments are also a key aspect of solving ocean acidification, with the use of these pigments, we could match the different colours of algae to the different Ph levels to see if the ocean is dangerously acidic.

Section 3:

Hypothesis:

Algae species such as carotenoids, phycobilins and chlorophylls contain special pigments that can change color in response to different pH levels. If these algae pigments can visibly change color across a pH scale, they could be used as natural pH indicators.

Materials and where they were sourced from:

- Different types of algae (spirulina, chlorella, red algae, brown algae) (Amazon)
- Isopropyl alcohol for pigment extraction (Pharmacy)
- Mortar and pestle (or blender) (kitchen-ware stores)
- coffee filters (Grocery store)
- household acids/bases (lemon juice, vinegar, baking soda, ammonia) (Various Grocery stores)
- Pipettes (Amazon)
- litmus papers and ph papers (Amazon)
- White ceramic plate and test tubes (used for the color comparisons) (Amazon)

Safety/waste disposal: Once I've completed the experiment, the algae that was used to test environmentally friendly substances (such as lemon juice) will be disposed of in the compost bin provided by the city. The algae that was used to test potentially environmentally harmful substances (like ammonia) will be disposed of in the garbage along with the pH and litmus

papers. The liquids that I will use will be neutralized (if needed) and disposed of in the sink. And the equipment I will use (ceramic dishes, pipettes) will be washed in the sink.

Procedure:

Step 1: Extract Pigments from Algae

1. Prepare the mixture:

- Add 2 grams of powdered algae to a small beaker.
- Pour in 10 mL of water and mix well.

2. Extract the pigments:

- Add 20 mL of isopropyl alcohol (at least 70%) to the algae mixture.
- Stir the solution thoroughly to ensure the pigments dissolve.

3. Allow the mixture to sit:

- Let the solution sit for 20–30 minutes, stirring occasionally.

4. Filter the extract:

- Pour the mixture through a coffee filter into a clean container to separate the liquid pigment extract from the solid algae residue.

Step 2: Test Pigment Reaction to pH Changes

1. Set up test wells:

- Using a ceramic plate with wells, distribute 1 mL of the pigment extract into each well.

2. Prepare pH solutions:

- Acidic Solution: Add 1 mL of vinegar to a well, one mL of tomato juice to a well, and add one mL of lemon Juice to a well.
- Neutral Solution: Add 1 mL of Milk to a well.
- Basic Solution: Add 1 mL of bleach to a well, and add one mL of soap to a well.

3. Observe reactions:

- Stir each mixture gently and observe any immediate color changes.
- Allow the solutions to sit for 5 minutes and note any further changes.

Step 3: Measure and Record Changes

1. Confirm pH levels:

- Use pH strips or litmus paper to verify the pH of each solution.

2. Compare color changes:

- Record the color of each algae extract at different pH levels in your table .

Section 4:

What results are expected:

- Chlorella (Green algae with chlorophyll-based extracts) may turn yellowish-brown in acidic environments.
- Spirulina (phycocyanin-based extracts) will probably shift from blue at neutral pH to reddish-yellow in acidic environments.

Why:

Due to the high amounts of chlorophyll present in both of the algae types, which happens to break down into Pheophytin, which turns the pigments any red, olive green, and brown.

Controlled Variables:

1. Types of algae used
2. Types of pH solutions used
3. Method of extraction
4. Cleanliness of workstation
5. Ph indicators used (Litmus papers, Ph papers)

Manipulated Variables:

1. The colour of the Algae Pigment

Measured/Responding Variables:

- The Colour of the algae pigment after its been added to the solutions of different pHs
- The Colour stability of the pigment in the solution

Experiment modifications:

- Increased the amount of time the algae pigment sat in isopropyl alcohol from 20 minutes to 25 minutes.
- Used liquid hand soap instead of baking soda as the basic solution.
- Had to research the Ph of clorox bleach because it bleached the Ph papers, litmus papers, and algaes so an accurate reading was not possible.

Results:

-The spirulina (Blue-green) algae shifted into reds and oranges for acids, yellowish greens for neutral substances, and darker greens for bases. The bleach bleached the colour of the algae and it turned white. The Chlorella (green) algae turned yellow-ish greens for acids (pH 2-6) pea green for neutrals (pH 7), and a similar, darker pea green for bases (pH 8). The bleach also bleached the chlorella algae, along with all of the ph papers and litmus papers.

-The spirulina algae turned into a thick, dark, gooey substance when it was mixed with the water. After the addition of the water, the sprirulina algae turned lighter and separated and the chlorella algae turned watery and dark. After the isopropyl alcohol was added, both of the algaes started to separate and created a light green film over top.

Challenges and shortcomings:

- I had to use bagged algae instead of fresh algae due to the inability to acquire algae cultivations.

- I only had access to two types of algae instead of the 4 that I would have liked. This was due to the cost and the unavailability of powdered algae in stores across calgary.

- I was only able to gather up 6 different household acids and bases to mix with the algae.

- There may have been experimental errors such as:

    -Contamination errors

-Accidental mis-measuring of solutions

-Timing errors

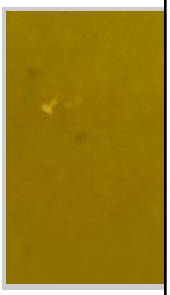
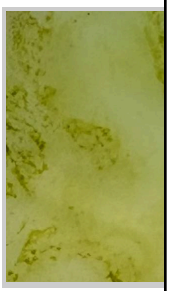
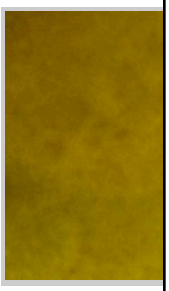
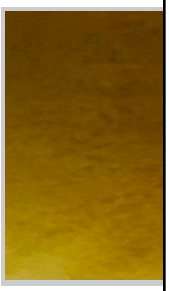
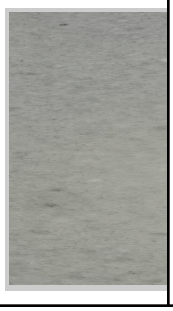
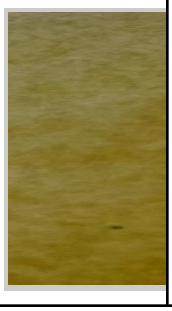
Advantages of the Experimental Methods:

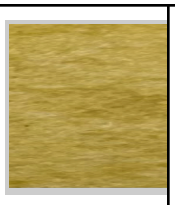
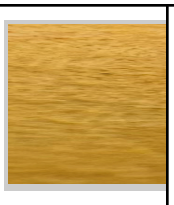
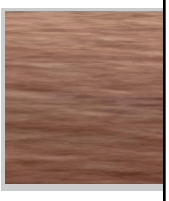
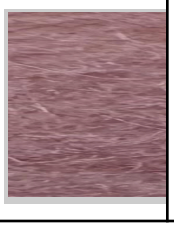
-It showcases the two types of algae that are most commonly used in the food industry. Which creates an advantage when it comes to finding commercial applications.

-Chlorella and spirulina algae both have strong chlorophyll pigments, giving them a good variety of colour changes.

-The yielded some valuable results that helped create a miniature algae pH comparison table.

Data Collection Table:

Type of Algae:	pH 3 (Acidic-Vinegar)	pH 7 (Neutral-Milk)	pH 13 (Basic-Bleach)	pH 6 (Acidic-Tomato juice)	Ph 2 (Acidic-Lemon-juice)	Ph 10 (Basic-soap)
Spirulina (green-blue algae)						
Chlorella (green algae)						

Ph Paper						
Blue litmus paper						
Red litmus paper						

Extra Application Ideas:

- Comparison to commercial pH indicators like litmus paper and Ph paper
- Test time stability — will the color remain stable or fade?
- Real-world applications: Could algae pigments be used in food safety, skincare, or environmental testing?

Trends in the results:

-Both algae types turned from darker greens to lighter greens, yellows, olive greens, and reds.

-The algae Ph indicator degraded faster in acidic environments (pH < 4, i.e. lemon juice) compared to alkaline environments.

-The spirulina and chlorella pigments changed to similar colours, which was unexpected, because Spirulina is a blue-green algae and chlorella is a green algae.

Section 5:

Conclusion:

Based on the experiment results, spirulina and chlorella algae pigments show promising potential as a natural, eco-friendly, cost effective, sustainable Ph indicator. They are useful in various scientific fields, like environmental sciences, food safety, cosmetics, and healthcare. These Ph indicators will be helpful with environmental monitoring such as water quality testing. They could be used to monitor the pH of lakes, oceans, and rivers especially in the areas that have been affected by industrial waste or acid rains. Algae pigments could also be very helpful in ocean acidification studies as certain pigments react strongly with acids and can help track changes in the sea waters pH.

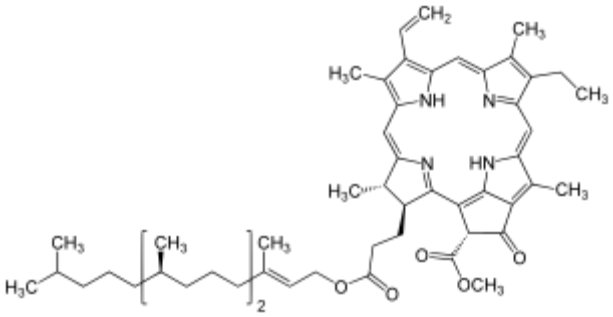
The food industry could also benefit from algae pigments. These pigments could function as natural food pH testers and could replace artificial pH indicators in food safety testing, like checking the freshness of dairy and meat products. Additionally, algae-based sensors that could be embedded inside of food packages that change colours if the food spoils.

Algae pH indicators could also have medical applications. They could be used as non-toxic Ph testers for bodily fluids like saliva or urine to detect health conditions such as acidosis or alkalosis as well as wound healing indicators. Another important application is in the field of cosmetics. Algae pigments could help develop pH-balanced skincare.

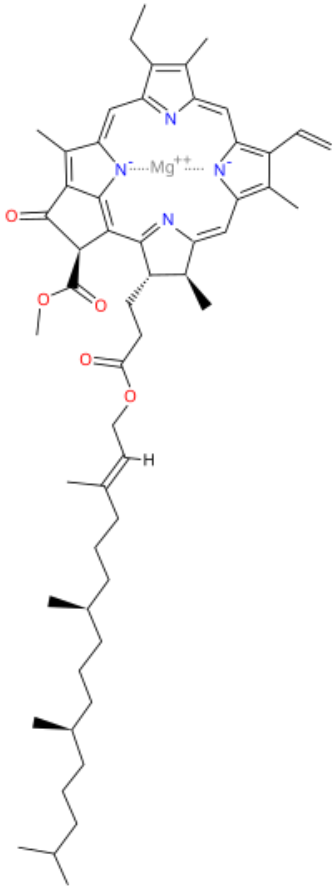
Finally algae pigments may be used as biodegradable pH test kits for schools. Schools could use algae pH kits in labs rather than chemical pH kits. They are safe, sustainable, and completely biodegradable. This will help divert chemical waste from the landfills. To conclude, Algae pigments will be useful in numerous different fields and could possibly revolutionise many different fields.

How this works:

Algae Pigments can change colours with various pH levels. This is due to their pH-dependent structure and chemical composition, as well as a process called Pheophytin Ionization. Pheophytin Ionization is the process of chlorophyll (a green pigment used to absorb light in plants) breaking down into pheophytin. This causes a colour change from green to an olive-brown due to acidification. Chlorella and Spirulina contain high amounts of chlorophyll. This is why I mainly found the colours shifting between greens, browns, and yellows with acidification. Pheophytin ionization occurs when chlorophyll's magnesium ions (Mg^{2+}) are replaced by two hydrogen ions (H+). This happens when it is added into acidic solutions. The process is highly accelerated by low pH. This can also occur when vegetables are canned and fermented and the pH is lowered. Pheophytin is also a reliable bio-indicator of environmental stress.



Chlorophyll A without the Mg^{2+} ion (Pheophytin ionization) [source: Wikipedia]



Chlorophyll A with Mg^{2+} ion [source: Wikipedia]

Section 6:

Future Steps:

In the future, I could use more than two different types of algae because different algae species contain unique pigments. That would show varied pH sensitivities, leading to a wider range of color changes. I would also use more than 6 different household acids and bases. Expanding the number of acids and bases gives a broader, more intricate, dataset allowing you to see if certain substances cause faster or stronger pH-related pigment changes. I would also add fresh algae instead of bagged, dried algae to get more pigmented results. Fresh algae retain more natural pigments, making the color changes more intense and accurate. With these changes, The Experiment will yield more accurate and stronger results.

CYSF Project Timetable

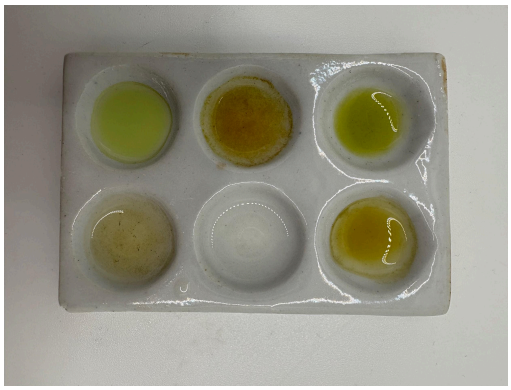
Task	Task Status	Date Completed	Notes
Choose a project	Finished	Dec 10th, 2024/January 7th,2025	I have chosen my project! I will figure out which algae species/environnement is the most ideal for producing a lot of lipids to create algae biofuels. Update- I have decided to change my project to to problems with the cost and timeline. I will now be Creating algae-based natural pH indicators out of different algae species and comparing them to

			commercial pH indicators such as litmus and pH paper.
Start background research	Finished	January 22nd, 2025	Today I've started doing some background research on algae-based ph indicators. I will use this to create a small pre-experiment write up and create the instructions for my experiment. I will continue to do research and add it as necessary.
Start the experiment	Finished	January 20th 2025	I have acquired all the necessary materials for my experiment and I've written down some instructions based on research for how I will extract the pigments.
Submit safety form	Finished	February 7th 2025	I finished my 2A safety form on the CYSF platform, it has an X next to it which makes me kind of worried considering that this is the last day to submit the form and if it does not work then I cannot participate in the CYSF.
Commercial applications	Finished	February 7th 2025	I finished writing up some possible commercial and environmental applications for algae pigment pH indicators and why they are important.
Finish Experiment	Finished	February 20th 2025	I have completed my experiment and found some interesting results.

			The colour changes were more drastic then I would have expected. The colours were a lot more vibrant. The materials for the experiment came a lot later than they should have, so I had to finish the experiment quicker.
Finish extra research	Finished	February 28th 2024	I have found a few facts about algae and I have researched why algae changes colours with acidification. I have also done some research about algae so I could learn more about the plant I was using for my experiment.
Finish additional applications	Finished		I have thought up some ways that algae pigments could be applied in commercial
Start trifold	Finished		I have started my trifold and am currently planning out how everything will fit on it.
Finish trifold	In progress	File	

Websites utilised for research:
-https://oceanconservancy.org/blog/2023/07/25/wild-facts-about-algae/ (Facts about algae)
-https://oceanservice.noaa.gov/facts/acidification.html (Ocean acidification)
-https://pmc.ncbi.nlm.nih.gov/articles/PMC3819078/ (what scientists are using algae for)



CLOROX
Splash-Less


FOUNDERS & FARMERS
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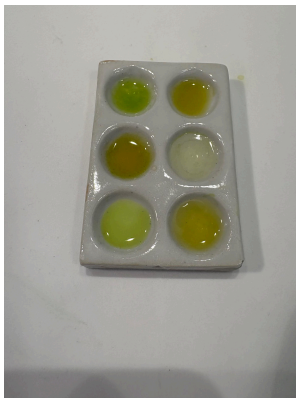
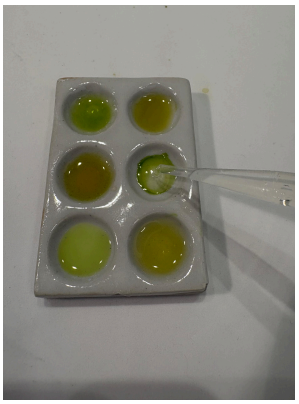
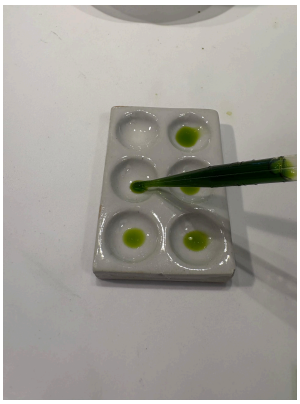

Organic traditions
CHLORELLA POWDER
POUDRE DE CHLORELLE

Organic traditions
SPIRULINA POWDER
POUDRE DE SPIRULINE

EXP 2029-NO



Lemon Juice
Bleach
Vinegar
Soap
Tomato Juice
Milk
Spirulina

Chlorella


Chlorella

Spirulina
Chlorella




life
Isopropyl Alcohol 99%
DANGER

Organic traditions
CHLORELLA POWDER
POUDRE DE CHLORELLE

Organic traditions
SPIRULINA POWDER
POUDRE DE SPIRULINE